Appendix A:

Final Risk Assessment For Diquat Bromide

The Water Quality Program

WASHINGTON STATE
DEPARTMENT OF
ECOLOGY

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TABLE OF CONTENTS

Diquat

Volume 4, Section 1

LABEL DESCRIPTION & HISTORY

29 PAGES

TABLE OF CONTENTS

1.0 REGISTRATION STATUS

This section describes the historic and current federal labels and typical uses. It also summarizes application rates, weeds controlled and reports on current practices undertaken by licensed Washington applicators. The final section describes research underway, including rate and application technology and proposed new labeling.

1.1 DIQUAT AS AN AQUATIC HERBICIDE

1.1.1 Registration Requirements

In order to register a pesticide with the EPA for use in the United States, the active ingredient and its formulations must be tested for mammalian toxicity, physical chemistry, environmental fate, effects on ground water, and eco-tox effects. Work must also be done to demonstrate the expected magnitude of residue on edible products and residues in water. After this data is generated, it is submitted to various branches of EPA for review. If EPA finds that the product does not pose significant risk to humans, livestock, or wildlife and has a favorable environmental persistence and degradation profile, a registration will be granted. With that registration, the manufacturer has permission to sell the product in the United States. However, each state may have its own separate registration process which may be more stringent than the EPA's registration process.

Washington State's registration procedure requires that the applicant submit a copy of the market label and a copy of the confidential statement of formula. The Washington State Department of Agriculture (WSDA) reviews these submittals for compliance with state and federal requirements. If these requirements are filled, the product will be registered by the state unless it presents an unusual hazard to the environment.

Studies conducted for submission to EPA since 1987 must be conducted in compliance with Good Laboratory Practice (GLP) regulations as specified in 40 CFR (Code of Federal Regulations) 160. These regulations are designed to improve the quality of records keeping and prevent fraud. They specify what records must be kept and how long they must be kept. They also specify how long analytical standards must be kept, how often they must be re-characterized and storage conditions. Furthermore they provide guidelines on how to determine the length of time that organic and inorganic reagents, solvents and biological samples can be kept, and under what conditions they should be stored. Also, GLPs provide guidance on how the integrity of the biological samples can be determined. For practical purposes, GLPs insure the integrity of the data. They allow for the reconstruction and interpretation of data within the study.

WSDA has registered diquat for use under several labels, but the only registered formulation for use in public waterways is Reward® Landscape and Aquatic Herbicide. This product contains 37.3% active ingredient (3.73 pounds diquat dibromide/gallon) which is equal to 20% cation equivalents (2.0 pounds diquat cation/gallon).

Diquat products registered by WSDA and except for Reward, are restricted in their use "for application to ponds, lakes and drainage ditches where there is little or no outflow of water and which are totally under the control of the products user include:

No.	Product Registered with the WSDA	EPA Registration No.	Name of Company
1	Misty Weed Trol® VF	8123-37-10807	Amrep Inc.

2	Opti-Gro Aqua Trim II	8123-37-66114	Check-Mark
3	Sea-Cide® Aquatic Weed Killer	8123-37-40208	Drummond Amer. Corp.
4	Weedtrine® D	8959-9	Applied Biochemists Inc.
5	Share Quick Kill® Non-Selective Vegetation Killer	10088-13-11547	Share Corp.
6	Total Solutions Eliminator®	10088-13-68562	Total Solutions
7	Zeneca* Diquat® Herbicide	10182-353	Zeneca* Ag Products Inc.
8	Reward® Landscape and Aquatic Herbicide	10182-404	Zeneca* Ag Products Inc.

* Zeneca Diquat products are now sold by Syngenta

Diquat® initially received Federal registration for control of submersed and floating aquatic weeds in 1962 and completed the Registration Eligibility Decision (RED) process on February 18, 2000. In addition to controlling watermilfoil, diquat is known to be particularly effective against water-lettuce (*Pistia stratiotes*), duckweed (*Lemna* spp.), cattails (*Typha* spp.), and *Salvinia* spp. Very few aquatic herbicides are effective in controlling these species of aquatic macrophytes (Gallagher and Haller, 1990 and Reward® LA label, 2000).

1.1.2 Registration and Permitting History

In the state of Washington, applications of aquatic herbicides and algaecides are performed by WSDA licensed applicators under a state permit system. Ecology manages this system and uses a 2001 Supplemental Environmental Impact Statement (SEIS) for diquat, copper compounds, glyphosate, endothall, 2,4-D and fluridone as well as manual, mechanical and biocontrol methods as guidance for writing permits for aquatic weed and algae control (Ecology, 2001). The state permitting system is a result of six agencies working together to develop a statewide integrated pest management system for aquatic plants and noxious emergent vegetation. The goal of this system is to ensure that the most effective and least environmentally damaging management alternatives will be used.

Ecology is responsible for issuing short-term modifications (STMs) to the water quality standards. These are required for management activities such as use of pesticides, mechanical or other control methods that might cause excess turbidity or violate other provisions of the water quality standards. Ecology is also responsible for ensuring consistency of proposals with rules and regulations designed to protect groundwater, shorelands, wetlands, air quality, and other elements of the environment.

Ecology evaluated diquat in the 1992 SEIS and determined it would not be permitted for use in Washington waters until critical information was available on toxicity, degradation, carcinogenicity, mutagenicity, and impacts to aquatic biota, specifically the clawed frog and other amphibians. Additionally, endothall, a less toxic contact herbicide, was available for use. From 1992 to present, there has been a considerable amount of research done to support the continuing registration of aquatic herbicides and algaecides containing diquat. As such, the most current data for these materials has not been considered or used in the issuance of permits to perform aquatic weed and algae control in Washington State (Resource Management, Inc., 1999). Specifically, the research needs and concerns for diquat identified in the 1992 SEIS included:

- Mutagenic effects – the 1992 SEIS found conflicting data
- EPA's carcinogenicity study was still pending in 1991
- Long-term low-level exposure has caused corneal opacity and cataracts in animals.

- A sub-chronic oral response dose (RfD) was not available for diquat in 1991
- Diquat was determined to be about ten times more toxic than endothall on an mg/kg body weight basis (PTI Environmental Services, 1991).

1.1.3 Risk Assessment (For a more detailed analysis see Section 4.1.10.2.5)

Formal reports to the EPA by the registrant Syngenta (formerly Zeneca), peer-reviewed literature, the EPA RED (EPA, 1995) and various EPA databases were reviewed in order to prepare this risk assessment: 1) The documents used by the registrant to support registration were those documents submitted to EPA in the course of the registration and re-registration process of diquat. They were conducted according to the EPA's current pesticide assessment guidelines and, if conducted after 1987, were also conducted under Good Laboratory Practice Regulations (40 CFR 160). 2). The bulk of the reviewed literature was completed more than 20-years ago. However, this literature is supported by the more recent unpublished literature submitted to EPA as part of the registration effort 3). A large portion of the toxicity data was collected from EPA's Brian database or the EPA's ECOTOX database, which are compilations of ecotoxicology data currently in use at EPA to generate and support ecological risk assessments. Information collected on work done before 1989 was collected from general review articles on the toxicity and environmental fate of diquat such as Shearer & Halter (1980) Campbell et al (1999), Hammer (1994), and Ecology (1980). Where possible, the toxicity values reported in the review articles were verified in the originally cited articles.

Herbicides used for aquatic weed control fall into one or more general categories: 1) Contact herbicides are chemicals that control weeds by direct contact with the foliage and destroy only those portions of the plant (generally the roots survive and plants regrow). 2) Systemic herbicides are applied to the foliage and/or stems of the plant and translocated to the roots or other portions of the plant, eventually resulting in the death of the entire plant. 3). Broad-spectrum herbicides kill most, if not all plants, if the dosage is appropriate. 4). Broadleaf herbicides generally kill dicot plants with broad leaves but there are exceptions; some broadleaf herbicides can kill monocots with broad leaf morphology and certain "narrowleaf" dicots are not harmed at concentrations that typically kill broadleaf plants. 5). Submerged (submersed), emerged (immersed) or floating indicates the way the plant typically grows. i.e., below the water line (submerged), from below the water line to above the waterline (emerged) and on the surface of the water and often un-rooted (floating). Pre-emergent and post-emergent weed control refers to whether control measures are taken prior to or after germination or first growth of the plant.

Diquat is a post-emergent, non-selective contact herbicide used primarily to control submerged weeds but it may also be used to control some floating and emergent weeds. The mode of action for diquat is inhibition of photosynthesis. Diquat is not typically used for algae control and most species of algae are not affected strongly by diquat (Shearer and Halter, 1980). Although the label claims no systemic action at the current labeled use rates, Hiltibran et al (1967) noted systemic action with Sago pondweed but not American pondweed.

The Risk Assessment in Section 4 indicates that diquat (diquat dibromide) may be used safely when most species of fish and invertebrates are present. The 96-hour LC50 for all verified studies on fish is greater than 0.5 ppm c.e. Therefore, the Risk Quotient for the most sensitive species of fish is below the acute levels of concern (0.1) for protection of the biota. The Acute Risk Quotient for diquat using early life-stage largemouth bass and striped bass is 0.04 (0.021 ppm c.e./~0.5 ppm c.e.). The very sensitive amphipod (*Hyallella azteca*) yields an acute risk quotient somewhat higher than the level of concern (RQ = 0.38 = 0.021ppm c.e. /0.048 ppm c.e.). However, when sediment is included in the test system the risk quotient drops to an acceptable level (RQ = 0.003 = 0.021 ppm c.e./6.8 ppm c.e.). These results are important since striped bass, largemouth bass and amphipods are considered to be extremely sensitive to most pesticides including diquat (Hughes, 1969, Wellborn, 1969, Wilson and Bond, 1969 and Shearer and Halter, 1980). Therefore, for all practical purposes, the level of concern (0.1) is not exceeded in acute risk assessment for protection of the biota. Since the application rate of diquat is low and residues generally disappear in less than a week, the conduct of a chronic risk assessment is not necessary to determine chronic safety to the biota (Campbell et al, 2000). Therefore, it should be possible to use diquat according to the label without significant acute or chronic risk to aquatic animals.

For fish, the results of acute risk assessments have been confirmed by field studies (Shearer and Halter, 1980 cites, and Hammer, 1994). Concentrations (0.5 to 3 ppm) that are somewhat higher than the current recommended maximum use rate (0.75 ppm c.e. = 4.48 Kg c.e./Ha in two feet of water) did not adversely impact fish. Lack of chronic risk has also been confirmed by field studies with warm water fish. Growth and reproduction of bluegill sunfish were unaffected by a single exposure to 1 to 3 ppm diquat. Furthermore, spawning success was not reduced due to diquat treatments in first generation post treatment.

Lorz et al (1979), found that the acute toxicity of diquat to salmon is low (11 to 300 ppm). However, exposure of Coho salmon to concentrations of diquat as low as 1.0 ppm for 10 hours did cause 15 to 87% of the smolts in a saltwater challenge test to die. Despite subsequent tests finding that smolts exposed to 3.0 ppm for 13 days survived saltwater challenge tests, fish exposed to these levels of diquat exhibited pathological symptoms including pathological changes in the eye, kidney, gill and liver. The gill ATP-ase enzyme complex was apparently unaffected in these fish which is consistent with surviving the saltwater challenge test. Coho salmon smolts exposed to diquat levels as low as 0.5 ppm for 96 hours were inhibited from migrating down stream. Failure to survive saltwater challenge tests and/or migrate down stream is an indication that diquat may interfere with the parr to smolt metamorphosis. More recent saltwater challenge tests with Chinook salmon indicate exposure to 0.5 ppm c.e. of diquat for 96 hours did not impair the osmo regulatory capability of newly smolted salmon. Furthermore, there were no mortalities or sublethal behavioral effects when Chinook smolts were challenged with seawater for 24 hours after exposure to diquat for 96 hours (Parametrix, 1997). Since the effects of diquat on smolting salmon appears to be uncertain, permits may be written to prohibit use of diquat products when smolts are present or in areas where salmon spawn.

Most species of aquatic invertebrate were not directly affected by diquat in the field (Shearer and Halter, 1980 site reviews by Shely, 1972 and Mullison, 1970). Zooplankton populations exposed to 0.5 ppm diquat were briefly depressed in Florida ponds, but recovered completely by day 11. Gilderhaus (1967 cited in Shearer and Halter, 1980) found that while benthic rotifers and copepods were not affected by multiple exposures to 1 to 3 ppm diquat in the field, cladoceran populations were temporarily affected. Populations of the cladoceran (*Simocephalus ventulu)* were eliminated for up to 64 days from ponds treated with 1 ppm diquat (Newbold, 1975 as reported in Shearer and Halter, 1980). The amphipod (*Hyalella azteca*) was killed in studies where sentinel animals were caged in the field and then exposed to 0.5 ppm diquat (The Water Investigations Branch, 1977 in Shearer and Halter, 1980). Temporary reductions in amphipod numbers were noted in shallow water where diquat concentrations could reach 0.7 ppm (Berry et al, 1975 in Shearer and Halter, 1980). However, in deep water, where diquat concentrations remained below 0.03 ppm, no reductions in amphipod numbers were noted. The data is somewhat conflicting. It is not clear whether these adverse affects are due to the direct affect of diquat poisoning or secondary effects due to loss of habitat (Hilsenhoff, 1966 in Shearer and Halter, 1980 and Hammer, 1994).

1.1.4 Registration Labels

1.1.4.1 Current Labels

There are currently thirty-four diquat labels registered for use in aquatic environments in Washington State. A number of these are registered for control only in impounded waters which cannot be considered to be either public waterways or waters that empty into public waterways. Therefore, Misty® Weed Trol VF, Aqua Trim® II Vegetation Control, Sea-Cide® Weedtrine® D Aquatic Herbicide, Quick Kill® Non-Selective Vegetation Killer, and Eliminator®, which are registered only for use in impounded water ways should not have an impact on public waterways. Zeneca Diquat® Herbicide is registered only for use in "non-flooded" portions of ponds, lakes and drainage ditches on farms and also should not have an impact on public waterways (i.e., "navigable waters" or "waters of the United States" see 33 U.S.C. SS 1311(a), 1362 (7)).

Reward® Landscape and Aquatic Herbicide may be applied to public waters in ponds, lakes, reservoirs, marshes, bayous, drainage ditches, canals, streams, rivers, and other slow-moving or still bodies of water for control of aquatic weeds. Reward® Landscape and Aquatic Herbicide is effective in controlling a variety of aquatic weeds listed in Table 2 and a couple of species of filamentous algae. The federal label for Reward® LA with potential for uses in Washington is attached in Appendix 1.

1.1.4.2 Historical Use and Labels

Diquat is used primarily to control aquatic macrophytes but also is registered to control certain species of filamentous algae in irrigation canals, ponds and lake/reservoirs. In 1966, EPA registered diquat for use in controlling submerged and floating aquatic macrophytes. At that time no other aquatic herbicides were registered for the control of water-lettuce, duckweed, *Azolla spp.* or *Salvinia* spp. Syngenta is currently the main manufacturer and distributor of diquat in the United States under the Reward® LA label.

Diquat is used widely in the United States but 60% of the total diquat applied for aquatic weed control (110,000 pounds/year) is used in Florida. In Florida the aquatic weed problem is related to the extent of surface water, climate, weed species and factors affecting dissemination of aquatic plants including recreational use and interconnections between waterways (Ritter et al, 2000).

For the purpose of historical significance, a diquat label from ~1990, corresponding to the current registered federal label for Reward® LA, is located in Appendix 2. This historical label indicates that the formulation, recommended uses and use rates have not changed significantly for diquat.

1.1.4.3 Label Restrictions

- **Label Restrictions**

  The label restrictions in place as of February 2000 are outlined below. However, label restrictions may change based on new data received by EPA. The current label contains the most up-to-date restrictions and the most current label that should be consulted when applying an herbicide.

There are a number of conditions on the current Reward® label that restrict the use of diquat when the maximum contaminant level goal (MCLG) of 0.02 ppm c.e. is exceeded. This includes restrictions on the use of treated water for drinking, live stock watering, agricultural sprays for food crops, turf and ornamental plants and irrigation for food crops, turf and ornamental plants. In absence of an approved analytical method, specific time periods specified in the label may be used to restrict water use after application of Reward® LA. The labels and permits that govern those restrictions may be periodically changed based on new information submitted to EPA and Ecology (Table 1).

Application of diquat products must be limited to a portion of the water body at any one time because decaying vegetation can deplete the dissolved oxygen content of the water and aquatic organisms need oxygen to survive. Treatment of dense weed areas may result in dissolved oxygen decreases due to the decomposition of dead weeds. A significant decrease in dissolved oxygen content may cause the suffocation of fish or other aquatic animals. No more than one-third to one-half of a water body should be treated at one time and a waiting period of 14 days between treatments is required. For actual size areas recommended for treatment or other restrictions, consult the label and the permit. Note that typical control measures often do not result in the treatment of an entire water body and typically only about 20% of a water body is treated based on areas designated for priority control. Control within areas of a water body can be designated a priority based on the impact aquatic weeds may have on specific areas of the water body. These priority impacts may include recreational use, water storage and withdrawal function, flood control, irrigation, property values, human health considerations, fish and wildlife habitat, ecological health and stability, biodiversity and effects on threatened and endangered species (Getsinger, Personal Communication, May, 2000).

There have been no field studies conducted with diquat that have shown that diquat is directly toxic to fish at standard maximum use rates (Shearer and Halter, 1980). Labels warn that diquat dibromide is toxic to aquatic invertebrates.

- **Additional Restrictions Imposed by Washington State**

Ecology uses a permit system to impose additional restrictions on the use of aquatic herbicides in Washington State. A sample boilerplate with the range of special and general conditions for the use of diquat can be found in Appendix A.

1.1.3.4 Labeled Use

Several diquat products are labeled for use in ponds and lakes by the US EPA. Reward® Landscape and Aquatic Herbicide is also registered for irrigation and drainage canal applications. Plants with floating leaves (Salvinia, water-lettuce and water-hyacinth) should be treated at the surface with Reward® LA in 75 to 200 gallons of water/acre plus the labeled rate of a 75% or greater nonionic surfactant. Emergent plant species (cattails) and certain floating species like pennyworts and duckweed should be treated by applying Reward® LA directly to the foliage in 50 to 150 gallons of water/acre plus the labeled rate of a 75% or greater nonionic surfactant. Reward® LA may be applied by air in 10 to 24 gallons of water/acre plus the labeled rate of a 75% or greater nonionic surfactant for the control of water-lettuce and water-hyacinth. Submersed vegetation (bladderworts, coontail, Elodea, Hydrilla and watermilfoils), Reward® LA should be applied in a water carrier with a thickener through boom trailing hoses with appropriate nozzle tips designed for application below the water surface. Reward® LA may alternatively be poured directly from the container in strips or as a spray in a sufficient carrier to assure complete coverage of the weed infested areas. If the weed population is mixed, the highest rate appropriate for the most tolerant species present should be used. Contact with non-target species by surface or air applied spray should be avoided so that drift does not adversely impact non-target or crop species. All equipment should be calibrated carefully to be sure of spreading the proper amount of herbicide.

When diquat products are used to control floating or emerged weeds, a non-ionic surfactant at the labeled use rate should be combined with the diluted liquid formulation to assist with sticking and penetration of the pesticide. This has the effect of reducing both the application rate and the cost of the application. Care should be taken to select a surfactant that has been approved for aquatic use. Surfactants approved for aquatic use may harm fish. Thickening agents like PolyControl® or one of the organosilicates are often added to herbicide solutions that are applied to the water surface in order to control drift (Kurt Getsinger, Army Corp of Engineers Interview, Appendix 5 of Endothall Section 1 Document).

When liquid formulations of diquat are applied by subsurface injection, thickening agents may be used to allow the liquid diquat product to drop lower in the water column where it will be more effective (Kurt Getsinger, Army Corp of Engineers Interview, Appendix 5 of Draft Endothall Section 1 Document).

1.1.3.5 Effectiveness Controlling Specific Aquatic Plant Species

In the control of aquatic weeds, diquat is a broad-spectrum general contact herbicide. The mode or action against macrophytes is the disruption of photosynthesis. Diquat is effective against a wide variety of aquatic weeds (Table 2) at label application rates of 0.5 to 2 gallons of formulation per surface acre, which gives typical water concentrations that range from 0.2 to 0.75 ppm c.e. According to the label, Reward® LA typically controls bladderwort (*Uticularia* spp.), coontail (*Ceratophyllum demersum*), Elodea (*Elodea* spp.), naiads (*Najas* spp.) pondweeds (*Potamogeton* spp. except *Potamogeton richardsonii*), watermilfoils (*Myriophyllum* spp.)

including parrotsfeather, Eurasian watermilfoil and northern watermilfoil, hydrilla (*Hydrilla verticillata*), water-lettuce (*Pistia stratiotes*), water-hyacinth (*Eichornia crassipes*), pennywort (*Hydrocotyle* spp.), frog's bit (*Limnobium spongia*), *Salvinia* spp., duck weed (*Lemna* spp.), cattails (*Typha* spp.) and certain species of filamentous algae (*Spirogyra* spp.and *Pithophora* spp.). For rates of applications to control these species, consult the Reward® TA Label (2000). Robinette (1998-1999) and Westerdahl et al. (1988) list a variety of other species and the relative effectiveness of Diquat® products to control the growth of these species (Table 2).

In addition to the macrophytes that Reward® LA is labeled to control, diquat has been observed to control certain other species of macrophyte for which there is no efficacy claimed on the label. These species include *Polygonum* spp. and fanwort, Water buttercup (*Ranunculus aquatilis*), giant duckweed (*Spirodela polyrhiza*) mosquito fern (*Azolla caroliniana*), slender duckweed (*Wolffiella floridana)*, watermeal (*Wolffia columbiana*), Brazilian elodea (*Egeria densa*), fanwort (*Cabomba, caroliniana*) and widgeon grass (*Ruppia maritima*) (Westerdahl et al, 1988). It is a violation of federal law to use a pesticide inconsistent with its labeling. However, Section 2(ee) of FIFRA does provide for the application of a pesticide that is applied to a plant, crop, animal or site that is specifically listed on the label. Therefore, these aquatic plants could be treated with Reward® at doses no greater than those specified on the label *provided* that the application was also consistent with Washington State Department of Ecology permitting/application requirements. Also, the pesticide may be applied at a dosage, concentration, or frequency less than that specified on the label.

Certain species of aquatic macrophytes are of particular interest to Ecology. They are *Myriophyllum spicatum* (Eurasian watermilfoil), *Lythrum salicaria*, (purple loosestrife), *Egeria densa* (Brazilian elodea), *Myriophyllum aquaticum* (parrotsfeather*), Cabomba caroliniana* (fanwort), *Hydrilla vertcillata* (hydrilla), *Tamarix ramosissima* (saltcedar), *Amorpha fruticosa* (indigobush), *Polygonum sachalinense*, (giant hogweed or giant knotweed), *Polygonum cuspidatum* (Japanese knotweed), *Lysimachia vulgaris* (garden loosestrife) and *Phalaris arundinacea* (reed canarygrass).

Some species of aquatic plants are known to resist or tolerate diquat. These species are *Nuphar* ssp. (spatterdock), *Nymphaea* spp. (fragrant water lilies), and many species of algae (Shearer and Halter, 1980). When the biomass of other aquatic species is decreased by diquat use, tolerant species have the potential to become dominant and decrease plant diversity in the treated area. Use of Reward® LA and other formulations for plants not listed on the label is not recommended because it may not be effective. However, the label does not prohibit its use.

1.1.3.6 Other Concerns

Although it is not the practice of Ecology to permit the use of tank mixes, diquat has been used in tank mixes to control a broader spectrum of weeds than is possible with diquat alone. For example, 2,4-D and diquat have been used as tank mixes for control of water hyacinth and water-lettuce. Also copper chelates and diquat have been used in tank mixes to simultaneously control algal epiphytes and vascular plant species.

Various non-ionic surfactants have been used with diquat for the control of floating and emergent aquatic vegetation. One of the main purposes in using a surfactant is to decrease the amount of active ingredient necessary to control floating and emergent aquatic vegetation. Some common surfactants used with aquatic herbicides are X-77®, CideKick®, PolyControl® and SunWet®. Non-Ionic aquatic surfactants registered for use with aquatic herbicides like diquat should not be toxic to fish or aquatic invertebrates.

The rate of herbicide use is controlled by a number of use pattern factors. These factors may include the species of plant to be controlled, the degree of infestation, the contact time between the treated water and the plants to be controlled, usage patterns including susceptibility of the treated plant species, depth of the water, temperature of the water and water exchange patterns. For example, water lettuce is more susceptible than duckweed, which in turn is more susceptible than Eurasian watermilfoil to diquat. Presence of a dense weed population may require a higher use rate to achieve control than a sparse weed population. Concentration/exposure time is usually proportional; that is a low concentration requires a longer exposure time to achieve control than a high concentration. Deep water (>2 feet) requires a higher treatment rate (4 lbs c.e./acre) than shallow water (<2 feet) which only requires a treatment rate of 2 lbs c.e./acre. Cold water (<50 to 60°F) may decrease the efficacy and immediacy of aquatic weed control. Water which has a high flow rate, large volume dilutions from ground water or surface water sources or extensive vertical and lateral mixing may require higher use rates than still or impounded water, which has little dilution or mixing.

1.1.4 Maintaining the Current Registration

Since the last Supplemental Environmental Impact Statement (1992), a number of additional studies compliant with the EPA's FIFRA Pesticide Assessment Guidelines and Good Laboratory Practice Standards have been completed and submitted to the US EPA for review. Studies that are compliant with current regulations not only add to the database but also increase the confidence of regulatory organizations, elected officials and the general public that the data supports, the most recent risk assessment (EPA RED, 1995, Campbell et al, Ritter et al, and Bartell et al, 2000a, 2000b, 2000c) and the Supplementary Environmental Impact Statements (State of Washington 1980, 1989, 1992, and 2001). These studies may result in the addition or removal of certain use restrictions depending upon their outcome. The changes brought by the development of new data will be assessed in later sections of this document.

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LIST OF TABLES

Table 1: Rate of Application, Effective Concentrations, Comments and Label Restrictions for Reward® Landscape and Aquatic Herbicide[1, 2]

Comments & Label Restrictions for Reward® Landscape and Aquatic Herbicide
For instructions on application rates to control specific weeds, see the Reward® LA label (2000). For the control of aquatic weeds, application rates vary from 0.5 to 2 gallons formulation/ha (0.2 to 0.75 ppm c.e.). Rates vary depending on the target species and treatment conditions. Treatment at water temperatures less than 50 to 60°F may result in reduced efficacy and immediacy of results may be reduced. Restricted water use periods for domestic purposes (drinking), watering livestock, irrigation and preparing agricultural sprays for non-food and food crops vary depending on use rate. The maximum contaminant level goal (MCLG) is 0.02 ppm c.e.; but until an approved analytical assay has been developed, the water use restriction periods specified in the label must be followed. There is no fishing or swimming restriction specified in the label. However, no applications are to be made in areas where commercial processing of fish resulting in production of fish protein concentrate or fishmeal is practiced. Restricted water use periods after treatment with 2 gallons Reward® LA/surface acre are 3 days for drinking, 3 day for livestock consumption, 3 days for spray tank applications and irrigation on non-food crops and 5 days for spray tank applications and irrigation on food crops. The restricted water use period after treatment with 0.75 to 1.0 gallons Reward® TA/surface acre are 2 days for drinking, 1 day for livestock consumption, 1 day for spray tank applications and irrigation on non-food crops and 5 days spray tank applications and irrigation on food crops. Restricted water use periods after treatment with 0.5 gallons Reward® TA/surface acre or use of <0.5 gallons/surface acre as a spot spray are 1 day for drinking, 1 day for livestock consumption, 1 day for spray tank applications and irrigation on non-food crops and 5 days spray tank applications and irrigation on food crops. Reward® LA is effective against *Potamogeton* species of pondweed except *P. richardsonii*. Application for control of *P. robbinsii* must be made when plants are in early stages of growth (spring and early summer). The labeled rate of a 75% nonionic surfactant must be used when spraying for control of floating or emergent plant species like Salvinia, watter-lettuce, water-hyacinth, pennywort, duck weed or cattails.

1 For Reward® TA, the label reports the active ingredient to be (diquat dibromide at 37.3%)

2 The concentration is also reported as 2 lbs diquat cation/gallon (20% diquat cation) or 3.73 lbs diquat dibromide/per gallon.

Table 2: Species Controlled, Effectiveness of Control and Registration Status for Listed Species (Continued)

Table 2: Species Controlled, Effectiveness of Control and Registration Status for Listed Species

Species Controlled	Not label restricted and/or Effectiveness of Control		
	Reward® Landscape and Aquatic (36.4% diquat dibromide; 20% diquat cation)	**Misty® Weed Trol Aquatic & Contact Herbicide (4.35% diquat dibromide; 2.35% diquat cation)**	**Aqua Trim II Vegetation Control (1.85% diquat dibromide; 1.00% diquat cation)**
Utricularia spp. (Bladderwort)[2]	Not label restricted Good Control[3.4]	Not label restricted Good Control[3.4]	Not label restricted Good Control[3.4]
Egeria densa (Brazilian Elodea)[2]	No Efficacy Claimed[8] Good Control[4]	No Efficacy Claimed[8] Good Control[4]	No Efficacy Claimed[8] Good Control[4]
Scirpus spp. (Bullrush)[1,7]	No Efficacy Claimed Fair Control[4]	No Efficacy Claimed Fair Control[4]	No Efficacy Claimed Fair Control[4]
Sparganium spp. (Bur reed)	No efficacy Claimed	No efficacy Claimed	No efficacy Claimed
Typha spp. (Cattails)[1,7]	Not label restricted Good Control[3.4]	Not label restricted Good Control[3.4]	Not label restricted Good Control[3.4]
Cabomba caroliniana (Fanwort)[2]	No Efficacy Claimed Good Control[4]	No Efficacy Claimed Good Control[4]	No Efficacy Claimed Good Control[4]
Ceratophyllum demersum (Coontail)[2]	Not label restricted Excellent Control[3,4]	Not label restricted Excellent Control[3,4]	Not label restricted Excellent Control[3,4]
Phragmites australis (Common reed)[7]	No Efficacy Claimed Not recommended[4]	No Efficacy Claimed Not recommended[4]	No Efficacy Claimed Not recommended[4]
Lemna spp. (Duckweed)[1]	Not label restricted Excellent Control[4]	Not label restricted Excellent Control[4]	Not label restricted Excellent Control[4]
Lemna minor. (Duckweed)[1]	Not label restricted Excellent Control[4]	Not label restricted Excellent Control[4]	Not label restricted Excellent Control[4]
Spirodela polyrhiza (Giant duckweed)[1]	No efficacy Claimed Excellent Control[4]	No efficacy Claimed Excellent Control[4]	No efficacy Claimed Excellent Control[4]
Wolffiella foridana (Slender Duckweed)[1]	No Efficacy Claimed Good Control[4]	No Efficacy Claimed Good Control[4]	No Efficacy Claimed Good Control[4]
Elodea spp. (Elodea)[2]	Not label restricted Excellent Control[4]	Not label restricted Excellent Control[4]	Not label restricted Excellent Control[4]
Elodea canadensis (American waterweed)[2]	Not label restricted Excellent Control[4]	Not label restricted Excellent Control[4]	Not label restricted Excellent Control[4]
Hygrophila polysperma (Hygrophila)	No Efficacy Claimed	No Efficacy Claimed	No Efficacy Claimed
Limnobium spongia (Frog’s bit)[6]	Not label restricted	No Efficacy Claimed	No Efficacy Claimed
Lythrum salicaria (Purple loosestrife)	No Efficacy Claimed	No Efficacy Claimed	No Efficacy Claimed

Species Controlled	Not label restricted and/or Effectiveness of Control		
	Reward® Landscape and Aquatic (36.4% diquat dibromide; 20% diquat cation)	**Misty® Weed Trol Aquatic & Contact Herbicide (4.35% diquat dibromide; 2.35% diquat cation)**	**Aqua Trim II Vegetation Control (1.85% diquat dibromide; 1.00% diquat cation)**
Lysimachia vulgaris (Garden loosestrife)	No Efficacy Claimed	No Efficacy Claimed	No Efficacy Claimed
Hydrilla verticillata (Hydrilla)[2]	Not label restricted Excellent Control[3], Good Control[4]	No Efficacy Claimed Excellent Control[3], Good Control[4]	No Efficacy Claimed Excellent Control[3], Good Control[4]
Amorpha fruticosa (Indigobush)	No Efficacy Claimed	No Efficacy Claimed	No Efficacy Claimed
Phaenicum hemitomon (Maidencane)[7]	No Efficacy Claimed Fair Control[4]	No Efficacy Claimed Fair Control[4]	No Efficacy Claimed Fair Control[4]
Azolla caroliniana (Mosquito fern)[1]	No Efficacy Claimed Good Control[4]	No Efficacy Claimed Good Control[4]	No Efficacy Claimed Good Control[4]
Myriophyllum spp. (Watermilfoil)[2]	Not label restricted Good Control[3], Good to Excellent Control[4]	Not label restricted Good Control[3], Good to Excellent Control[4]	Not label restricted Good Control[3], Good to Excellent Control[4]
Myriophyllum aquaticum (Parrotsfeather)[2]	Not label restricted Good Control[3], Excellent Control[4]	Not label restricted Good Control[3], Excellent Control[4]	Not label restricted Good Control[3], Excellent Control[4]
Myriophyllum spicatum (Eurasian watermilfoil)[2]	Not label restricted Good Control[3], Excellent Control[4]	Not label restricted Good Control[3], Excellent Control[4]	Not label restricted Good Control[3], Excellent Control[4]
Myriophyllum heterophyllum (Variable leaf milfoil)[2]	Not label restricted Good Control[3]	Not label restricted Good Control[3]	Not label restricted Good Control[3]
Najas spp. (Naiad)[2]	Not label restricted Excellent Control[3,4]	Not label restricted Excellent Control[3,4]	Not label restricted Excellent Control[3,4]
Najas minor (Slender naiad)[2]	Not label restricted Excellent Control[3]	Not label restricted Excellent Control[3]	Not label restricted Excellent Control[3]
Najas quadalupensis (Southern naiad)[2]	Not label restricted Excellent Control[3]	Not label restricted Excellent Control[3]	Not label restricted Excellent Control[3]
Hydrocotyle spp. (Pennywort)[1,7]	Not label restricted Fair Control[3]	Label Use Excellent control[3]	Labeled Use Excellent control[3]
Hydrocotyle umbellata (Pennywort)[1,7]	Not label restricted Excellent control[3]	Not label restricted Excellent control[3]	Not label restricted Excellent control[3]
Polygonum sachalinense (Giant hogweed or Giant knotweed)[7]	No Efficacy Claimed Fair Control[4]	No Efficacy Claimed Fair Control[4]	No Efficacy Claimed Fair Control[4]
Polygonum cuspidatum (Japanese knotweed)[7]	No Efficacy Claimed Fair Control[4]	No Efficacy Claimed Fair Control[4]	No Efficacy Claimed Fair Control[4]

Table 2: Species Controlled, Effectiveness of Control and Registration Status for Listed Species (Continued)

Species Controlled	Not label restricted and/or Effectiveness of Control		
	Reward® Landscape and Aquatic (36.4% diquat dibromide; 20% diquat cation)	**Misty® Weed Trol Aquatic & Contact Herbicide (4.35% diquat dibromide; 2.35% diquat cation)**	**Aqua Trim II Vegetation Control (1.85% diquat dibromide; 1.00% diquat cation)**
Potamogeton spp. (Pondweeds)[2]	Not label restricted Good Control[3,4] Except for *P. richardsonii*	Not label restricted Good Control[3,4] Except for *P. robbinsii*	Not label restricted Good Control[3,4,5] Except for *P. robbinsii*
P. amplifolius (Bass weed or Big-leaf pondweed)[2]	Not label restricted Good Control[3]	Not label restricted Good Control[3]	Not label restricted Good Control[3]
P. diversifolius[2]	Not label restricted Good Control[3]	Not label restricted Good Control[3]	Not label restricted Good Control[3]
P. natans (Floating-leaf pondweed or American pondweed)[2]	Not label restricted Good Control[3]	Not label restricted Good Control[3]	Not label restricted Good Control[3]
P. pectinatus (Sago pondweed)[2]	Not label restricted Good Control[3]	Not label restricted Good Control[3]	Not label restricted Good Control[3]
P. pusillus (Thin-leaf pondweed)[2]	Not label restricted Excellent Control[3]	Not label restricted Excellent Control[3]	Not label restricted Excellent Control[3]
P. zosteriformis (Flat-Stem pondweed)[2]	Not label restricted Good Control[3]	Not label restricted Good Control[3]	Not label restricted Good Control[3]
Zannichellia palustris (Horned pondweed)[2]	No Efficacy Claimed Not Recommended[4]	No Efficacy Claimed Not Recommended[4]	No Efficacy Claimed Not Recommended[4]
Phalaris arundinacea (Reed canarygrass)	No Efficacy Claimed Not Recommended (Eadey & Renney, 1965)	No Efficacy Claimed Not Recommended (Eadey & Renney, 1965)	No Efficacy Claimed Not Recommended (Eadey & Renney, 1965)
Tamarix ramosissima (Saltcedar)	No Efficacy Claimed	No Efficacy Claimed	No Efficacy Claimed
Salvinia spp. (Salvinia)[1]	Not label restricted Excellent Control[4]	Probable Labeled Use; Excellent Control[4]	Probable Labeled Use; Excellent Control[4]
Salvinia rotundifolia (Salvinia)[1]	Not label restricted Excellent Control[4]	Not label restricted Excellent Control[4]	Not label restricted Excellent Control[4]
Polygonum spp. (Smartweeds)[7]	No Efficacy Claimed Fair Control[4]	No Efficacy Claimed Fair Control[4]	No Efficacy Claimed Fair Control[4]
Spartina spp. (Smooth cordgrass)	No Efficacy Claimed	No Efficacy Claimed	No Efficacy Claimed
Ranunculus aquatilis (Water buttercup)[2]	No Efficacy Claimed Excellent Control[4]	No Efficacy Claimed Excellent Control[4]	No Efficacy Claimed Excellent Control[4]

Species Controlled	**Not label restricted and/or Effectiveness of Control**		
	Reward® Landscape and Aquatic (36.4% diquat dibromide; 20% diquat cation)	**Misty® Weed Trol Aquatic & Contact Herbicide (4.35% diquat dibromide; 2.35% diquat cation)**	**Aqua Trim II Vegetation Control (1.85% diquat dibromide; 1.00% diquat cation)**
Eichhornia crassipes (Water-hyacinth)[1]	Not label restricted Good Control[3], Excellent Control[4]	Not label restricted Good Control[3], Excellent Control[4]	Not label restricted Good Control[3], Excellent Control[4]
Pistia stratiotes (Water-lettuce)[1]	Not label restricted Excellent Control[4]	Not label restricted Excellent Control[4]	Not label restricted Excellent Control[4]
Nuphar spp. (Spatterdock)[7]	No Efficacy Claimed Tolerant[6], Not Recommend[4]	No Efficacy Claimed Tolerant[6], Not Recommend[4]	No Efficacy Claimed Tolerant[6], Not Recommend[4]
Nymphaea spp. (Yellow water lilies)[7]	No Efficacy Claimed Tolerant[6], Not Recommended[4]	No Efficacy Claimed Tolerant[6], Not Recommended[4]	No Efficacy Claimed Tolerant[6], Not Recommended[4]
Wolffia columbian (Watermeal)[1]	No Efficacy Claimed Good Control[4]	No Efficacy Claimed Good Control[4]	No Efficacy Claimed Good Control[4]
Paspalum fluitans (Water paspalum)[7]	No Efficacy Claimed Fair control[4]	No Efficacy Claimed Fair control[4]	No Efficacy Claimed Fair control[4]
Ludwigia uruguayensis (Water primrose)[7]	No Efficacy Claimed Fair Control[4]	No Efficacy Claimed Fair Control[4]	No Efficacy Claimed Fair Control[4]
Brasenia schreberi (Waterhield)[7]	No Efficacy Claimed Fair Control[4]	No Efficacy Claimed Fair Control[4]	No Efficacy Claimed Fair Control[4]
Justica americana (Waterwillow)[7]	No Efficacy Claimed Fair Control[4]	No Efficacy Claimed Fair Control[4]	No Efficacy Claimed Fair Control[4]
Heteranthera spp. (Water stargrass)	No Efficacy claimed	No Efficacy claimed	No Efficacy claimed
Ruppia maritima (Widgeongrass)[2]	No Efficacy Claimed Good Control[4]	No Efficacy Claimed Good Control[4]	No Efficacy Claimed Good Control[4]
Vallisneria americana (Wildcelery)[2]	No Efficacy Claimed Fair Control[4]	No Efficacy Claimed *Fair Control*[4]	No Efficacy Claimed *Fair Control*[4]
Trapa natans (Water chestnut)[7]	No Efficacy Claimed Not Recommended[4]	No Efficacy Claimed Not Recommended[4]	No Efficacy Claimed Not Recommended[4]
Filamentous Algae species[2]	Not label restricted Good Control[3]	Not label restricted Good Control[3]	Not label restricted Good Control[3]
ora spp.[2]	Not label restricted	Not label restricted	Not label restricted
Spirogyra spp.[2]	Not label restricted	Not label restricted	Not label restricted
Other Species of algae	No Efficacy Claimed	No Efficacy Claimed	No Efficacy Claimed

[1] Floating or emersed plants. Requires a non-ionic surfactant mixed with diquat to achieve control.

Table 2: Species Controlled, Effectiveness of Control and Registration Status for Listed Species (Continued)

[2] Submersed plants or algae. Apply directly from the container, apply in a water carrier or apply as an invert emulsion. Application method should follow the label instructions.

[3] (Robinette, 1998-1999)

[4] (Westerdahl et al., 1988)

[5] P. Robbinsii applications must be made when the plants are in early stage of growth such as in spring or ear summer.

[6] Shearer and Halter, 1980.

[7] Emersed plants.

[8] Not listed as a controlled species on the label.

LIST OF APPENDICES

APPENDIX 1: Current Labels

PULL HERE TO OPEN

REWARD®

Landscape and Aquatic Herbicide

TO PREVENT ACCIDENTAL POISONING, NEVER PUT INTO FOOD, DRINK, OR OTHER CONTAINERS, AND USE STRICTLY IN ACCORDANCE WITH ENTIRE LABEL.

DO NOT USE THIS PRODUCT FOR REFORMULATION.

Active Ingredient: Diquat dibromide [6,7-dihydrodipyrido (1,2-a:2′,1′-c) pyrazinediium dibromide]	37.3%
Other Ingredients:	62.7%
Total:	100.0%

Contains 2 lbs. diquat cation per gal. as 3.73 lbs. salt per gal.

KEEP OUT OF REACH OF CHILDREN.

WARNING/AVISO

Si usted no entiende la etiqueta, busque a alguien para que se la explique a usted en detalle. (If you do not understand the label, find someone to explain it to you in detail.)

See additional precautionary statements and directions for use inside booklet.

EPA Reg. No. 100-1091
EPA Est. 100-TX-001

Product of United Kingdom
Formulated in the USA
SCP 1091A-L2 0901
154290

2.5 gallons

U.S. Standard
Measure

syngenta

SCP 130-1091A-L2

Reward®

FIRST AID	
If swallowed	• Call a Poison Control Center or doctor immediately for treatment advice. • Immediately give water or milk to drink and induce vomiting by inserting finger in throat. • Do not induce vomiting or give anything by mouth to an unconscious person. • Take person and product container to the nearest hospital or physician fast. • PROMPT TREATMENT IS ESSENTIAL TO COUNTERACT POISONING and should be initiated before signs and symptoms of injury appear.
If on skin or clothing	• Take off contaminated clothing. • Rinse skin immediately with plenty of water for 15-20 minutes. • Call a Poison Control Center or doctor for treatment advice.
If in eyes	• Hold eye open and rinse slowly and gently with water for 15-20 minutes. • Remove contact lenses, if present, after the first 5 minutes, then continue rinsing eye. • Call a Poison Control Center or doctor for treatment advice.
If inhaled	• Move person to fresh air. • If person is not breathing, call 911 or an ambulance, then give artificial respiration, preferably mouth-to-mouth, if possible. • Call a Poison Control Center or doctor for further treatment advice.
NOTE TO PHYSICIAN **CALL SYNGENTA MEDICAL EMERGENCY ASSISTANCE 1-800-888-8372** at any hour to obtain toxicology information and a diquat analysis. To be effective, treatment for diquat poisoning must begin **IMMEDIATELY**. Treatment consists of binding diquat in the gut with suspensions of activated charcoal or bentonite clay, administration of cathartics to enhance elimination, and removal of diquat from the blood by charcoal hemoperfusion or continuous hemodialysis.	
Have the product container or label with you when calling a poison control center or doctor, or going for treatment.	
HOT LINE NUMBER For 24-Hour Medical Emergency Assistance (Human or Animal) or Chemical Emergency Assistance (Spill, Leak, Fire, or Accident), Call **1-800-888-8372**	

PRECAUTIONARY STATEMENTS

Hazards to Humans and Domestic Animals

WARNING/AVISO

May be fatal if absorbed through skin. Harmful if swallowed or inhaled. Causes substantial, but temporary, eye injury. Causes skin irritation. Contact with irritated skin, or a cut, or repeated contact with intact skin may result in poisoning. Do not get in eyes, on skin, or on clothing. Avoid breathing vapor or spray mist. Do not feed forage from treated crops to livestock. Keep livestock and pets out of treated fields and crop areas.

Personal Protective Equipment (PPE)

Applicators and other handlers must wear:

- Coveralls over short-sleeved shirt and short pants or coveralls over long-sleeved shirt and long pants
- Waterproof gloves
- Chemical-resistant footwear plus socks
- Protective eyewear
- Chemical-resistant headgear for overhead exposure
- Chemical-resistant apron when cleaning equipment, mixing, or loading

Exception: After this product has been diluted with at least 50 gallons of water, applicators for AQUATIC SURFACE APPLICATIONS must, at a minimum, wear (Note – Mixers and Loaders for this application method must still wear the Personal Protective Equipment (PPE) as described in the above section):

- Long-sleeved shirt and long pants
- Shoes plus socks
- Waterproof gloves
- Protective eyewear

Exception: At a minimum, applicators for AQUATIC SUBSURFACE APPLICATIONS must wear (Note – Mixers and Loaders for this application method must still wear the Personal Protective Equipment (PPE) as described in the above section):

- Short-sleeved shirt and short pants
- Waterproof gloves
- Chemical-resistant footwear plus socks

Discard clothing and other absorbent materials that have been drenched or heavily contaminated with this product's concentrate. Do not reuse them. Follow manufacturer's instructions for cleaning/maintaining PPE. If no such instructions for washables, use detergent and hot water. Keep and wash PPE separately from other laundry.

Engineering Control Statements

When handlers use closed systems, enclosed cabs, or aircraft in a manner that meets the requirements listed in the Worker Protection Standard (WPS) for agricultural pesticides [40 CFR 170.240(d)(4-6)], the handler PPE requirements may be reduced or modified as specified in the WPS. Mixers, loaders, and applicators using closed systems who meet these requirements may wear: long-sleeved shirt and long pants, protective eyewear, waterproof gloves, shoes plus socks, and a chemical-resistant apron when mixing, loading, or cleaning equipment. If handling tasks are performed from inside an enclosed cab or aircraft with enclosed cockpits that meet these requirements may wear: long-sleeved shirt, long pants, shoes and socks for the labeling-specified PPE. All labeling-specified PPE must be immediately available for use in an emergency. All applicable requirements as specified in 40 CFR 170.240(d)(4-6) must be followed.

User Safety Recommendations

Users should:

- Wash hands before eating, drinking, chewing gum, using tobacco, or using the toilet.
- Remove clothing immediately if pesticide gets inside. Then wash thoroughly and put on clean clothing.
- Remove PPE immediately after handling this product. Wash the outside of gloves before removing. As soon as possible, wash thoroughly and change into clean clothing.

Environmental Hazards (Terrestrial and Aquatic Uses)

This pesticide is toxic to aquatic invertebrates. **For Terrestrial Uses**, do not apply directly to water, or to areas where surface water is present, or to intertidal areas below the mean high water mark. Do not contaminate water when disposing of equipment wash waters. **For Aquatic Uses**, do not apply directly to water except as specified on this label. Treatment of dense weed areas may result in oxygen loss from decomposition of dead weeds. This loss of oxygen may cause fish suffocation. Therefore, treat only $^1/_3$-$^1/_2$ of the water body area at one time, especially if dense areas of weeds and/or algae exist, and wait 14 days between treatments.

Necessary approval and/or permits should be obtained prior to application if required. Consult the responsible State Agencies (i.e., Fish and Game Agencies or Department of Natural Resources) before making applications to public waters.

CONDITIONS OF SALE AND LIMITATION OF WARRANTY AND LIABILITY

NOTICE: Read the entire Directions for Use and Conditions of Sale and Limitation of Warranty and Liability before buying or using this product. If the terms are not acceptable, return the product at once, unopened, and the purchase price will be refunded.

The Directions for Use of this product should be followed carefully. It is impossible to eliminate all risks inherently associated with the use of this product. Crop injury, ineffectiveness or other unintended consequences may result because of such factors as manner of use or application, weather or crop conditions, presence of other materials or other influencing factors in the use of the product, which are beyond the control of SYNGENTA CROP PROTECTION, Inc. or Seller. All such risks shall be assumed by Buyer and User, and Buyer and User agree to hold SYNGENTA and Seller harmless for any claims relating to such factors.

SYNGENTA warrants that this product conforms to the chemical description on the label and is reasonably fit for the purposes stated in the Directions for Use, subject to the inherent risks referred to above, when used in accordance with directions under normal use conditions. This warranty does not extend to the use of the product contrary to label instructions, or under abnormal conditions or under conditions not reasonably foreseeable to or beyond the control of Seller or SYNGENTA, and Buyer and User assume the risk of any such use. SYNGENTA MAKES NO WARRANTIES OF MERCHANTABILITY OR OF FITNESS FOR A PARTICULAR PURPOSE NOR ANY OTHER EXPRESS OR IMPLIED WARRANTY EXCEPT AS STATED ABOVE.

In no event shall SYNGENTA or Seller be liable for any incidental, consequential or special damages resulting from the use or handling of this product. **THE EXCLUSIVE REMEDY OF THE USER OR BUYER, AND THE EXCLUSIVE LIABILITY OF SYNGENTA AND SELLER FOR ANY AND ALL CLAIMS, LOSSES, INJURIES OR DAMAGES (INCLUDING CLAIMS BASED ON BREACH OF WARRANTY, CONTRACT, NEGLIGENCE, TORT, STRICT LIABILITY OR OTHERWISE) RESULTING FROM THE USE OR HANDLING OF THIS PRODUCT, SHALL BE THE RETURN OF THE PURCHASE PRICE OF THE PRODUCT OR, AT THE ELECTION OF SYNGENTA OR SELLER, THE REPLACEMENT OF THE PRODUCT.**

SYNGENTA and Seller offer this product, and Buyer and User accept it, subject to the foregoing Conditions of Sale and Limitations of Warranty and of Liability, which may not be modified except by written agreement signed by a duly authorized representative of SYNGENTA.

Reward®

DIRECTIONS FOR USE

It is a violation of Federal law to use this product in a manner inconsistent with its labeling.

Do not apply this product in a way that will contact workers or other persons, either directly or through drift. Only protected handlers may be in the area during application. For any requirements specific to your State or Tribe, consult the agency responsible for pesticide regulation.

READ ENTIRE LABEL. USE STRICTLY IN ACCORDANCE WITH PRECAUTIONARY STATEMENTS AND DIRECTIONS, AND WITH APPLICABLE STATE AND FEDERAL REGULATIONS.

DO NOT APPLY THIS PRODUCT THROUGH ANY TYPE OF IRRIGATION SYSTEM.

AGRICULTURAL USE REQUIREMENTS

Use this product only in accordance with its labeling and with the Worker Protection Standard, 40 CFR part 170. This Standard contains requirements for the protection of agricultural workers on farms, forests, nurseries, and greenhouses, and handlers of agricultural pesticides. It contains requirements for training, decontamination, notification, and emergency assistance. It also contains specific instructions and exceptions pertaining to the statements on this label about personal protective equipment (PPE), and restricted-entry interval. The requirements in this box only apply to uses of this product that are covered by the Worker Protection Standard.

Do not enter or allow worker entry into treated areas during the restricted-entry interval (REI) of 24 hours.

PPE required for early entry to treated areas that is permitted under the Worker Protection Standard and that involves contact with anything that has been treated, such as plants, soil, or water is:

- Coveralls over short-sleeved shirt and short pants, or coveralls over long-sleeved shirt and long pants
- Waterproof gloves
- Chemical-resistant footwear plus socks
- Protective eyewear
- Chemical-resistant headgear for overhead exposure

NON-AGRICULTURAL USE REQUIREMENTS

The requirements in this box apply to uses of this product that are NOT within the scope of the Worker Protection Standard for agricultural pesticides (40 CFR part 170). The WPS applies when this product is used to produce agricultural plants on farms, forests, nurseries, or greenhouses.

Keep all unprotected persons out of operating areas or vicinity where there may be drift.

For terrestrial uses, do not enter or allow entry of maintenance workers into treated areas, or allow contact with treated vegetation wet with spray, dew, or rain, without appropriate protective clothing until spray has dried.

For aquatic uses, do not enter treated areas while treatments are in progress.

STORAGE AND DISPOSAL

Prohibitions

Do not contaminate water, food, or feed by storage, disposal, or cleaning of equipment. Open dumping is prohibited.

Storage

Keep pesticide in original container. Do not put concentrate or dilute into food or drink containers. Do not contaminate feed, foodstuffs, or drinking water. Do not store or transport near feed or food. Store at temperatures above 32°F. For help with any spill, leak, fire, or exposure involving this material, call **1-800-888-8372.**

Pesticide Disposal

Pesticide wastes are toxic. Improper disposal of excess pesticide, spray mixture, or rinsate is a violation of Federal law. If these wastes cannot be disposed of by use according to label instructions, contact your State Pesticide or Environmental Control Agency, or the Hazardous Waste representative at the nearest EPA Regional Office for guidance.

Container Disposal

Do not reuse container. Triple rinse (or equivalent). Then offer for recycling or reconditioning, or puncture and dispose of in a sanitary landfill, or incineration, or if allowed by State and local authorities, by burning. If burned, stay out of smoke.

CONTAINER IS NOT SAFE FOR FOOD, FEED, OR DRINKING WATER!

DIRECTIONS

Reward Landscape and Aquatic Herbicide is a nonvolatile herbicidal chemical for use as a general herbicide to control weeds in noncrop and aquatic areas. Absorption and herbicidal action is usually quite rapid with effects visible in a few days. Reward Landscape and Aquatic Herbicide controls weeds by interfering with photosynthesis within green plant tissue. Weed plants should be succulent and actively growing for best results. Rinse all spray equipment thoroughly with water after use. Avoid Spray Drift to crops, ornamentals, and other desirable plants during application, as injury may result. Application to muddy water may result in reduced control. Minimize creating muddy water during application. Use of dirty or muddy water for Reward Landscape and Aquatic Herbicide dilution may result in reduced herbicidal activity. Avoid applying under conditions of high wind, water flow, or wave action.

Spray Drift Management

Avoiding spray drift at the application site is the responsibility of the applicator and the grower.

The interaction of many equipment- and weather-related factors determine the potential for spray drift. The applicator and the grower are responsible for considering all these factors when making decisions.

The following drift management requirements must be followed to avoid off-target movement from aerial applications to agricultural field crops. These requirements do not apply to forestry applications, public health uses, or to applications using dry formulations.

The distance of the outermost nozzles on the boom must not exceed 3/4 the length of the wingspan or rotor.

Nozzles must always point backward parallel with the air stream and never be pointed downward more than 45 degrees.

Where states have more stringent regulations, they should be observed.

Droplet Size

The most effective way to reduce drift potential is to apply large droplets. The best drift management strategy is to apply the largest droplets that provide sufficient coverage and control. Applying larger droplets reduces drift potential, but will not prevent drift if applications are made improperly, or under unfavorable environmental conditions (See **Wind, Temperature and Humidity**, and **Temperature Inversions**).

Controlling Droplet Size

- **Volume** – Use high flow rate nozzles to apply the highest practical spray volume. Nozzles with higher rated flows produce larger droplets.
- **Pressure** – Do not exceed the nozzle manufacturer's recommended pressures. For many nozzle types, lower pressure produces larger droplets. When higher flow rates are needed, use higher flow rate nozzles instead of increasing pressure.
- **Number of Nozzles** – Use the minimum number of nozzles that provide uniform coverage.
- **Nozzle Orientation** – Orienting nozzles so that the spray is released parallel to the airstream produces larger droplets than other orientations and is the recommended practice. Significant deflection from horizontal will reduce droplet size and increase drift potential.
- **Nozzle Type** – Use a nozzle type that is designed for the intended application. With most nozzle types, narrower spray angles produce larger droplets. Consider using low-drift nozzles. Solid stream nozzles oriented straight back produce the largest droplets and the lowest drift.

Boom Length

For some use patterns, reducing the effective boom length to less than 3/4 of the wingspan or rotor length may further reduce drift without reducing swath width.

Application Height

Applications should not be made at a height greater than 10 ft. above the top of the target plants, unless a greater height is required for aircraft safety. Making applications at the lowest height that is safe reduces exposure of droplets to evaporation and wind.

Swath Adjustment

When applications are made with a crosswind, the swath will be displaced downwind. Therefore, on the up and downwind edges of the field, the applicator must compensate for this displacement by adjusting the path of the aircraft upwind. Swath adjustment distance should increase with increasing drift potential (higher wind, smaller drops, etc.).

Wind

Drift potential is lowest between wind speeds of 2-10 mph. However, many factors, including droplet size and equipment type, determine drift potential at any given speed. Application should be avoided below 2 mph due to variable wind direction and high inversion potential.
Note: Local terrain can influence wind patterns. Every applicator should be familiar with local wind patterns and how they affect spray drift.

Temperature and Humidity

When making applications in low relative humidity, set up equipment to produce larger droplets to compensate for evaporation. Droplet evaporation is most severe when conditions are both hot and dry.

Temperature Inversions

Applications should not occur during a temperature inversion because drift potential is high. Temperature inversions restrict vertical air mixing, which causes small suspended droplets to remain in a concentrated cloud. This cloud can move in unpredictable directions due to the light variable winds common during inversions. Temperature inversions are characterized by increasing temperatures with altitude and are common on nights with limited cloud cover and light to no wind. They begin to form as the sun sets and often continue into the morning. Their presence can be indicated by ground fog; however, if fog is not present, inversions can also be identified by the movement of smoke from a ground source or an aircraft smoke generator. Smoke that layers and moves laterally in a concentrated cloud (under low wind conditions) indicates an inversion, while smoke that moves upward and rapidly dissipates indicates good vertical air mixing.

Sensitive Areas

The pesticide should only be applied when the wind is blowing away from adjacent sensitive areas (e.g., residential areas, bodies of water, known habitat for threatened or endangered species, non-target crops).

Commercial Greenhouses And Nurseries

For general weed control in commercial greenhouses (beneath benches), (field grown and container stock), and other similar areas, Reward Landscape and Aquatic Herbicide may be applied preplant or postplant preemergence in field grown ornamental nursery plantings or postemergence as a directed spray. Reward Landscape and Aquatic Herbicide may also be applied preemergence in ornamental seed crops (U.S., except CA). Avoid contact with desirable foliage as injury may occur. Do not use on food or feed crops.

Spot spray: 1-2 qts. Reward Landscape and Aquatic Herbicide plus the labeled rate of a 75% or greater nonionic surfactant per 100 gals. of water, or 0.75 oz. (22 mls.) Reward Landscape and Aquatic Herbicide plus the labeled rate of a 75% or greater nonionic surfactant per 1 gal. of water.

Broadcast: 1-2 pts. Reward Landscape and Aquatic Herbicide in a minimum of 15 gals. of water per acre. Add the labeled rate of a 75% or greater nonionic surfactant per 100 gals. of spray mixture. Use an adequate spray volume to insure good coverage.

Ornamental Seed Crops (Flowers, Bulbs, Etc.) U.S., Except CA

For preharvest desiccation of ornamental seed crops. NOT FOR FOOD OR FIBER CROPS.

Broadcast (Air or Ground): 1.5-2 pts. Reward Landscape and Aquatic Herbicide plus the labeled rate of a 75% or greater nonionic surfactant per acre in sufficient water (minimum of 5 gals. by air; 15 gals. by ground) for desiccation and weed burndown. Repeat as needed at no less than 5-day intervals up to three applications. Do not use seed, screenings, or waste as feed or for consumption.

Directions For Landscape, Industrial, Recreational, Commercial, Residential, and Public Areas

Reward Landscape and Aquatic Herbicide provides fast control of broadleaf and grassy weeds in industrial, recreational, golf course, commercial, residential, and public areas.

Reward Landscape and Aquatic Herbicide is a nonselective herbicide that rapidly kills undesirable above ground weed growth in 24-36 hours. Avoid application of Reward Landscape and Aquatic Herbicide to desirable plants.

Reward Landscape and Aquatic Herbicide is a contact/desiccant herbicide; it is essential to obtain complete coverage of the target weeds to get good control. Improper application technique and/or application to stressed weeds may result in unacceptable weed control. For best results, apply to actively growing, young weeds.

Difficult weeds (such as perennial or deeply-rooted weeds) can often be controlled by tank mixing Reward Landscape and Aquatic Herbicide with other systemic-type herbicides. Refer to other product labels for specific application directions.

For residual weed control, tank mix Reward Landscape and Aquatic Herbicide with a pre-emergent herbicide labeled for the intended use site. When mixing Reward Landscape and Aquatic Herbicide with another herbicide, it is recommended to mix just a small amount first to determine if the mixture is physically compatible before proceeding with larger volumes.

Syngenta has not tested all possible tank mixtures with other herbicides for compatibility, efficacy or other adverse effects. Before mixing with other herbicides Syngenta recommends you first consult your state experimental station, state university or extension agent.

Grounds maintenance weed control: Reward Landscape and Aquatic Herbicide can be used as a spot or broadcast spray to control weeds in public, commercial and residential landscapes, including landscape beds, lawns, golf courses and roadsides. Reward Landscape and Aquatic Herbicide can also be used for weed control around the edges and nonflooded portions of ponds, lakes and ditches.

Trim and edge weed control: Reward Landscape and Aquatic Herbicide can be used to eliminate undesired grass and broadleaf plant growth in a narrow band along driveways, walkways, patios, cart paths, fence lines, and around trees, ornamental gardens, buildings, other structures, and beneath noncommercial greenhouse benches. Vegetation control with Reward Landscape and Aquatic Herbicide is limited to the spray application width. Do not exceed the labeled rate of Reward Landscape and Aquatic Herbicide as excessive rates may result in staining of concrete-based materials.

Reward Landscape and Aquatic Herbicide, since it does not translocate systemically, can be used as an edging or pruning tool when precisely applied to select areas of grass or to undesirable growth on desirable ornamental bedding plants, ground covers, etc.

Reward®

Industrial weed control: Reward Landscape and Aquatic Herbicide can be used as a spot or broadcast spray either alone or in combination with other herbicides as a fast burndown or control weeds in rights-of-ways, railroad beds/yards, highways, roads, dividers and medians, parking lots, pipelines, pumping stations, public utility lines, transformer stations and substations, electric utilities, storage yards, and other noncrop areas.

Spot spray: 1-2 qts. of Reward Landscape and Aquatic Herbicide plus the labeled rate of a 75% or greater nonionic surfactant per 100 gals. Water, or 0.75 oz. (22 mls) Reward Landscape and Aquatic Herbicide plus the labeled rate of a 75% or greater nonionic surfactant per 1 gal. of water.

Broadcast: 1-2 pts. Reward Landscape and Aquatic Herbicide in a minimum of 15 gals. of water per acre. Add the labeled rate of 75% or greater nonionic surfactant per 100 gals. spray mixture. Use an adequate spray volume to insure good coverage. Greater water volumes are necessary if the target plants are tall and/or dense. It is recommended that 60 gals. or greater water volume be used to obtain good coverage of dense weeds.

Turf Renovation (All Turf Areas Except Commercial Sod Farms)

To desiccate golf course turf and other turf areas prior to renovation, apply 1-2 pts. of Reward Landscape and Aquatic Herbicide per acre plus the labeled rate of a 75% or greater nonionic surfactant in 20-100 gals. of water (4 teaspoons of Reward Landscape and Aquatic Herbicide plus the labeled rate of a 75% or greater nonionic surfactant per 1 gal. of water) using ground spray equipment. Apply for full coverage and thorough contact with the turfgrass. Apply only when the turf is dry, free from dew and incidental moisture. For enhanced turf desiccation, especially in the case of thick turfgrass, water volumes should approach 100 gals. of water per acre.

For **suppression** of regrowth and quick desiccation of treated turfgrass, Reward Landscape and Aquatic Herbicide may be mixed with other systemic nonselective or systemic postemergence grassy weed herbicides. Refer to other product labels for specific application directions and restrictions.

Avoid spray contact with, or spray drift to, foliage of ornamental plants or food crops.

Do not graze livestock on treated turf or feed treated thatch to livestock.

Dormant Established Turfgrass (Bermudagrass, Zoysiagrass), Nonfood or Feed Crop

For control of emerged annual broadleaf and grass weeds, including Little Barley*, Annual Bluegrass, Bromes including Rescuegrass, Sixweeks fescue, Henbit, Buttercup, and Carolina Geranium in established dormant bermudagrass lawns, parks, golf courses, etc.

Apply 1-2 pts. Reward Landscape and Aquatic Herbicide per acre in 20-100 gals. of spray mix by ground as a broadcast application. Add the labeled rate of a 75% or greater nonionic surfactant per 100 gals. of spray mixture.

Bermudagrass must be dormant at application. Application to actively growing bermudagrass may cause delay or permanent injury. Users in the extreme Southern areas should be attentive to the extent of dormancy at the time of application.

*For control of Little Barley, apply Reward Landscape and Aquatic Herbicide prior to the mid-boot stage.

Aquatic Use Directions

New York – Not for Sale or Use in New York State without Supplemental Special Local Needs Labeling.

Necessary approval and/or permits should be obtained prior to application if required. Consult the responsible State Agencies (i.e., Fish and Game Agencies or Department of Natural Resources). Treatment of dense weed areas may result in oxygen loss from decomposition of dead weeds. This loss of oxygen may cause fish suffocation. Therefore, treat only $^1/_3$-$^1/_2$ of the water body area at one time and wait 14 days between treatments.

For application only to **still water** (i.e. ponds, lakes, and drainage ditches) where there is minimal or no outflow to public waters.

and/or

For applications to **public waters** in ponds, lakes, reservoirs, marshes, bayous, drainage ditches, canals, streams, rivers, and other slow-moving or quiescent bodies of water for control of aquatic weeds. For use by:

- Corps of Engineers; or
- Federal or State Public Agencies (i.e., Water Management District personnel, municipal officials); or
- Applicators and/or Licensees (certified for aquatic pest control) that are authorized by the State or Local government.

Reward®

Treated water may be used according to the following table or until such time as an approved assay (example: PAM II Spectromatic Method) shows that the water does not contain more than the designated maximum contaminant level goal (MCLG) of 0.02 mg./l. (ppm) of diquat dibromide (calculated as the cation):

Water Use Restrictions Following Applications With Reward Landscape And Aquatic Herbicide (Days)

Application Rate	Drinking	Fishing and Swimming	Livestock Consumption	Spray Tank Applications** and Irrigation to Turf and Ornamentals	Spray Tank Applications** and Irrigation to Food Crops
2 gals./surface acre	3 days	0	1 day	3 days	5 days
1 gal./surface acre	2 days	0	1 day	2 days	5 days
0.75 gal./surface acre	2 days	0	1 day	2 days	5 days
0.50 gal./surface acre	1 day	0	1 day	1 day	5 days
Spot Spray* (< 0.5 gal./surface acre)	1 day	0	1 day	1 day	5 days

*Rates refer to total surface area.

**For preparing agricultural sprays for food crops, turf or ornamentals (to prevent phytotoxicity), do not use water treated with Reward Landscape and Aquatic Herbicide before the specified time period.

When the contents of more than one spray tank is necessary to complete a single aquatic application, no water holding restrictions apply between the consecutive spray tanks.

No applications are to be made in areas where commercial processing of fish, resulting in the production of fish protein concentrate or fish meal, is practiced. Before application, coordination and approval of local and/or State authorities must be obtained.

Apply Reward Landscape and Aquatic Herbicide in Accordance With the Following Table

Weed Species	Subsurface or Bottom Placement Gals./Surface Acre*	Surface Gals./Surface Acre*
Bladderwort (*Utricularia* spp.)	1-2	2
Coontail (*Ceratophyllum demersum*)	2	2
Elodea (*Elodea* spp.)	2	2
Naiad (*Najas* spp.)	1-2	2
Pondweeds[1] (*Potamogeton* spp.)	2	2
Watermilfoils (*Myriophyllum* spp.)	1-2	2
Hydrilla (*Hydrilla verticillata*)	2	2
Waterlettuce[2] (*Pistia Stratiotes*)	NA	0.5 - 0.75
Waterhyacinth[2] (*Eichhornia crassipes*)	NA	0.5 - 0.75
Pennywort[3] (*Hydrocotyle* spp.)	NA	0.5 - 0.75
Frog's Bit[6] (*Limnobium spongia*)	NA	0.5 - 0.75
Salvinia[2] (*Salvinia* spp.)	NA	0.5 - 0.75
Duckweed[4] (*Lemna* spp.)	NA	1
Cattails[3] (*Typha* spp.)	NA	1-2
Algae[5] (*Spirogyra* spp. & *Pithophora* spp.)	1-2	2

*For water less than or equal to 2 ft. in average depth of treatment area, use a maximum of 1 gal. Reward Landscape and Aquatic Herbicide per surface acre. Lowest rates should be used in shallow areas where the water depth is considerably less than the average depth of the entire treatment area, for example, shallow shoreline areas. At water temperatures below 50°-60°F, efficacy and immediacy of results may be reduced.

[1] Reward Landscape and Aquatic Herbicide controls *Potamogetan* species except Richardson's pondweed (*P. richardsonii*). For control of *P. robbinsii,* applications must be made when the plants are in the early stages of growth such as in Spring and early Summer.

[2] For salvinia, waterlettuce, and water hyacinth, use the labeled rate of Reward Landscape and Aquatic Herbicide in 75-200 gals. water plus the labeled rate of a 75% or greater nonionic surfactant per acre for surface sprays, and for aerial application for waterlettuce and water hyacinth control, apply the labeled rate of Reward Landscape and Aquatic Herbicide in 10-24 gals. of water plus the labeled rate of a 75% or greater nonionic surfactant per acre.

[3] For Pennywort and cattail control, apply in 50-150 gals. of water plus the labeled rate of a 75% or greater nonionic surfactant per acre for full coverage and thorough weed contact. Repeat treatments may be necessary to control regrowth. For best results, apply before flowering (cattail).

[4] For duckweed control, apply as an overall spray in 50-150 gals. of water plus the labeled rate of a 75% or greater nonionic surfactant per acre. Retreatment may be necessary for plants missed in previous applications and regrowth.
[5] For suppression of certain filamentous algae species including *Spirogyra* and *Pithophora,* apply according to the submersed use directions.
[6] Not for use in California.

Application: In mixed weed populations, use the high rate of application as indicated by weeds present.

Subsurface Applications: Where the submersed weed growth, especially Hydrilla, has reached the water surface, apply either in a water carrier or an invert emulsion through boom trailing hoses carrying nozzle tips to apply the dilute spray below the water surface to insure adequate coverage.

Bottom Placement: Where the submersed weeds, especially Hydrilla, Bladderwort, and Coontail growth, have reached the water surface or where water is slowly moving through the submersed weed growth that has reached the water surface, especially Hydrilla, Bladderwort, and Coontail, control may be enhanced when applied in an invert emulsion carrier injecting diluted Reward Landscape and Aquatic Herbicide near the bottom with weighted hoses. The addition of a copper-based algaecide will improve control. Where algae are present along with the submersed weeds, pretreatment with copper-based algaecide at recommended rates is advised for best results.

Surface Application: For submerged aquatic weeds, apply Reward Landscape and Aquatic Herbicide either as concentrate slowly poured directly from the container in strips or as a spray in sufficient carrier. Applications should be made to ensure complete coverage of the weed areas. In mixed weed populations, use the high rate of application as indicated by weeds present.

If posting is required by your state or tribe – consult the agency responsible for pesticide regulations for specific details.

General Recommendations for "Posting Notification"*

- **Flowing water:** "post" the restricted area (within/at 1,600 ft. downstream of treatment) for the duration of the water use restriction.
- **Standing water:** "post" the restricted area (within/at 1/4 mile of treatment) for the duration of the water use restriction.
- **No "posting" is necessary where water use is greater than 1,600 feet downstream of treated water in flowing water bodies or where water use is greater than 1/4 mile from treated water in standing water bodies.**

*"Posting" should be removed at the end of the restriction period.

For non-emergency (e.g., current product information), call
Syngenta Crop Protection at 1-800-334-9481.

Product of United Kingdom
Formulated in the USA

Syngenta Crop Protection, Inc.
Greensboro, North Carolina 27409
www.syngenta-us.com
SCP 1091A-L2 0901
154290

Reward®

REWARD®

Landscape and Aquatic Herbicide

TO PREVENT ACCIDENTAL POISONING, NEVER PUT INTO FOOD, DRINK, OR OTHER CONTAINERS, AND USE STRICTLY IN ACCORDANCE WITH ENTIRE LABEL.

DO NOT USE THIS PRODUCT FOR REFORMULATION.

Active Ingredient: Diquat dibromide [6,7-dihydrodipyrido (1,2-a:2',1'-c)pyrazinediium dibromide]	37.3%
Other Ingredients:	62.7%
Total:	100.0%

Contains 2 lbs. diquat cation per gal. as 3.73 lbs. salt per gal.

See directions for use in attached booklet.

AGRICULTURAL USE REQUIREMENTS

Use this product only in accordance with its labeling and with the Worker Protection Standard, 40 CFR part 170. Refer to supplemental labeling under "Agricultural Use Requirements" in the Directions for Use section for information about this standard.

EPA Reg. No. 100-1091
EPA Est. 100-TX-001

Product of United Kingdom
Formulated in the USA

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Syngenta Crop Protection, Inc.
Greensboro, North Carolina 27409
www.syngenta-us.com

SCP 1091A-L2 0901
154290

KEEP OUT OF REACH OF CHILDREN.

WARNING/AVISO

Si usted no entiende la etiqueta, busque a alguien para que se la explique a usted en detalle. (If you do not understand the label, find someone to explain it to you in detail.)

Precautionary Statements

Hazards to Humans and Domestic Animals

May be fatal if absorbed through skin. Harmful if swallowed or inhaled. Causes substantial, but temporary, eye injury. Causes skin irritation. Contact with irritated skin, or a cut, or repeated contact with intact skin may result in poisoning. Do not get in eyes, on skin, or on clothing. Avoid breathing vapor or spray mist. Do not feed forage from treated crops to livestock. Keep livestock and pets out of treated fields and crop areas.

FIRST AID

If swallowed: Call a Poison Control Center or doctor immediately for treatment advice. Immediately give water or milk to drink and induce vomiting by inserting finger in throat. Do not induce vomiting or give anything by mouth to an unconscious person. Take person and product container to the nearest hospital or physician fast. PROMPT TREATMENT IS ESSENTIAL TO COUNTERACT POISONING and should be initiated before signs and symptoms of injury appear.

If on skin or clothing: Take off contaminated clothing. Rinse skin immediately with plenty of water for 15-20 minutes. Call a Poison Control Center or doctor for treatment advice.

If in eyes: Hold eye open and rinse slowly and gently with water for 15-20 minutes. Remove contact lenses, if present, after the first 5 minutes, then continue rinsing eye. Call a Poison Control Center or doctor for treatment advice.

If inhaled: Move person to fresh air. If person is not breathing, call 911 or an ambulance, then give artificial respiration, preferably mouth-to-mouth, if possible. Call a Poison Control Center or doctor for further treatment advice.

NOTE TO PHYSICIAN: CALL SYNGENTA MEDICAL EMERGENCY ASSISTANCE 1-800-888-8372 at any hour to obtain toxicology information and a diquat analysis. To be effective, treatment for diquat poisoning must begin IMMEDIATELY. Treatment consists of binding diquat in the gut with suspensions of activated charcoal or bentonite clay, administration of cathartics to enhance elimination, and removal of diquat from the blood by charcoal hemoperfusion or continuous hemodialysis.

Have the product container or label with you when calling a poison control center or doctor, or going for treatment.

HOT LINE NUMBER: For 24-Hour Medical Emergency Assistance (Human or Animal) or Chemical Emergency Assistance (Spill, Leak, Fire, or Accident), Call **1-800-888-8372**

Environmental Hazards (Terrestrial and Aquatic Uses)

This pesticide is toxic to aquatic invertebrates. For **Terrestrial Uses,** do not apply directly to water, or to areas where surface water is present, or to intertidal areas below the mean high water mark. Do not contaminate water when disposing of equipment wash waters. For **Aquatic Uses,** do not apply directly to water except as specified on this label. Treatment of dense weed areas may result in oxygen loss from decomposition of dead weeds. This loss of oxygen may cause fish suffocation. Therefore, treat only $^1/_3$-$^1/_2$ of the water body area at one time, especially if dense areas of weeds and/or algae exist, and wait 14 days between treatments.

Necessary approval and/or permits should be obtained prior to application if required. Consult the responsible State Agencies (i.e., Fish and Game Agencies or Department of Natural Resources) before making applications to public waters.

STORAGE AND DISPOSAL

Prohibitions

Do not contaminate water, food, or feed by storage, disposal, or cleaning of equipment. Open dumping is prohibited.

Container Disposal

Do not reuse container. Triple rinse (or equivalent). Then offer for recycling or reconditioning, or puncture and dispose of in a sanitary landfill, or incineration, or if allowed by State and local authorities, by burning. If burned, stay out of smoke.

CONTAINER IS NOT SAFE FOR FOOD, FEED, OR DRINKING WATER!

2.5 gallons
U.S. Standard Measure

syngenta

Diquat

Volume 4, Section 2

CHEMICAL CHARACTERISTICS

10 PAGES

TABLE OF CONTENTS

2.0 CHEMICAL CHARACTERISTICS

The physical/chemical data in the following section are those required by USEPA when a product is registered for use in the US as a pesticide. These characteristics assist in the basic understanding of the molecule and are later used in predicting environmental behavior or are considered when higher tiered studies are designed or requested. Pure active ingredient or technical grade active ingredient (TGAI) refers to the active compound(s), which cause the desired biological effect when applied to a target system. Manufacturing Use Products (MUP) are the TGAI with additional added ingredients. Typically, the added ingredients are solvents, safeners or diluents. The technical grade active ingredient and the manufacturing use products are typically formulated into end-use products, also known as formulated products. The end-use products consist of a known percentage active ingredient plus a solvent or solid carrier and may include surface active components to aid in dissolution, emulsification, suspension, *etc.*, of the active ingredient. The technical grade product of diquat dibromide consists of the diquat dibromide molecule dissolved in water.

2.1 DIQUAT

Diquat dibromide [6,7-dihydrodipyrido(1,2-a:2',1'-c) pyrazinediium dibromide] is the active component in aquatic herbicides used in static and flowing water to control aquatic weeds. It is also registered for a number of terrestrial uses including broadleaf and grass plant control and desiccation of potatoes and certain crops. Diquat is a non-selective contact herbicide, desiccant, defoliant and growth regulator for seed crops. It is rapidly absorbed by green plant tissues, which are killed on exposure to light. It possesses some systemic properties. Its mode of action is described as a photosynthetic electron flow diverter. Compounds in this group result in rapid disruption of cell membranes and very rapid kill. The bipyridyliums penetrate into the cytoplasm, cause the formation of peroxides and free electrons (light is required) which destroy the cell membranes almost immediately. Rapid destruction of cell membranes prevents translocation to other regions of the plant. (EPA, 1995) (Purdue, 2000)(Worthing, 1991)

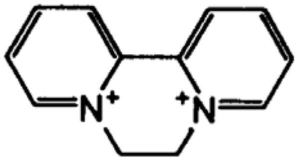
Diquat is formulated as a solution in water. The primary concentrated end-use product is Reward® Landscape and Aquatic Herbicide (EPA Reg. No. 10182-404). Reward® may be used to control a number of submerged species as well as several species of emergent weeds.

2.1.1 Composition

Diquat dibromide is a quaternary ammonium compound containing two bridged pyridine rings. In its pure form it is a pale yellow solid with a melting point of 335-340°C. In commercial trade, however, diquat dibromide is found as an aqueous solution.

- **Active Ingredient**

Common name:	Diquat dibromide
CAS Registry No.:	85-00-7
Chemical name:	6,7-dihydrodipyrido[1,2-a:2',1' -c]pyrazinediium dibromide
Empirical formula:	$C_{12}H_{12}Br_2N_2$
Molecular weight:	344.07
Structure:	



- **Impurities**

There are no known impurities identified by the manufacturers or the US EPA which are known to be of toxicological or environmental concern. The US EPA has established guidelines that require that impurities of concern, such as N-nitrosoamines, ethylenedibromide and chlorinated dioxins and furans must be disclosed. No such compounds are present in diquat dibromide products.

The primary intentionally added inert or “other” ingredient in diquat dibromide formulations is water. Other formulation ingredients, when included in the end-use products, have been reviewed by the USEPA and approved when used for their intended purpose, however, these are not reported, as they are confidential manufacturing information.

The USEPA has established a category listing system for the “other” (inert) compounds used in pesticide formulations. The lists are designated 1, 2, 3, 4a and 4b. Compounds are assigned to the various lists according to their toxicological concern and to the extent their safety has been reviewed by the Agency. In the case of each list, if USEPA determines that a compound is no longer used in any pesticide formulation, it will be removed from the list.

List 1 contains eight compounds, which, due to their toxicological profile, require special labeling if used in a pesticide formulation. These compounds are generally not used in pesticidal formulations any longer. There are no List 1 compounds in the diquat formulations used in the State of Washington.

List 2 compounds are those for which USEPA has not yet determined a full profile but is reviewing existing information. At the completion of their evaluation, it is expected that the compounds still in use in pesticide formulations will be moved to List 1 or to List 4. T here are no List 2 compounds in the diquat formulations used in the state of Washington.

List 3 contains those compounds which have not been fully evaluated, but which have profiles of lesser concern in the USEPA evaluation scheme. It is expected that most of these compounds will be moved to List 4 once their evaluation by the Agency is complete. There are no identified List 3 inert compounds in the diquat products.

List 4 is divided into two categories. List 4A contains compounds generally regarded as safe for use in pesticide formulations and includes such compounds as corn cobs and attapulgite clay. List 4B contains those compounds that have sufficient data on file at EPA to substantiate that they can be used safely in pesticide products.

In addition to the above-mentioned review by the USEPA, all registered pesticidal end-use products (the products actually applied to the environment to control weeds or pests) must undergo a series of toxicological tests to establish their safety. Because these tests are performed on the actual end-use formulation, the effects of the "other" ingredients are effectively tested simultaneously. This toxicological screen of the "other" compounds affords an additional opportunity to examine comparative data on the active ingredient versus the end-use product to determine if there is a need to test each of them in a complete testing battery.

- **Intentionally added inert ingredients**

  Intentionally added inert or "other" ingredients in diquat formulations include water, which serves as the primary diluent/solvent in the liquid product.

2.1.2 Color

Color is an end-point observation of the product used to assist in identification.

	Color	Citation
Diquat Dibromide MUP	Very Dark Brown	(Hart, 1987)
Reward ® Landscape and Aquatic Herbicide	Very Dark Brown	(Hart, 1987)

2.1.3 Physical State

Physical state is an end-point observation of the product, solid, liquid or gaseous used to assist in identification.

	Physical State	Citation
Diquat Dibromide MUP	Clear Liquid	(Hart, 1987)
Reward ® Landscape and Aquatic Herbicide	Liquid	(Hart, 1987)

2.1.4 Odor

Odor is an end-point observation of the product used to assist in identification. Odor may also serve as a warning in cases where odorants are added as a safety factor.

	Odor	Citation
Diquat Dibromide MUP	Earthy	(Hart, 1987)
Reward ® Landscape and Aquatic Herbicide	Earthy	(Hart, 1987)

2.1.5 Melting Point

The melting point is a physical end point observation used for identification of pure compounds and may provide some indication of thermal stability. For the pure acid active ingredient (diquat dibromide) the melting point is not defined as the product is a liquid. Melting point is not applicable to the formulations because they are liquids. However, Extoxnet reports it decomposes at temperatures above 333° C.

2.1.6 Boiling Point

The boiling point is a physical end point observation for identification of pure compounds. The boiling point for the pure acid active ingredient is essentially that of water as the MUP consists of diquat dibromide dissolved in water. The boiling points for the liquid formulations are essentially the same as water, 100°C as they are aqueous based.

2.1.7 Density, Bulk Density or Specific Gravity

Bulk density is a measure of the weight per unit volume of the product and is useful for physical identification or differentiation of two similar products. The value may also be needed to calculate application rates in some instances. Density is typically reported as grams per cubic centimeter at 25°C. The density of the end-use product is essentially the same as the MUP as there is only a small difference in amount of water in the two products.

	Density (g/cc)	Citation
Diquat Dibromide MUP	1.26	(Hart, 1987)
Reward ® Landscape and Aquatic Herbicide	~1.26	(Hart, 1987)

2.1.8 Solubility

Solubility is a physical end point useful for understanding potential environmental impact. High water solubility is frequently associated with mobility and affects distribution in water and soil. This endpoint is determined for the active ingredient in a product and is typically reported as grams per 100 ml water at 25°C.

	Solubility in Water @ 25°C (g/100 ml)	Citation
Diquat Dibromide MUP	>70	(Hart, 1987)
Reward ® Landscape and Aquatic Herbicide	>70	(Hart, 1987)

2.1.9 Vapor Pressure

Vapor pressure is a physical end point useful for understanding the distribution of the active ingredient between water/soil and air. High volatility is an indication of potential impact in the air compartment. This endpoint is determined for the active ingredient in a product and is typically reported as mm mercury (Hg) at a specified temperature.

	Vapor Pressure @ 24.3°C (mm Hg)	Citation
Diquat Dibromide MUP	$< 10^{-7}$	(Worthing, 1991)
Reward ® Landscape and Aquatic Herbicide	$< 10^{-7}$	(Worthing, 1991)

1.1

2.1.10 Disassociation Constant

Disassociation constant is a physical end point used to assess the distribution of the product in aqueous media. There are no dissociable hydrogens associated with this molecule; therefore there is no data to report.

2.1.11 Octanol/Water Partition Coefficient

Octanol/Water partition coefficient is a physical end point used to assess the potential of a compound to bioaccumulate in the environment. The value represents the ratio of product in octanol versus water at equilibrium at 25°C. Values less than 10 indicate little or no likelihood of bioaccumulation.

	Octanol/Water Coefficient (Kow)	Citation
Diquat Dibromide MUP	0.000025	(Worthing, 1991)
Reward ® Landscape and Aquatic Herbicide	0.000025	(Worthing, 1991)

2.1.12 pH

pH is a physical end point used to identify the product and to assess the potential effect of the equilibrium in the environment. For diquat dibromide MUP, the value is reported for the undiluted product at 21°C. The value for the end-use product is essentially the same as there is only a small difference in the amount of water in the two products.

	pH	Citation
Diquat Dibromide MUP	6.68	(Hart, 1987)
Reward ® Landscape and Aquatic Herbicide	6.68	(Hart, 1987)

2.1.13 Stability

Stability is a chemical evaluation of the product to assess the potential effect of heat, light, metals and metal ions on the active ingredient. In the case of diquat dibromide MUP there may be significant corrosion of metals including iron, aluminum and zinc. Additional discussion of compatibility may be found in section 2.1.20, Corrosion Characteristics below. (Hart, 1987).

2.1.14 Oxidizing or Reducing Action

Oxidizing or reducing action is an assessment of the potential for a compound to react with common oxidizers or reducers. Diquat dibromide is not reactive toward water or carbon dioxide, however, it reacts with both zinc and potassium permanganate. It can act as both an oxidizer and a reducer. (Hart, 1987)

2.1.15 Flammability

Determination of flammability is measurement of the temperature that will sustain a flame and is used to classify the product for hazard in storage and shipping. Determination of flammability is not required for technical grade products. The formulated products are aqueous and will not support combustion. (Hart, 1987)

2.1.16 Explodability

Determination of explodability is measurement of the potential for a compound to explode when exposed to physical or thermal shock. Determination of explodability is not required for technical grade products or manufacturing-use products. The formulated products are aqueous based and are not explosion hazards. Additionally, the diquat dibromide molecule contains no oxygen or explodable functional groups. It does not become explosive when the water is removed. (Hart, 1987)

2.1.17 Storage Stability

Storage stability is the physical determination of the stability of the active ingredient when stored in its commercial packaging over extended time periods, usually one to two years or more. Diquat dibromide has been shown to be stable for at least 1.5 years when stored in polyethylene at 25°C. (Hart, 1987).

2.1.18 Viscosity

Viscosity is a physical end-point measurement used to identify the product and to assess the ability of the product to be poured or pumped. The measurement is not required on technical grade products or on solid products. The viscosity is reported in centipoise at 20°C.

	cP	Citation
Diquat Dibromide MUP	1.77	(Fletcher, 1995)
Reward ® Landscape and Aquatic Herbicide	1.77	(Fletcher, 1995)

2.1.19 Miscibility

Miscibility is a physical assessment of the ability of a formulated product to mix with spray oils for use during application. Since the diquat aquatic products are not labeled for application in oil, this data requirement is not applicable.

2.1.20 Corrosion Characteristics

Corrosion characteristics require the physical observation/measurement of the effects of the product on the commercial packaging. Measurements of the weight, deformation and strength of the packaging are reported. For diquat dibromide there is significant corrosion of metals such as iron, zinc and aluminum. In addition, it may cause stress cracking in some plastics. The following materials have been determined to be compatible with diquat dibromide: Atlac 382 and Derakane 470 (plastics), polyethylene, butyl rubber, chlorobutyl rubber, bromobutyl rubber, Mebonite, titanium, glass, enamel or glass lined mild steel and stainless steels AISI 316, 321 and 304. (Hart, 1987)

2.1.21 Dielectric Breakdown Voltage

Dielectric breakdown voltage is the physical measurement of the effect of an electric arc on the stability of the formulated product. This requirement applies only to formulations that are applied around electrical equipment or apparatus. As there is no likelihood of open electrical apparatus in the aquatic environment, this test is not applicable.

2.1.22 Photodegradation

EPA reports that diquat will photodegrade in surface layers of water in 1-3 or more weeks when not adsorbed to particulate matter. http://www.epa.gov/ogwdw/dwh/t-soc/diquat.html .

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Diquat

Volume 4, Section 3

ENVIRONMENTAL FATE

81 PAGES

TABLE OF CONTENTS

3.0 DIQUAT

Diquat has been used as an herbicide in both terrestrial and aquatic weed control for a number of years. Due to its high profile and increasing use as an aquatic herbicide, there have been a number of studies conducted since the original diquat Environmental Impact Statement (EIS) was issued in 1993, to determine the fate and behavior of diquat in aquatic environments. For ease of reference, this update incorporates data from the earlier EIS in addition to information from pre-1990 sources not cited in that document, and from references published since 1990. In addition, several "registration" studies performed by and for registrants are cited.

The Washington State Department of Ecology is only considering diquat dibromide for use in aquatic weed management. Most work has been conducted on diquat dibromide or diquat dibromide monohydrate. Diquat cation, is the active substance in diquat dibromide, diquat dibromide monohydrate and diquat chloride. Attention should be paid to the differences in hydrolysis, photolysis, and other results in this section. Data generated from sources of diquat ion other than diquat dibromide or diquat dibromide monohydrate should be interpolated with caution when predicting diquat behavior in the environment.

3.1 VOLATILIZATION

No test data were found regarding diquat dibromide volatilization. The following information was obtained from the 9th edition of the Pesticide Manual (Worthing and Hance et al, 1991) and the EPA (EPA, RED, 1995) summarizes the volatility issue well as follows.

Available data indicate that the vapor pressure of diquat dibromide is essentially zero (ARS, 1995) and diquat dibromide can be considered non-volatile. At 25°C, the vapor pressure of diquat dibromide is $<4\times10^{-9}$ mmHg (EPA RED, 1995). Since the solubility of diquat dibromide is reported as 7.00×10^{5} ppm (700g/L) (Worthing and Hance, 1991), the Henry's Law constant for diquat dibromide was calculated to be $<2.6\times10^{-15}$ atm• meter^3/mole. This is somewhat misleading since diquat is not believed to partition significantly from water to air. Given these characteristics, the volatilization half-life for diquat is essentially infinite in an aquatic system at 25°C and 1-meter depth. So diquat dibromide can be considered non-volatile (Reinert and Rogers, 1987 and Wauchope et al, 1992).

No additional studies were found regarding the volatility and water solubility of diquat dibromide or diquat dibromide monohydrate.

3.2 HYDROLYSIS

Summary: Diquat dibromide was stable to hydrolysis in sterile buffered water. Half-lives cannot be measured since degradation of diquat dibromide was reported as being insignificant. Therefore, the hydrolysis half-life can be presumed to be greater than 30 days and probably greater than a year in sterile buffered water at pHs of 5, 7 and 9. No data were found regarding a specific time for the diquat cation half-life, but it is generally regarded as stable to hydrolysis. Since the pH of most natural water ranges from 6 to 9, with higher values during higher bio-productivity in summer, diquat dibromide and diquat cation will not be hydrolyzed when used in rivers, canals, reservoirs, lakes and ponds.

Hydrolysis refers to the chemical interaction of the agrochemical with water as a mechanism of agrochemical breakdown. While aqueous or aquatic (the terms are synonymous in this review) persistence studies are sometimes conducted in natural water bodies, true hydrolysis studies are conducted in laboratories using sterile distilled or deionized buffered water so that the chemical effects of an aqueous environment can be isolated from biological, sunlight, or sediment interactions. Aquatic persistence in natural water is addressed in Section 3.2.5.

Laboratory hydrolysis studies for EPA submission are typically performed with radioactive ^{14}C-diquat dibromide at three pH values (pH 5, pH 7, pH 9 corresponding to slightly acid, neutral, and mildly alkaline, respectively) in sterile buffered water for a period of 30 days at 25°C (EPA, 1995). Sampling for breakdown products and the remaining concentration of parent material occurs at frequent intervals.

Diquat dibromide is very soluble in water and immediately dissociates to diquat cation. Therefore, diquat cation is immediately available as a contact herbicide dissolved in the water column. Since diquat cation is not hydrolyzed, it remains available as a contact herbicide until it is removed from the water by other physical methods (photolysis, sorption to soil, sorption to sediment, sorption to plant material or biological breakdown by microorganisms).

3.2.1 Half-life

EPA (1985) reviewed diquat environmental fate literature. They cited early references indicating that diquat acid is stable in water. Diquat dibromide may be removed from water by sorption to clay containing sediments (Weber et al 1965 in Reinert and Rogers, 1992), sorption to seston (suspended sediment) found in the water column (Bowmer, 1982), sorption onto plants and algae, degradation by microbes found in soil and potentially also associated with the surfaces of plants and the organic components of sediment (Baldwin and Calderbank, 1968 and Burns and Audus, 1970 in Simsiman et al, 1976) or energy inputs such as photolysis ultraviolet light (EPA RED, 1995). In an aqueous photolysis study mimicking the effects of Florida spring sunlight, diquat dibromide can be considered to be photolytically stable; diquat degraded with a calculated half-life of 74 days (EPA RED, 1995). However, diquat dibromide was very stable in buffered water shielded from all light sources.

Table 3.2.1 illustrates the pH-dependency of diquat dibromide. Half-lives for diquat dibromide could not be measured because it is stable to hydrolysis in buffered water at pHs 5, 7 and 9. No degradation of diquat dibromide was seen during the course of the study (EPA RED, 1995). If degradation occurs, the hydrolysis half -life can be predicted by the following pseudo first-order rate equation:

$$T(1/2) = 0.693/(K_b)$$

where K_b = pseudo first order hydrolysis rate constant (day^{-1})

Since the [OH] concentration is high relative to the concentration of diquat dibromide, its concentration can be ignored.

The pseudo first order hydrolysis rate constant is essentially zero since diquat dibromide does not readily hydrolyze. However, it has been reported by Gangstad (1986) that diquat dibromide is not stable under alkaline conditions; however, since no hydrolysis occurs between pH 5 and pH 9, it is assumed that only extremely high pH or alkalinity would hydrolyze diquat. The pH of most natural waters is approximately 6 to 9, particularly during the summer months. Therefore, diquat dibromide can be expected to degrade very slowly (if at all) due to hydrolysis, when applied to lakes and ponds. It is possible that the bacteria associated with plant surfaces may be able to degrade diquat dibromide. Simsiman and Chesters (1976) determined that bacteria associated with dead and dying plants were able to degrade 32% of applied diquat to water-soluble metabolites within 22 days. The remainder of the applied test material (^{14}C-diquat dibromide) was either volatilized (48%) or bound to the sediment (19%). These plant-associated bacteria degraded over 80% of the applied test substance to volatile metabolites and water-soluble metabolites in 22 days. However, after diquat dibromide is released from plant tissue and becomes bound to sediment, further degradation by microbes occurs very slowly. The rate of degradation after association with sediment that might be typically found in the environment was estimated by Wauchope (1992) to be very low (<0.00069 day^{-1}). The half-life of diquat dibromide under these bound conditions may be in excess of 1,000 days. This form of diquat is not biologically active.

3.2.2 Degradation Products

There are no significant degradation products due to the hydrolysis of diquat dibromide at pH 5, 7 and 9. Bacterial degradation products that occurred on the surface of plants consisted of unidentified water-soluble metabolites. In weed-free systems, carbon dioxide was produced under aerobic conditions with small volumes of overlying water. However, this accounted for only about 8% of the diquat dibromide after degradation for 180 days in Lake Mendota (Wisconsin) sediment. The remainder of the sediment-bound residue was unchanged diquat cation. These experiments are typically conducted with ^{14}C-radiolabeled diquat, which makes the pesticide and its degradates easier to detect and quantify (Simsiman and Chesters, 1976). No data were located regarding the specific hydrolysis half-life of diquat cation or its hydrolysis products. From the structure of the molecule, little hydrolysis is expected in the pH ranges found in natural waters before the cation is degraded by other mechanisms, such as photolysis and microbial metabolism.

Table 3.2
Hydrolysis of Diquat (Laboratory Studies)

Matrix	Compound	pH	Temp	Half-life (DT_{50})	Reference
Sterile buffered water*	Diquat dibromide	5	~25°C	Very long Cannot be measured. No degradation in 30 days	EPA RED, 1995
Sterile buffered water*	Diquat dibromide	7	~25°C	Very long Cannot be measured. No degradation in 30 days	EPA RED, 1995
Sterile buffered water*	Diquat dibromide	9	~25°C	Very long Cannot be measured. No degradation in 30 days	EPA RED, 1995

* EPA guideline study

3.3 AQUEOUS PHOTOLYSIS

Summary: Several reports on the photolysis of diquat were reported. However, only minimal data was presented on the effects of sunlight, or light with the same spectrum as sunlight. In a study that followed EPA guidelines, ^{14}C-diquat was considered to be pholytically stable in the environment; diquat was degraded with a half-life of 74 days under artificial light that was similar to Florida spring sunlight. In another experiment, diquat was exposed to sunlight from May to June under a cloudless Saskatchewan sky; under these conditions a 5 ppm solution of diquat degraded to 10% of the original concentration in 5 weeks producing no volatile degradates. Other studies exposed diquat to artificial light that was of a much shorter wave length (<290 nm) than typical sunlight. These exposures produced up to 9 photodecomposition products. However, except for one of the products, these decomposition products may not be environmentally relevant

The major product of diquat photodecomposition is TOPPS (1,2,3-tetrahydro-1-oxo-pyridol [1,2-a]-5 pyrazinium salt), which constituted 12% of the applied radioactivity after 32 days. Although small amounts of picolinamide, picolinic acid and water-soluble volatile fragments can be produced from TOPPS after exposure to sunlight, the concentrations of these metabolites were considered to be inconsequential.

The presence of adsorbents in sandy loam, kaolinite clay or montomorollonite clay appears to retard the degradation of diquat. Although there was little loss of radioactivity from these sorbents, no effort was made to determine whether there was any change from the original molecule to ^{14}C-labeled degradates.

As with hydrolysis, photolysis testing is carried out in a laboratory. Vessels containing solutions of the herbicide in sterile distilled or deionized and buffered water are irradiated with either a mercury vapor lamp or natural sunlight. Identical vessels are kept in the dark for the duration of the study and also sampled in order to compensate for the effects of any hydrolysis occurring. Testing is usually carried out at 25°C, at pH 5, 7 and 9, but this is not always the case, particularly with compounds that are hydrolytically stable or unstable at all pHs. Other photolysis testing, such as photolysis of a pesticide on the surface of a soil, is also required by the EPA for products that might be incidentally applied to soil, as is the case for diquat.

The purpose of photolysis experiments is to isolate the effect of sunlight, specifically the ultraviolet and near-ultraviolet part of the spectrum, on the degradation of an herbicide without biological or chemical interactions. Natural sunlight's visible spectrum covers wavelengths from about 800 nm (deep red) to about 300 nm (deep violet). Generally speaking, only light in the violet and ultraviolet end of the spectrum has enough energy to initiate or influence chemical reactions ("photochemical reactions"). Air, as well as ozone, strongly filters near-ultraviolet and ultraviolet radiation, and cuts off nearly all radiation below 290 nm wavelength. Water is transparent to radiation down to approximately 180 nm (far ultraviolet), assuming that there are no suspended solids or dissolved colored material, such as humic acids, to impair passage of the light. Aqueous photolysis experiments done under natural sunlight are influenced by the season, latitude, time of day, depth of the water column and ozone thickness (Zepp and Cline, 1977).

3.3.1 Half-life

Table 3.2.2 summarizes photolysis data for diquat. Photolysis testing is normally carried out on diquat dibromide, and only a few studies were found that addressed diquat dibromide in absence of adsorbents. The half-life for diquat exposed for 32 days to artificial light with the intensity and wavelength of Florida spring sunlight was 74 days. This value allows one to conclude that diquat can be considered to be photolytically stable.

Studies conducted under actual sunlight with 5-weeks exposure to May and June sunlight typically found in Saskatchewan under cloudless skies yielded a half-life of 11.2 days (Smith and Grove, 1969 and Reinert and Rogers, 1987). Such a short photolysis half-life indicates that sunlight may be important in the degradation of diquat under circumstances of long days in the late spring to early summer.

Environmental characteristics, such as degree on cloud cover, season, latitude and ozone thickness can impact the rate of degradation on half-life of a pesticide exposed to sunlight. However, experimental parameters such as the type of buffers used and the size and depth of the photolysis chamber may also influence the rate of degradation. Since the photolyisis chamber used by Smith and Grove (1969) was a shallow petri dish and not the standard 250-ml beaker, the rate of hydrolysis may have been faster than in the study reported by EPA (EPA RED, 1995).

Photo-degradation of diquat dibromide has also been reported on soils. EPA (EPA RED, 1995) has reported that ^{14}C-diquat did not degrade after exposure to artificial sunlight with the equivalent energy of 32 days of natural sunlight. The only compound that was identified after extraction and chromatography was diquat. This indicates that soil/sediment will protect diquat from the degradative effects of sunlight.

Funderburk et al (1966) found that ^{14}C-diquat was stable when sorbed onto five grams of, sandy loam, kaolinite clay or montmorillonite clay and then exposed to UV light (240 to 260 nm). Very little (13, 8 and 6%, respectively) loss of applied radiation occurred during the 14-day exposure period. However, degradation of ^{14}C-diquat was extensive when no sorbents were present; over half of the applied radiation was lost in 2 days and 75% was lost in 4 days. This experiment was somewhat flawed in design since no attempt was made to determine the composition of the radioactivity in the adsorbents and the wavelengths of light used are dissimilar from natural sunlight. However, it does show that adsorption of diquat to soil or clay slows photolyisis.

Data indicates that since the hydrolysis half-life and vapor half-life are very long, other mechanisms may be more important in the degradation and dissipation of diquat from the aquatic environment. While photolysis may be important for the degradation of diquat in shallow, non-turbid waters with low levels of weed growth, it is likely that microbial degradation, adsorption into target plants, and adsorption onto sediment and seston (suspended matter) as well as dilution from untreated waters are the major modes of dissipation in the field.

The principal degradate is TOPPS (1,2,3,4-tetrahydro-1-oxo-pyridol [1,2-a]-5 pyrazinium salt), which is discussed below. The EPA (EPA RED, 1995) reported that 12% of the recovered radiation was due to TOPPS after 32 days of photolytic degradation at pH 7.0. Smith and Grove (1969) also reported TOPPS to be a major product of aqueous photolysis.

3.3.2 Degradation Products

EPA (EPA RED, 1995) and Smith and Grove (1969) found that the major product produced during photolysis of diquat was TOPPS. Approximately 12% of the recovered radiation was due to the presence of TOPPS. Although Smith and Grove (1969) did not attempt to quantify the TOPPS concentration after 5-weeks exposure to sunlight, they concluded that TOPPS was a major water-soluble degradate produced under aqueous photolysis. Other significant degradates produced by the photodecomposition of diquat were picolinamide and picolinic acid with picolinic acid being the dominant product. Other significant metabolites included water-soluble volatile fragments, which accounted for one third of the applied radiation. Other minor metabolites degraded by a minor pathway directly from diquat included pyridine compounds, which could potentially degrade to water-soluble volatile fragments.

Photolysis of diquat also occurs on the surfaces of plants treated with diquat. The rate of degradation is related to the intensity of the sunlight falling on the foliage of treated plants. This phenomenon could occur on treated crop plants and on floating and emergent weeds that may be treated with diquat. Any diquat bound to dead and dying, floating or emerged plant debris may be exposed to sunlight leading to photo decomposition before it is incorporated into soil or sediment. Any diquat reaching soil or sediment, either directly or released from incorporated organic materials (decaying plant material), is subject to adsorption and protection from photodegradation. In natural systems, photochemical reactions may compete with adsorption and microbial decomposition and are likely to be comparatively slow. The avenues of loss of diquat in soils and sediment seem to be through photochemical and microbial degradation as well as desorptive processes. Since sorptive capacity and sorption itself tends to slow the rate of these processes, it is likely that diquat will accumulate in soils and sediments, particularly in those containing montmorillonite clay.

Table 3.3: Photolysis of Diquat (Laboratory Studies)

Matrix	Compound	Initial Conc	pH	Temp (°C)	Half-life (DT_{50})	Reference

Sterile buffered water (Artificial sunlight)	Diquat dibromide	NR[1]	7	Probably around ~25°C	74 days	EPA RED, 1995
Sterile buffered water Natural sunlight (May to June in Saskatchewan)	Diquat dibromide	1000 ppm	N/A[2]	Probably around ~25°C	1.6 days	Smith and Grove, 1969
Loam Soil Artificial sunlight (equivalent to 32 days of natural sunlight)	diquat dibromide	NR[1]	NR	20.5 to 29.1°C	n.s.d.[3] No degradation in 32 days	EPA RED, 1995
Neet diquat	Diquat	Dry powdered diquat	NA	28°C	~2 days	Funderburk and Bozarth, 1967

Note: Unless otherwise stated, all experiments utilized artificial light, usually mercury vapor lamps on an approximately 12 hours light/12 hours dark cycle.
NR = not reported.
NA = not available.
n.s.d. = no significant degradation.

3.4 DEGRADATION AND PERSISTENCE - SOIL

Summary: *When soil is contacted by diquat, diquat is almost immediately adsorbed. After adsorption, diquat is not readily photolyzed or metabolized by bacteria. Some species of soil microbes are known to metabolize diquat. These species include the bacteria like* Pseudomonas fluorescens*,* Rhizobium *spp.,* Aerobacter aerogenes *and the yeast* Lipomyces starkyi. *Unidentified mixed species of soil microorganism in a nutrient solution were shown to degrade diquat to carbon dioxide. The adsorptive media had a strong influence on the rate of adsorption. Diquat adsorbed to kaolinite was completely available for microbial decomposition. However, diquat adsorbed to montmorillonite clay was only available for degradation if the concentration of diquat was great enough to overcome the CEC sorptive capacity of the montmorillonite clay. Expanding lattice clays with greater numbers of inter-lamellar sorptive sites adsorb and reduce the biological availability, degradative rate and tendency to desorb in the presence of high salt concentrations. For example, diquat adsorbs to montmorillonite > smectite > bentonite > vermiculite > kaolinite. In theory, diquat should be more biologically available when sorbed on the organic matter in soil than when sorbed on the mineral component of soil. However, diquat seems to be strongly adsorbed to peat, muck and organic soils as well as organo-clay complexes and humic substances as well as higher plants, lignin and cellulose. Although these tendencies have been noted, laboratory studies indicate that diquat did not degrade in an aerobic sandy loam incubated at 25°C in the dark for 9 months. Furthermore, in the field diquat did not degrade for 3 years after application to two plots in New York at rates of 0.25 to 0.5 lbs a.i./acre. Concentrations in the top 15 cm of soil varied from 0.01 in loam soil with a potato overcrop to 0.32 ppm in clay loam soil without an overcrop. No data were found pertaining to half-life in terrestrial soil. Half-lives of diquat dibromide in soil could not be measured since no significant degradation of diquat was seen in laboratory aerobic soil studies for 9 months or long-term terrestrial field studies for 3 years.*

No significant metabolites of diquat were seen on soils treated with diquat. However, if diquat is converted to TOPPS and then 2-picolinamide by the action of sunlight on treated plant surfaces, at least one species of bacteria has been shown to degrade diquat to picolinic acid. This bacterium also has the biochemical capacity to convert picolinic acid to 6-hydroxypicolinate and eventually to 2,5-dihdroxy- pyridine, maleamate, maleate, fumarate and carbon dioxide. This kind of extensive degradation has only been seen when sunlight and a short gram-negative rod-shaped bacterium jointly degrade diquat. So, it is not entirely clear if this extensive degradation can occur on soil under normal conditions. However, a number of soil bacteria are known to metabolize diquat in pure culture. The metabolites produced by these soil bacteria have not been identified

Although only the aquatic uses of diquat are considered in this document, the compound is registered for terrestrial applications, which account for the largest use. Data regarding diquat persistence in soil are therefore required to be submitted to the EPA. This information has a relevance to accidental terrestrial overspray on lake or stream shorelines, and peripherally as an indication of possible fate on near-shore lake bottoms exposed by drought or draw-down following a diquat application.

3.4.1 Half-life

No soil degradation data regarding diquat dibromide half-life were found during this review (EPA RED, 1995). Broadly summarizing the data, persistence of diquat dibromide indicates that the half-life of this material is very long on soils (Table 3.2.4).

In the laboratory, diquat dibromide is not readily degraded on sandy loam soil under aerobic conditions. Diquat is not degraded after application at 3 ppm for 9 months at 25°C (EPA RED, 1995). Diquat sorbed on kaolinite clay incubated in aqueous solutions with added nutrient media was degraded extensively to carbon dioxide. The rate of degradation in this system was as high or slightly higher than in systems with no added kaolinite clay. However, diquat adsorbed to montmorillonite clay and incubated in similar aqueous solutions only degraded to the extent that the concentration of diquat exceeded the cation exchange capacity's (CEC's) strong adsorption capacity (SAC) of the montmorillonite clay (Weber and Coble, 1968).

In a field study, diquat applied at 0.25 to 0.5 lb a.i./acre did not degrade for three years after application to two plots in New York. The concentration of diquat in the upper 15-cm soil depth was 0.32 ppm in clay loam soils with no overcrop at the time of application, and as low as 0.01 ppm in loam soil with a cover crop of potatoes at the time of application. Concentrations at the New York site were 0.01 to 0.03 ppm at 15 to 22.5-cm soil depths. Similar applications to loam soil at two sites in Idaho did not degrade for three years after application. Furthermore, the concentrations of diquat ion ranged from 0.01 to 0.13 ppm in the upper 35-cm soil depth (EPA RED, 1995). No residues were recovered from below 35-cm soil depth.

Since no degradation of diquat was seen at long-term terrestrial sites, or in laboratory experiments with soil, it was not possible to determine the half-life of diquat on soils.

3.4.2 Degradation Products

A few soil microorganism have been noted, which are capable of degrading diquat. For example, the bacteria *Pseudomonas fluorescens*, *Rhizobium* sp. and *Aerobacter aerogenes*, and the yeast *Lipomyces starkeyi* are capable of degrading diquat in culture medium. However, the products produced by this degradation were not isolated or identified (Simsiman et al ,1976 and Funderburk, 1969).

It has been noted that mixed soil microorganisms from Norfolk sandy loam grown in nutrient media are capable of degrading diquat to carbon dioxide. However, intermediate products were not isolated or identified. This degradation was largely unaffected when diquat was adsorbed to kaolinite clay but was largely eliminated when diquat was adsorbed to montmorillonite clay. The only case where these microbes degraded diquat adsorbed to montmorillonite clay occurred when the diquat concentration was high enough to more than saturate the cation exchange capacity seen in the montmorillonite clay. This adsorption onto montmorillonite clay is often referred to as strong adsorption capacity (SAC), and is due to the combined effects of coulombic and van der Waals forces. However, the binding of diquat to kaolinite clay, vermiculite clay and organic materials is due primarily to coloumbic forces which are much weaker (Simsiman et al, 1976).

There has been one report of a documented complete metabolic pathway for diquat. It involves the photolysis of diquat to TOPPS followed by further photolysis to picolinamide. From this point, a short, gram-negative rod-shaped bacterium is capable of degrading picolinamide to picolinate and then subsequently to 6-hydroxypicolinate, 2, 5-dihydroxypyridine, and then to maleamate, maleate and fumarate. Since maleate and fumarate can enter the Krebb's cycle, the end products of this pathway should be carbon dioxide. It is not clear if picolinamide can be generated from sources other than photolysis of diquat. If it can be, this may be a primary mode of degradation in soil solutions (soil/water slurries), but probably not in soil itself. This may be important since it is known that diquat disappears slowly, although it seems relatively resistant to biological degradation. This may be an important terrestrial pathway where diquat is sprayed on plants and then degraded to picolinamide prior to the plants dying and being incorporated into the soil (Orpin et al, 1972).

3.4.3 Physical and Chemical Factors

There are several physical and chemical factors influencing the rate of binding for diquat dibromide and its subsequent breakdown in soil. Among those investigated are pH, temperature, the presence of other soil constituents and soil moisture, soil microbial population, and prior treatment with diquat. The last of these is discussed in more detail in Section 3.2.6.

- **Temperature and pH**

  The rate of chemical reactions and most biological metabolic processes doubles for every 10°C increase in temperature. However, since the rate of degradation on soil can not be determined for diquat, this is a mute point. For purely physical aspects, temperature has been found not to significantly influence the binding of diquat to soil in most cases. Although pH is not believed to strongly affect the adsorption of diquat to soils, it has been noted that the related herbicide (paraquat) binds less readily to soil that has recently been limed. This probably has more to do with the presence of Ca^{++}, which competes for cation exchange sites than the change in soil pH due to liming. Furthermore, paraquat treated soils that had been heavily limed (20 tons/acre) were seen to be more phytotoxic than lightly limed soils (1.5 tons/acre) (Weber and Best, 1972 in Weber and Weed, 1974). It is interesting to note that even under conditions of heavy liming, microbial decomposition of paraquat proceeds very slowly and probably did not contribute significantly to the observed effects.

  As verification to the above observations, it has been observed that paraquat was very toxic to blueberry plants in loamy sand soils containing very little clay mineral matter. Subsequent bioassays of the same soils with cucumber seedlings shows that paraquat was very active and persistent in soil long after the blueberry plants had been removed (Monaco and Webber, 1972 in Weber and Weed, 1974). However, the paraquat treatment rate for these experiments was very high (50 lbs/acre) while typical treatment rates with paraquat are approximately 2 lbs/acre.

- **Soil Moisture**

  Degradation on soils with normal levels of moisture have been noted to be insignificant (EPA RED, 1995). However, clay/water slurries have been noted to degrade ^{14}C-diquat when mixed populations of soil microbes from Norfolk sandy loam and incubate in 50 mL of nutrient solution. Degradation was noted by the production of $^{14}CO_2$. If a sorbent is added to this mixture, it may eliminate metabolism of diquat if the diquat is adsorbed by the presence of montmorillonite clay or other clay which strongly and irreversibly adsorbs diquat. If a sorbent like kaolinite clay is added, which does not strongly adsorb diquat, the degradation of diquat appears to be unaffected (Weber and Coble, 1968).

- **Organic Matter and Clay Minerals**

  Clay minerals, particularly expanding lattice clays like montmorillonite, smectitie and bentonite, which have the bulk of their CEC sorptive sites located in inter-lamellar regions generally have a high adsorptive capacity. Diquat or paraquat sorbed to such minerals will not be biologically available to plants or microorganisms (Weber and Weed, 1974). Limited expanding lattice clays and non-expanding lattice clays like vermiculite, kaolinite and illite, and organic soils like loam, sand and muck also adsorb diquat. However, this binding is much looser and is readily desorbed with high salt solutions; also diquat and paraquat can often be considered to be biologically available for uptake by plants and microorganisms on soils where only loose binding of these pesticides takes place (Weber and Weed, 1974). However, small amounts of expanding lattice clays will adsorb diquat and paraquat and make them biologically unavailable to plants and microbes.

- **Soil Microbes**

  Since diquat has not been noted to degrade in soils (EPA, 1995), characteristics of initial lag times, and recruitment by selection of bacterial populations effective in degrading diquat is not an issue that needs to be addressed. There is probably little that can be done to improve the ability of soil microbes to degrade diquat since the ability to degrade any pesticide is genetically controlled within the bacteria. Mutations would be necessary in order for a bacterial species to utilize the pesticide as a new food source. If this occurs, bacterial populations can increase and the breakdown of the herbicide may commence. Often repeat treatments in subsequent years would activate the mutant bacteria and breakdown would occur immediately. The consequence of this more rapid breakdown may be that greater use rates may be necessary for the control of the same weeds (Newbold, 1975). However, as previously discussed, bacteria do not appear to be able to degrade diquat once it has been bound to soil.

  Several species of soil microorganisms have been seen to degrade diquat in pure culture including the bacteria *Pseudomonas fluorescens*, *Rhizobium* sp. and *Aerobacter aerogenes*. As discussed previously, mixed populations of microorganisms have been seen to degrade unbound diquat to carbon dioxide in nutrient solutions (Weber and Coble, 1968). However, if the diquat is bound to a sorbent like montmorillonite clay, these microbes are ineffective in degrading diquat. Also a short, gram-negative rod-shaped bacterium is capable of degrading the diquat photolysis product (picolinamide) to picolinic acid and other degradates in nutrient solutions (Oprin et al, 1972). This bacteria is capable of growing on picolinamide and is probably capable of degrading picolinamide to carbon dioxide. In addition to these species, the yeast *Lipomyces starkeyi* is capable of degrading both diquat and paraquat (Baldwin et al, 1966 in Wright, 1971 and Funderburk, 1969). When soil is fractionated into an organic and a mineral component, *Lipomyces stakeyi* is able to degrade paraquat on the organic component but not on the mineral component (Burns and Audis, 1970 in Simsiman et al, 1976). However, work showing that soil microorganisms are capable of degrading diquat on soil that is not in an aqueous solution is not available.

  The available data indicates that after diquat has adsorbed to soil it is predominately biologically unavailable. Generally speaking, diquat that has been bound to soil is biologically unavailable for the control of weeds or for breakdown by microorganisms (Weber and Scott, 1966; Weber and Coble, 1968 and Weber et al, 1969). For example, Coats et.al. (1966) found that diquat treated soils were effective in inhibiting the growth of wheat seedlings depending on the soil type and how tightly bound the diquat was to that soil. On sandy loam, concentrations of diquat or paraquat as high as 100 ppm c.e. had no effect on the growth of wheat seedlings; on kaolinite clay, concentrations of diquat or paraquat as high as 1,000 ppm c.e. inhibited the growth of wheat seedlings by approximately 40% and 50%, respectively; on bentonite (a form of montmorillonite), diquat and paraquat at concentrations as high as 50,000 ppm c.e. had no effect on the growth of wheat seedlings. These concentrations are highly in excess of the concentrations that would typically be used to treat for terrestrial weeds (0.25 to 0.89 lbs a.i./ acre = 0.19 to 0.66 ppm a.i. = 0.10 to 0.35 ppm c.e. in the top 15 cm of soil). Therefore, wheat would probably not be affected if grown in soil that had previously been treated with diquat. In the case of diquat, adsorption will bind the chemical so tightly that microorganisms cannot use it and non-target plant species will not typically be able to adsorb it when they are planted in plots previously treated with diquat. It has been estimated by Knight and Tolimson (1967 in Simsiman et al, 1976) that over 10,000 kg/ha of diquat would be required to exceed the typical strong adsorbance capacity of a typical sandy clay loam agricultural soil. For a similar product (paraquat), it was determined that 2,250 pounds/acre would be necessary to saturate the cation exchange capacity of a 6-inch soil layer containing 1 percent montmorillonite clay; and this would assume no degradation of the herbicide by microorganisms. Therefore, diquat and paraquat adsorbed to typical agricultural soils would not have significant impact on crops planted in these soils. This data was confirmed by the effects of paraquat in inhibiting the growth of cucumber seedlings when grown in hydroponic solutions containing montmorillonite or kaolinite clay. While kaolinite clay reduced growth inhibition to 55% when it was present in high enough quantities (600 mg) to completely bind 10 μmoles of paraquat, montmorillonite clay at 30 mg reduced growth inhibition in cucumber seedlings treated with 10 μmoles paraquat to zero (Weber and Scott, 1966).

Table 3.4: Diquat Persistence in Soil

Experiment	Compound & Application Rate	Half-life (DT_{50})	Time to Residues < 0.05 ppm	Reference
Lab aerobic sandy loam soil microcosm*	^{14}C-Diquat dibromide 3 ppm	> 9 months at ~25°C	NR[1] but presumably more than 9 month	EPA RED, 1995
Long-term Field Dissipation Clay loam soil with no crop cover New York*	Diquat dibromide 2 x 0.25 lbs c.e.[2]/acre 0.32 ppm c.e. in 0 to 15 cm soil depth	>3 years	NR but presumably more than three years	EPA RED, 1995
Long-term Field Dissipation Loam soil with cover crop of potatoes in New York*	Diquat dibromide 2 x0.25 lbs c.e./acre 0.01 ppm c.e. in 0 to 15 cm soil depth	>3 years	Concentration immediately after spraying foliage was <0.01 ppm c.e.	EPA RED, 1995
Long-term Field Dissipation Loam soil with no cover crop in Idaho*	Diquat dibromide 2 x 0.25 lbs c.e./acre 0.13 ppm c.e. in 0 to 35 cm soil depth	> 3 years	NR but presumably more than three years	EPA RED, 1995
Long-term Field Dissipation Loam soil with cover crop of potatoes in Idaho*	Diquat dibromide 2 x0.25 lbs c.e./acre 0.01 ppm c.e. in 0 to 35 cm soil depth	>3 years	Concentration immediately after spraying foliage was <0.01 ppm c.e.	EPA RED, 1995

[1] NR = not reported
[2] c.e. = Diquat cation equivalent
* EPA guideline study

3.5 DEGRADATION AND PERSISTENCE - AQUATIC SYSTEMS

Summary: Microorganisms associated with the surfaces of plants are the primary cause of diquat degradation. After treatment of simulated ponds containing water, sediment and aquatic plants (Eurasian watermilfoil and American waterweed), incubation of a system treated with 1.5 ppm diquat c.e. produced 48% volatile products (probably to CO_2*), 32% water soluble degradation products with the remaining 19% of the applied test substance remaining as diquat bound to the sediment. Diquat dibromide is very stable in sediment and once bound to the sediment, diquat is stable for 180 days or more. Two laboratory studies in sterile buffered water found no significant degradation after 30 days of incubation. In a 31-day aerobic aquatic study, a half-life could not be calculated since 95% to 99 % of the diquat residues were associated with the soil-sediment and no significant amounts of diquat were degraded. In a 9-month anaerobic aquatic study, a half-life for diquat could not be determined since 89% to 100 % of the diquat residues were associated with the soil-sediment; but approximately 5% of the applied radioactivity had been degraded to unknown metabolites after 9 months of incubation at 25 ℃. According to the analysis done in a review article by Reinert and Rogers (1987), other aerobic and anaerobic laboratory aquatic studies in weed-free systems indicate half-lives of 31.9 and 49.5 days, respectively. However, the original report by Simsiman and Chesters (1976) found no evidence for degradation of diquat within these weed-free systems. This indicates that the bulk of the diquat (>90%) remains bound to the sediment for more than 180 days. As would be expected in a test system that contained soil with 39% clay and a CEC of 0.24to 0.34 me/g soil. However, the total residue concentration in water for this system dissipated quickly with a half-life of less than one day. It has been estimated that the diquat half-life within a typical aquatic ecosystem would be over 1,000 days due to the tight and essentially irreversible binding of diquat to the hydrosoil (sediment). Although diquat is known to degrade slowly, it has been determined in laboratory studies that lag time is seen before species of microbes are recruited that will effectively degrade diquat. It has also been determined that subsequent applications of diquat are degraded more rapidly than the initial application in ponds containing sand sediment and sandy clay sediment. Although microorganisms have been identified that degrade diquat in nutrient medium, the metabolites produced have not been identified, although certain species have been identified including soil bacteria like* Pseudomonas fluorescens*,* Rhizobium *sp. and* Aerobacter aerogenes *and the yeast* Lipomyces starkeyi. *Other species of bacteria and microbes that degrade diquat are known to exist but they have not been isolated or identified. It is unclear if these microbe species typically found in soils can also be found in aquatic ecosystems. However, since* Pseudomonas fluorescens *can tolerate up to 2,500 ppm of diquat, it may be a likely candidate as one of the more important organisms involved with the potential degradation of diquat in the aquatic environment.*

Most outdoor studies reported that diquat is removed rapidly from the water column with a half-life ranging from <1.0 day to ~ 4.0 days. When diquat is used at typical use rates (0.5 to 1.0 ppm c.e.), dissipation half-lives in water can vary from <1 day to ~ 4 days. At sites where diquat dissipation from water is rapid, a combination of factors is involved, which may contribute to the rapid dissipation of diquat. These factors may include sediment with high amounts of montmorillonite or bentonite clay, seston (suspended matter) containing suspended sediment with a high proportion of clays in them, the presence of phytoplankton which may adsorb diquat to high levels and the presence of aquatic macrophytes that may also adsorb diquat. Therefore, the persistence in water at concentrations higher than the limit of detection ranges from < 1 day to ~ 35 days.

The concentration of diquat in sediment usually starts out low (<0.5 ppm a.e.) but can build up over time as treated aquatic weeds decay and release diquat back into the water column or additional treatments occur. For example, treatment of ponds at a water column concentration of

0.62 ppm c.e. can result in initial (Day 1) sediment concentrations of diquat that are less than 0.5 ppm c.e.. Concentrations of diquat only build up slowly reaching 7.4 ppm c.e at 18 days. However, after the aquatic weeds die and release diquat back into the water between the 24th and 56th day, concentrations can rise to as high as 37 ppm c.e. Furthermore, dissipation from the sediment can be slow with 24 ppm c.e. remaining in the sediment 160 days after application. The half-life of diquat on these treated sediments has been estimated to be greater than 160 days at many of the treatment sites. Therefore, the persistence of diquat in sediment can be longer than 4 years with sediment concentrations remaining higher than 1.7 ppm c.e after treatment with 0.27 lbs/acre.

Diquat is dissipated rapidly from the water column and is retained on sediment for very long periods of time. In an experiment designed to mimic the worst case in the field, two ponds in Florida were treated four times with 4 lbs c.e./ surface treatment acre with diquat. Diquat was removed rapidly from the water column by adsorbing to the sediment. Diquat concentrations in water typically dropped from ~0.1 ppm c.e. on the day of application to < 0.004 to 0.027 ppm after five days of dissipation, < 0.004 to 0.021 ppm c.e. by day 10, and <0.004 ppm by day 29. The pattern of dissipation was similar after each treatment. However, the dissipation rate after the first treatment was about half as fast as after subsequent treatments. These water dissipation half-lives range from 0.72 to 2.3 days. On the sediments (sandy clay loam and sand sediments), the concentration of diquat was variable and ranged to a maximum of 1.2 ppm c.e. diquat 30 days after the last application. There was no discernable pattern of diquat concentrations in sediment even after 180-days of dissipation when the concentrations ranged from<0.01 ppm to 0.44 ppm c.e. The findings at these two sites were in agreement with findings from other field dissipation studies conducted under a variety of environmental conditions; and were also comparable to data obtained from laboratory studies.

Laboratory studies show that diquat significantly decreases in aquatic systems that contain water, sediment and aquatic weeds. For example, a microcosm made up of Lake Mendota water, and sediment augmented with a heavy growth of Myriophyllum spicatum *and* Elodea canadensis *broke down ^{14}C-diquat in 22 days to volatile components (48%) and water soluble degradates (32%). The bulk of the remaining diquat was associated with the sediment (19%) with only small amounts remaining in the water (1%). After 180 days of dissipation, 77% of the applied compound consisted of volatile components, 10% consisted of water-soluble degradation products and 13 % consisted of diquat bound to sediment. This phenomenon was observed after treating the microcosm with 1.5 ppm c.e. ^{14}C-diquat dibromide. It is not clear to what degree diquat may be degraded in the field. However, it is apparent the diquat degradation occurs when it is associated with microorganisms found on decaying and dying vegetation. After the plants have decayed, the remainder of the diquat will strongly adsorb to the sediment. Many soil/sediment types have been tested for their adsorption capacity of diquat and it is apparent that typical sediments adsorb and tightly bind to diquat up to concentrations of 250 to 2,500 ppm. Sediments that contained clay minerals with a high cation exchange capacity sorbed diquat to a higher level. For these reasons the overall field half-life of diquat in aquatic ecosystems has been estimated to be longer than 1,000 days.*

Numerous physical and chemical factors can affect the persistence and fate of diquat in the aquatic environment. Temperature influences the rate of both chemical and biological processes. Since diquat is not readily hydrolyzed at typical environmental temperatures, it appears unlikely that temperature will influence the degradation of diquat by hydrolysis, and generally the adsorption to montmorillonite and kaolinite clays is unaffected by temperatures between 10°C and 55°C, which is typical of sorbents that depend on coulombic and van der Waals forces for adsorption. However, adsorption to vermiculite was somewhat retarded at lower temperatures *since adsorption to vermiculite appears to be endothermic. Most importantly, the degradation of diquat by microflora will be inhibited at low water temperatures. Water pH does not appear to play a significant role in the hydrolysis of diquat. Except at extremely high pH (>9) (Simsiman et al, 1976), the hydrolysis of diquat appears to be unaffected by pH. The amount of oxygen dissolved in a water body has a direct effect on the speed of diquat adsorption and on metabolism. On soils containing significant quantities of montmorillonite clay, for anaerobic soil/water systems it took 7 days to reach adsorption equilibrium while it took 21 days for aerobic soil/water systems to reach adsorption equilibrium. However, the final amounts of diquat adsorbed in such systems appeared to be similar (typically more than 90%). Although extensive degradation of diquat generally does not occur after adsorption to sediment, degradation was much faster in aerobic conditions with a half-life of 31.9 days compared with that in anaerobic conditions (49.5 days). Although no work was done to test the hypothesis that temperature will affect the rate of microbial degradation, it seems reasonable to expect that within a very broad range of normal environmental temperatures (~5 ° C to ~45 ℃) the rate of microbial degradation will increase by two-fold for every 10 ℃ increase in temperature. This temperature effect assumes that the microbial species degrading diquat remain viable throughout this temperature range. At temperature extremes (<5 ° C and >45 ° C) it can be expected that microbial degradation will slow up and eventually cease. In many cases, eutrophic and even mesotrophic lakes are more likely to support large populations of microorganisms that can metabolize unadsorbed diquat more rapidly than lakes with lower nutrient levels. On the other hand, if carbon sources are not abundant, competition for the carbon in diquat may favor the growth of the microbiota that can utilize diquat exclusively. It is generally believed that the adsorption of diquat onto sediments makes it biologically unavailable; and that once adsorption occurs, microbes are unable to degrade diquat and algae, plants and animal biota will not adsorb it. This phenomenon appears to be dependent upon the type of sediment that is present. However, sediments with even very low levels of montmorillonite clay appear to irreversibly adsorb diquat typically to concentrations of more than 250 ppm c.e.*

Probably the most important physical process affecting diquat persistence in larger water bodies is transport of treated water away from the treated area and replacement with untreated water through lateral circulation or vertical movement of water. In this regard, the larger the lake, the more wind blowing across the lake surface, and the more water exchange through inlet and outlet streams or rivers, the more likely that diquat residues will be rapidly dispersed and diluted to below detection limits. In small lakes, detectable concentrations of diquat may be carried a significant distance down an outlet stream if the flow is sufficient and degradation is slow.

The disappearance of diquat from a lake or other natural water body is influenced by a number of factors as discussed in earlier in Section 3.1.4.3. Various water chemistry conditions, physical conditions such as temperature, adsorption to the sediment, and the extent of water currents and dilution can all have very pronounced effects on diquat persistence. This section reviews the disappearance times reported for natural water bodies and for artificial laboratory microcosm and mesocosm studies (small and medium scale simulations) and also looks at the reported factors that can influence such times.

3.5.1 Half-life and Disappearance Time

Table 3.2.5 summarizes the half-lives of diquat as reported in research papers, as well as the time to non-detection or very low levels as specified in the table. A half-life is the time required for an herbicide to reach half of its initial concentration immediately following application. Depending on the type of study and the data collected, a half-life may be mathematically calculated using several analyses over time, or may be interpolated from tabular data or figures given in a cited paper as was sometimes necessary in this review.

Time to disappearance is the time necessary for an herbicide's concentration to drop below the lower limit of analytical detection. This value is usually ~0.01 ppm c.e. for diquat in sediment and ~0.004 to ~0.008 ppm c.e. in water (Fujie, 1988). Because of the variety of analytical techniques used over time (chemical analysis, bioassay), the Limit of Detection (LOD), the lowest herbicide concentration that can be reliably quantified, has varied over time.

Half-life values are important for estimating persistence, but can be misleading if the herbicide remains in the environment at significant concentrations after the half-life time. Times to disappearance are useful tools for predicting impacts on biota and wildlife, particularly when used with calculated or estimated half-lives (Waterborne, 1995 and Ritter et al, 2000). The persistence of diquat varies widely depending on the conditions of the system being tested. Therefore, it is not surprising that a wide range of half-life and disappearance times has been reported in the literature.

The majority of scientific literature describes laboratory microcosm/flask and aquarium studies as opposed to natural water body studies. Laboratory studies allow more control over water chemistry and temperature as well as the determination of degradates formed if desired. They are useful for isolating the effect of specific factors on diquat persistence. On the other hand, studies in natural lakes and ponds subject the herbicide to 1) temperature and pH variations, 2) a greater variety of microorganisms, 3) a greater water: sediment surface ratio (thus affecting adsorption), and 4) to dilution and movement throughout the lake. There are few reported data concerning diquat persistence in sediment; most of the water/sediment studies report half-lives in the water phase and the sediment phase separately. However, Reinert and Rogers (1987) have reported the half-life for the two phases as a whole from work conducted by Simsiman and Chesters (1976) in weed-free flask systems. The reported half-life values for these systems were 31.9 days under aerobic conditions and 49.5 days under anaerobic conditions. However, in the original paper these weed-free flask systems do not appear to degrade significant portions of the applied diquat with more than 90% irreversibly binding to the sediment.

Two EPA guideline aquatic metabolism studies were conducted at 25°C using ^{14}C-labeled diquat dibromide. In the 31-day aerobic study (EPA RED, 1995), between 95% and 99% of the diquat residues were associated with the soil/sediment portion of the system and no significant metabolites were produced. The aerobic aquatic half-life could not be calculated. In the 9-month anaerobic study (EPA RED, 1995), one unidentified degrade comprised approximately 5% of the applied radioactivity. Throughout the anaerobic study, 89% to 100% of the diquat residues were associated with the solid sediment portion of the system. No anaerobic aquatic half-life could be calculated. Although the characterization of the sediments were not reported in these studies, it is likely that the soil had enough montmorillonite or bentonite clay to completely adsorb the diquat applied to the test system. Weber and Coble (1968) have reported that monmorillonite clay, which has adsorbed diquat, protects it from microbial degradation. It is apparent from these studies that, in the absence of aquatic weeds, diquat has a very long half-life. The half-life for the aerobic aquatic metabolism study is >31 days and the half-life from the anaerobic study is much longer than 9 months.

Two Wisconsin sediments, containing 39% to 58% clay with monmorillonite clays tending to predominate, were mixed with water at ratios of 5mL:200 mL (sediment:water) or 5mL: 20mL (sediment:water). These systems were maintained under either anaerobic or aerobic conditions (Simsiman and Chesters, 1976). Under these conditions, the sediment containing 39% clay came to adsorption/equilibrium in 7 days under anaerobic conditions and in 21 days under aerobic conditions. However, the soil containing 58% clay came to adsorption equilibrium at about 14 days. The soil/sediment typically adsorbed more than 95% of the diquat and once adsorbed, it appeared to remain unchanged for longer than 65 to 180 days. The slight amount of degradation (0.02 to 3.57%) appeared to be faster under aerobic conditions than anaerobic conditions and appeared to be faster under conditions where the sediment:water ratio was 5 ml: 20mL. The faster degradation rate under aerobic conditions was expected. However, it was not expected that the system containing less water would exhibit a higher level of degradation; this observation was explained by noting that more intimate contact was achieved between the overlying water and the sediment in systems where the sediment to water ratio was narrower. An alternative hypothesis that seems more credible is that there was a greater amount of suspended sediment in the 5:200 (sediment to water systems) which removed more diquat and made it unavailable for degradation by aquatic microbes. Therefore, 5:20 sediment to water system degraded diquat more readily. Although Reinert and Rogers (1987) reported diquat half-lives of 31.9 days for the aerobic aquatic system and 49.5 days for the anaerobic aquatic system, it is not clear how this calculation was made and it is believed to be much longer than this (more than 180 days for the aerobic aquatic system and more than 95 days for the anaerobic aquatic system). However, the dissipation half-life of diquat in the overlying water was very short and is estimated to be less than 2 days in every test.

However, a mesocosm containing sediment (8 L), water (60L) and a heavy growth of Eurasian watermilfoil and American waterweed showed extensive biodegradation in 22 and 180 days. After applying 1.5 ppm c.e. ^{14}C-diquat dibromide, 48% of the applied radiation consisted of volatilized components (probably $^{14}CO_2$), and 32 % water soluble degradation products. The rest of the residues remained as unchanged diquat with 1% in the water and 19% in the sediment. After 180 days of degradation, the amount of volatilized $^{14}CO_2$ was 77% of the applied radiolabel and the amount of water soluble residue was 10%. The remainder of the applied radiolabel (13%) was recovered from the sediment as unchanged diquat. In the presence of aquatic weeds degradation of diquat can be extensive. It is believed however, that this degradation of diquat is due to the presence of aquatic microbes that are associated with the dead and dying aquatic weeds; it is not believed that the aquatic weeds are responsible for the degradation of diquat. It is interesting that the bacterial degradation appears to lag for approximately four days before it becomes a major factor in the degradation of diquat. Prior to this time, adsorption appears to be the main factor in removing diquat from the water column of this artificial aquatic system (Simsiman and Chesters, 1976). A lag-phase prior to the microbial degradation of a herbicide appears to be common in other systems as well and was observed in mesocosms treated with endothall and 2,4-D. [See Sections 3 and 4 of the Endothall and 2,4-D Supplemental Environmental Impact Statements (Ecology, 2000)].

Hiltibran (1972) found that sediment-free systems consisting of tap water, aged tap water, lake water or fresh tap water with fish degraded dissolved diquat at 1.0 ppm c.e. with a half-life of about 7 days. Complete dissipation of diquat in these systems occurred in 28 to 65 days when the concentration of diquat dropped to levels of 0.01 to 0.19 ppm c.e. Such concentrations can be still higher than the drinking water tolerance of 0.02 ppm c.e. However, the half-life and persistence of diquat is usually much shorter in the field than the laboratory. The half-life and persistence time in the laboratory appears to increase for diquat as the initial concentration increases to 5.0 ppm c.e. At 5 ppm c.e., the half-life of diquat in a similar experiment increased to 14 to 65 days with persistence at concentrations of 0.25 to 2.71 ppm c.e. being greater than 65 days.

In a laboratory aquatic system containing demineralized water with no sediment, a treatment with 1.44 ppm c.e. diquat did not significantly dissipate in 6 days. However, in a model ecosystem containing 11.4 gallons of water from Lake Chautauqua (New York), 1 gallon of bottom sediment, plants, and animals, the water dissipation half-life of a 1.5-ppm c.e. diquat treatment was 21.9 hours. Dissipation to concentrations of less than the drinking water standard (0.02 ppm) occurred between 96.5 and 120 hours. After 195.5 hours, the concentration of diquat in water was virtually non-detectable (<0.001 ppm c.e.) and the sediment contained 51.475 ppm c.e. By this time the concentration of diquat in these sediments had risen to 51.475 ppm c.e. and the diquat concentration in plants had risen to 0.04 to >0.15 ppm c.e. This was expected because the plants would have decayed extensively at this point and released their sorbed diquat, which would typically be immediately adsorbed by the sediment (Shaw and Hopke, 1975).

Several authors have conducted field studies with diquat and determined the levels of diquat in water and sediment at several different times after treatment. For example, Fujie (1988), in a study designed to show the worst case dissipation of diquat, treated two ponds with a four foot depth in Florida with diquat dibromide at 4 lbs c.e./acre (0.37 ppm c.e.). Fujie treated half of each pond on four different dates (6/9/87, 7/9/87, 8/8/87 and 9/9/87) with the maximum use rate. The sediment in these ponds was characterized as sandy clay loam or sand sediment and the sorptive capacity of these sediments appeared to be fairly low. Samples of water were taken at 3 separate depths and in 4 separate areas of the pond, but by the end of the first day, concentrations throughout the ponds were similar. The dissipation rate in water was two-fold higher in the 2nd , 3rd and 4th treatment than in the 1st treatment. The water dissipation half-life for these ponds ranged from 0.72 to 1.6 days at a pond located in Bivens Arm Florida and 0.76 to 2.3 days at the pond located at Shongaloo Fisheries. Within 3 days the concentration of diquat in the water columns of these ponds fell to 0.005 to 0.047 ppm c.e. and by 7 days the concentration of diquat fell to less than the US EPA drinking water standard of 0.02 ppm c.e. By ten days after treatment, the water concentrations of diquat in these ponds fell to levels that were typically less than the level of detection (<0.004 ppm c.e).; but occasional detects as high as 0.021 ppm c.e. were seen. Concentrations in the sediment of these ponds remained fairly low throughout the study. These concentrations were similar after each treatment with concentrations ranging around 0.3 to 0.6 ppm c.e. in the 0 to 5 cm sediment depth; but the lowest to highest concentration of diquat in the sediment raged from <0.01 to 1.2 ppm. No consistent pattern for concentrations of diquat in the sediment could be detected and 180 days after the last treatment, the diquat concentration in sediment ranged from <0.01 to 0.44 pmm c.e. Concentrations of diquat in the sediment were also measured in the 5 to 10 cm segment of the sediment with only sporadic detects of diquat above 0.01 ppm c.e being observed.

Gilderhaus (1966) found that when diquat was applied one time at concentrations of 1.0 or 3.0 ppm c.e. the half-life in water was ~3 and ~4 days, respectively. The persistence in the water column ranged from 10 to 21 days for the 1.0 ppm c.e. application and 21 to 42 days for the 3.0 ppm c.e. application. Diquat concentrations in the sediment reach a maximum of 4.0 to 11.4 ppm c.e. 12 weeks after a single 1.0 ppm c.e application and 22.8 to 45 .8 ppm c.e. 12 weeks after a single 3.0 ppm c.e. application. These concentrations persisted in sediment for the duration of a 24-week monitoring period. Multiple applications at 1.0 ppm c.e. and 3.0 ppm also persisted for the duration of the experiment and appeared to accumulate to levels that were 3- to 7-fold higher than the systems that received only one application.

Frank and Comes (1967) saw a very short half-life for diquat (0.63 days) and persistence in the water column of between four and eight days after a treatment of a pond with 0.62 ppm diquat. The concentration of diquat in the water column was 0.01 ppm c.e. after 4 days of dissipation. The bulk of this diquat had adsorbed to the sediment and aquatic weeds. The concentration in the sediment was low (<10 ppm) for the first 18 days after treatment but accumulated to 37 ppm 56 days after treatment and did not dissipate significantly by 160 days after treatment. At that time the concentration of diquat in sediment was still above 24 ppm c.e. The authors indicated that this phenomenon was due to the fact that plants sorbed diquat and held it away from the sediment until extensive decay had occurred. After decay had occurred, diquat was released from the humic material and incorporated into the sediment from which release back into the water column was very slow.

In a similar experiment, Grzenda et al (1966) found that diquat plus paraquat at 2.5 plus 2.5 ppm c.e. displayed a diquat half-life of ~0.9 to ~3.0 days depending on conditions of the treated ponds. Also, the persistence in ponds to concentrations less than 0.01 ppm was between 7 and 14 days in the former case and between 27 and 30 days in the latter case. The difference in the persistence in these two situations appears to be correlated with turbidity and the presence of thick stands of *Nitella* spp. Turbid water, due to the suspension of finely divided organic sediment probably sorbed diquat more readily than when suspended sediment, was not present. Furthermore, *Nitella* spp. probably sorbed a significant amount of diquat from the water column. After decomposition, *Nitella* spp. released diquat, which was readily adsorbed to the sediment. However, this last hypothesis cannot be verified because sediment analysis for diquat was not possible since diquat could not be extracted from the sediments found in these South Carolina ponds with even the most rigorous means.

In Lake Seminole, Georgia, only trace concentrations of diquat could be found 24 hours after treatment for control of Eurasian watermilfoil. Diquat was not detected in any of the soil/sediment samples. However, diquat was seen at significant concentration in Eurasian watermilfoil seven days after treatment. Eurasian watermilfoil was not controlled well by the application of diquat (Daly et al, 1968).

When diquat and Cutrine® were used at rates of 1.0 and 0.46 ppm in Florida, Louisiana, and Texas to control hydrilla, approximately two-thirds of the diquat and copper dissipated from the water column within one day. It was believed that this dissipation was due to adsorption of diquat and copper by aquatic weeds, soil and detritus. Diquat dissipated to below detectable limits within 14 days of application (Ganstad, 1986 and Gangstad and Stanley, 1987). Hydrilla and other plants have been noted by other authors to resist treatment with diquat as the season progresses due to the accumulation of clay particles, epiphytic algae and aufwuchs on leaf surfaces. The accumulation of these living organisms and detritus on the leaf surfaces and in the water column prevents the adsorption of diquat by the target plant (Bowmer, 1982 and Hodson et al, 1984).

Other work by Gangstad (1986) and Stanley and Gangstad (1987) in Californian reservoirs indicates that three treatments with diquat plus copper at 0.1 ppm and 0.3 ppm copper in mid May, mid July and mid August had water dissipation half-lives of less than one day. Furthermore, the time to disappearance was typically around one day. Rapid dissipation of diquat was attributed to rapid adsorption by aquatic weeds, soil and detritus. Reservoirs with the shortest diquat half-life had more planktonic organisms than reservoirs with longer diquat half-lives.

In Nomini Creek (Virginia), diquat was not detected in the water column at any time after treatment with 4 lbs c.e./acre (0.35 ppm c.e.) (~0.4 days to 36 days). However, extensive diquat was detected in the sediment from 9 (1.21 to 3.70 ppm c.e.) to 356 days (1.19 to 4.07 ppm c.e.). The sediment concentration of diquat did not vary significantly within a single sampling site. The low levels of diquat in the water column may have been due to several factors, including high dilution from a tidal stream and tidal currents that reached speeds of 0.4 feet/sec, the presence of watermilfoil at the time of treatment (diffuse to 80% coverage), extensive suspended sediment (36 ppm) and a sediment layer consisting of 79 to 89% silt clay and ~13% organic matter. It is interesting that the lowest sediment concentrations of diquat occurred at the sampling station where the watermilfoil coverage was most extensive (Haven, 1969).

Hiltibran et al (1972), found that diquat could not be detected in the water column 24 hours or 72 hours after treatment of whole small ponds at rates of 1.0 ppm c.e. diquat. The limit of detection for diquat in these experiments was ~0.01 ppm c.e. This rapid dissipation was believed to be due to high turbidity because of a heavy phytoplankton bloom.

Similar observations were made on Millers Pond microcosms treated at 0.5, 1.0 and 5.0 ppm during the late fall in 1969. The half-life and persistence of diquat in the water column was very short. The half-life and maximum persistence of diquat in the 0.5 ppm c.e. treatment was <1.0 to <2.0 days and between 2.0 and 5.0 days (water concentration =< 0.01 to 0.03 ppm on day 4), respectively. The half-life and maximum persistence of diquat in the 1.0 ppm c.e. treatment was <1.0 to <2.0 days and between <1.0 and <6.0 days (water concentration = ≤ 0.07 on day 5), respectively. The half-life and maximum persistence of diquat in the 5.0 ppm c.e. treatment was <1 to <4 days and between 3 and 5 days (water concentration = ≤ 1.29 ppm c.e. on day 4), respectively. These short half-life and persistence times were found in turbid water with a dark green color characteristic of a phytoplankton bloom. The short half-lives and persistence times were believed to be correlated with the turbidity of the treated water.

However, treatments applied in the spring (1970) to the same site had much longer half-lives and persistence times. The half-life and maximum persistence of diquat in the 0.5 ppm c.e. treatment was <4.0 to 6.0 days and >13 days (water concentration = 0.21 ppm on day 13), respectively. The half-life and maximum persistence of diquat in the 1 ppm c.e. treatment was >13 days and >13 days (water concentration = 1.75 ppm on day 13), respectively. The half-life and maximum persistence of diquat in the 5 ppm c.e. treatment was ~8 days and > 8 days (water concentration = 2.4 ppm c.e. on day 8), respectively. The water in these microcosms was clear water and although aquatic plants were present, they did not appear to impact the concentration of diquat in the water column to a significant degree. Another study was conducted in the fall of 1970, and the results were similar.

Factors that influenced dissipation of diquat from the water column of Millers Pond included the time it takes for diffusion of diquat through the water column to occur, and adsorption of diquat by plants, algae, fish or other biota. The higher concentrations presented here (5 ppm c.e.) are much higher than is necessary to control most aquatic weeds (0.5 ppm to ~1.0 ppm c.e.). Hiltibran et al (1972) presented evidence that both susceptible (Sago pondweed) and non-susceptible weeds (American pondweed) can remove 70% to 80% of the applied diquat within a 48-hour period. Since diquat was usually not detected within 24 hours of application at normal rates, aquatic plants and algae must rapidly take up diquat from the water. Turbidity from either suspended sediment or the presence of algae have been shown to be significant factors in the dissipation of diquat from the water column (Selman and Upchurch, 1963 in Yao, 1967 and Bowmer, 1982).

In whole pond treatments near Wake (North Carolina), 1.0 ppm diquat applied on May 1st was found to dissipate to concentrations of 0.2 to 0.4 ppm within six hours of application and by 24 hours after application, diquat was not detected at concentrations higher than 0.03 ppm. After 36 hours of dissipation, diquat was not detected (LOD = 0.003 ppm). The half-life of diquat at the three treated ponds was <6 hours and the DT95 was approximately 20 hours. The authors attributed the rapid disappearance of diquat to dense infestations of filamentous algae (*Pithophora*) and water meal (*Wolffia* sp.). They also claimed that the rapid disappearance of diquat was due to rapid adsorption of diquat to high clay particulate content that is characteristic of Piedmont water impoundments and to photolysis. It is unlikely that photolysis plays a significant role in the degradation of diquat unless photoactivators generated from natural sources are present (Langeland and Warner, 1986).

After treating Orange Lake (Florida) with diquat, diquat + 1% polyacrylamine or diquat plus 1% polycarboxylate, it was found that the average concentration of diquat was higher in the top third of the water column than in the middle and bottom thirds of the water column. Furthermore, the concentrations in the top of the water column were higher when thickeners like polyacrylamine or polycarboxylate were used than when they were not used. This conclusion is contrary to the prevailing wisdom that thickeners allow herbicides to sink more deeply into the water column where they will be most effective. At least for diquat, under the treatment conditions at Orange Lake (4 acres of 1.3 to 1.7 meter depth treated with 4 lbs c.e. diquat/acre = ~0.27 ppm c.e.), thickeners did not aid in sinking. The water dissipation half-life of diquat under all treatment conditions was 25 to 39 hours (Langeland et al, 1994). Diquat did not move significantly outside the treatment area with 90% of all water samples taken at the edge of the treatment plot containing less than 0.05 ppm c.e. diquat. At 200 feet outside the treatment area, all water samples contained less than 0.05 ppm c.e., which indicates that for this treatment situation a setback distance of more than 200 feet is not necessary. The results for potential setback distances at Orange Lake were very similar to the results obtained from the modeling work done by Waterborne (1995).

In the Guntersville Reservoir (Tennessee), a 20 acre plot (3 feet deep) was treated with 0.4 ppm diquat plus 0.4 ppm Komeen®. Diquat dissipated to water concentrations of 0.011 ppm c.e. within 8 days, and it was not detected (<0.05 ppm c.e.) 15 days after treatment. Concentrations of diquat in sediment were much lower than in many of the other reviewed studies with 0.67 ppm c.e. diquat found 6 hours after treatment and 1.67 ppm c.e. diquat found 29 days after treatment. Diquat concentrations in plant tissue were 231 ppm at 6 hours after treatment, 100 ppm at 48 hours after treatment and 3.16 ppm c.e. 29 days after treatment. The half-life of diquat in water, soil and plants was 3.18, 73.8 and 5.38 days. Sorption and microbial degradation were believed to be the major factors affecting the dissipation of diquat. It is apparent from this data that adsorption by both the sediment and the resident aquatic plants may strongly influence the concentration of diquat in the water column. However, the importance of microbial degradation is not adequately demonstrated in this field study.

In studies conducted in Washington State, Serdar (1997) found that the half-life of diquat in the water column at Lake Steilacoom ranged from 2.6 days to 3.4 days. Lake Steilacoom was treated in its entirety (except for areas immediately around the inlet and outlet and in water more than 15 to 17 feet deep). Concentrations of diquat were highest in a treated embayment and in Chambers Creek (just below the dam) for the first three days after treatment at 0.13 ppm c.e. on June 5, 1996. Twenty-four hours after treatment, the concentration of diquat in the embayment was 0.087 ppm c.e. However, the concentration of diquat in the embayment dropped to 0.059 ppm c.e. 3 days after treatment. The concentrations of diquat in Chambers Creek were 0.091, 0.074 and 0.036 ppm c.e. at 1, 2 and 3 days, respectively. Concentrations of diquat were lower in open areas 300 feet off shore and within 300 feet of the mouth of Ponce de Leon Creek, which provides the dilution water for Lake Steilacoom. The concentration for the first 3 days after treatment at these sampling sites ranged from 0.033 to 0.057 ppm c.e. and did not vary significantly. By 7 days after treatment, the concentrations of diquat in water had dropped to less than the drinking water standard (0.02 ppm c.e.) and by 12 days the highest concentrations of diquat ranged from 0.003 to 0.007 ppm c.e. Since the concentration of diquat in water exceeds the maximum contaminant level goal (0.02 ppm c.e.) for the first 3 days after treatment, water in Lake Steilacoom should not be used for drinking, livestock watering, or irrigation for the first 3 days after treatment at 0.13 ppm c.e. (4 lbs a.e./acre). Furthermore, some of the more susceptible invertebrates (like *Hyallella azteca*) and fish (striped bass) may be adversely impacted in the presence of diquat in the water column. However, labeling and use of this product as a restricted use herbicide may prevent damage of the fish biota.

At Gravelly Lake, only 8 acres of the south shore and 2 acres of an embayment on the NE shore were treated for the control of *Elodea* and pondweeds at 0.11 ppm c.e. Except for the first four hours, the concentrations of diquat in the water of both treated and untreated areas of Gravelly Lake was <0.0028 ppm c.e. Diquat could not be detected at concentrations higher than 0.0005 ppm after 6 days of dissipation. Treatments of this type at Gravelly Lake should not impact recreational use or the use of diquat-treated water for drinking, irrigation or livestock watering. Furthermore, the concentrations of diquat found in Gravely Lake are not likely to adversely impact aquatic organisms (Serdar, 1997).

Treatment of the Kelowna Yacht Basin (3.7 acres) on Lake Okanogan at concentrations of 0.75 ppm diquat plus paraquat produced diquat water concentrations of ~0.5 ppm c.e. immediately after treatment. These concentrations fell to 0.21 to 0.24 ppm c.e, 12 hours after treatment and 0.044 to 0.071 ppm c.e 36 hours after treatment. Complete dissipation of diquat from the water column occurred in 60 hours (LOD = 0.05 ppm c.e.). Diquat was not found in the sediment until 2 weeks after treatment and then the detected concentrations ranged from <0.05 to 0.54 ppm c.e. In a similar experiment at Kin Beach (Lake Okanogan), 30 x 45 foot plots were treated with 1.0 plus 1.0 ppm c.e. of diquat plus paraquat. No diquat was detected in the water column after 43 hours. Diquat appeared to be rapidly eliminated from the water column due to extensive uptake by Eurasian watermilfoil and adsorption and inactivation by contact with soil/sediment (Water Investigations Branch, 1975).

Treatment of a large 10 acre cove in Findley Lake (New York) with the maximum use rate of diquat (4.0 lbs c.e./acre) completely controlled *Myriophyllum* spp. within 10 days of application. The water half-life of diquat within this cove was one to two days and fell below detectable levels (0.005 ppm c.e.) between 4 and 8 days after treatment. Concentrations of diquat were ~0.08, ~0.05 and ~0.02 ppm c.e. at one, two and four days, respectively after treatment. One day after treatment, and at all subsequent times, the concentrations of diquat in the water column did not vary according to depth at 6, 24 or 42 inches below the surface.

Treatments of the perimeter of Chautauqua Lake (New York) at concentrations ranging from 3 to 4 lbs c.e./acre did not persist beyond the day of treatment. The water half-life in these perimeter treatments was less than one day with initial concentrations varying between 0.145 and 0.881 ppm c.e. The initial concentrations of diquat varied according to depth with the concentrations being ~0.3, ~0.4 and ~0.7 ppm c.e. at 6, 24 and 42 inches in the area treated with 4 lbs c.e./acre. In the other treatment area of Chautauqua Lake the initial concentration varied with depth with concentrations being ~0.9, ~0.17 and 0.015 ppm c.e. at 6, 24 and 42 inches below the surface. No explanation was offered for these variations, but more rapid dissipation can be anticipated in spot or peripheral treatment areas due to extensive lateral mixing. (Sewell, 1969).

Modeling work with EXAMS II models making worst case assumptions of infinite water and plant half-lives and the maximum treatment rate (4 lbs a.e./acre) led Waterborne (1995) to conclude that diquat will not be seen in reservoirs more than 400 feet beyond the treatment area and that concentrations of diquat will fall below 0.02 ppm c.e. after treatment of a 70 x 200 to 70 x 1,000 foot area in 4 to 8 hours. Complete dissipation will occur in 24 to 48 or 48 to 120 hours for treated areas of 70 x 200, 70 x 600 or 70 x 1,000 feet, respectively. Similar results were obtained for riverine systems except that in slow moving water (2.4 to 3.5 inches/min), a 24-hour water use restriction with a 1,200 foot setback distance would be necessary to avoid exceeding 0.02 ppm c.e. after treatment of an area that is 200 feet long x 15 feet wide x 2 feet deep. If rapid moving water of 6-9 feet/min water is considered, a 4-hour water use restriction with a 1,400 foot setback distance is necessary. Under some of the modeled riverine systems the required setback distance maybe up to 1,600 feet. For this riverine environment, the bottom of the river was assumed to be sandy and the suspended sediment had a concentration of 1.0 mg/L.

Yeo (1967) found that treatment of reservoirs in California with diquat at concentrations varying from 0.125 to 1.0 ppm could produce water concentrations of diquat that varied considerably. Furthermore, this dissipation appeared to be controlled by turbidity and aquatic plant density with little impact from hardness, pH or temperature. The DT95 (time to 95% dissipation) of diquat in the water column was <4.0 days, 0.5 hours, ~ 8 days and <4 days in reservoir water treated with 0.125, 0.250, 0.500 and 1.0 ppm c.e. diquat. At the 1.0 ppm c.e. treatment concentrations, the diquat at all sites had dissipated to less than 0.06, 0.02 and 0.009 ppm in 4, 8 and 12 days, respectively. Therefore, no interference with water use for recreation, agriculture or home use should occur after 8 days of dissipation. After the death of treated aquatic plants (4 days), diquat was released and may become available to other aquatic plants. "When diquat is used for the control of aquatic weeds, the chief factor resulting in loss of the herbicide is uptake by weeds and adsorption to suspended silt and mud. This process is rapid and can account for removal of 90% of applied diquat within 24 hours." However, conditions of heavy weed infestation and high turbidity must be present for this to occur. If few weeds and little suspended sediment are present, the half-life for diquat may be fairly long. Plastic growth pools treated with 1.0 ppm c.e. diquat showed concentrations at 8 and 12 days that were ~0.04 and ~ 0.018. The concentration of diquat in growth pools at 12 days after treatment with 4.0 ppm diquat was 0.6 to 0.9 ppm c.e. Although the treatments of 0.5 to 1.0 ppm may be used for recreational, agricultural and home purposes, the 4.0 ppm treatment is still higher than the maximum contaminate level goal on the 12th day after treatment.

It is apparent that diquat has a very long system half-life (>1,000 days on sediment) and that accumulation can occur to very high levels (1.2 to ~70 ppm c.e.) on sediment. However, under conditions where diquat does not bind to sediment, microbial degradation may be important. It also appears that in most systems diquat will not desorb from sediment at high enough concentrations to cause significant damage to plant biota. It has been estimated that it would take 10 to 50 years of single season treatments to overcome the sorptive capacity of typical sediments for diquat before a release occurs. However, the time for sorptive capacity to be overcome is likely to be many times this number of treatments because this assumption assumes complete adsorption of diquat to the sediment and no dissipation due to photolysis, microbial degradation, and adsorption removal by hydraulic mechanisms prior to sorption to the sediment (Birmingham and Colman, 1983).

3.5.3 Physical and Chemical Factors

Few studies were found that were designed to ascertain the effects of various water and sediment parameters on the persistence of diquat in aquatic systems. Most were conducted under a controlled set of conditions, or were field studies under uncontrolled conditions. In most studies with variable conditions, it was not possible to separate out the effects of the specific variables discussed below. Although there is some dispute amongst researchers on the relative importance of photolysis, microbial degradation and adsorption of diquat to sediment, suspended sediment and plants, it is apparent that these factors play a significant role in the dissipation of diquat under certain conditions. The most important mechanism of dissipation from the water column appears to be adsorption to suspended and bottom sediment, adsorption to suspended sediment and algae with increased turbidity of the water column, removal of the pesticide by hydraulic means prior to adsorption and microbial degradation. Several of the factors discussed earlier that affect soil persistence can also be expected to influence persistence in aquatic systems.

- **Temperature**

  Temperature has a pronounced effect on the rate of chemical reactions and metabolic processes. In the case of diquat, where biological degradation predominates, temperatures outside the optimum range for diquat degradation are likely to result in decreased microbial degradation. Although microflora have been shown to degrade diquat to water soluble metabolites and volatiles (probably CO_2) (Simsiman and Chesters, 1976, Simsiman et al, 1976, Weber and Coble, 1968), the studies demonstrating the degradation of diquat were done at constant temperatures (25° or 37°C), so no effects were noted on the increase of microbial degradation with increased temperature. Water temperatures high enough to inhibit diquat metabolism in bacteria and fungi are unlikely to occur in Washington lakes. In this moderate climate, cooler temperatures would cause the most likely effect at night and at greater lake depths. Because of the high specific heat of water, it is a good thermal insulator, so the temperature of average size lakes does not vary much from night to day at the surface and even less at greater depths. Water temperatures of perhaps 10°C to 21°C may be expected in medium size lakes during the times when aquatic weed control is a concern. Smaller or shallow lakes may be expected to be warmer than larger lakes.

  For the most part, temperature has had little effect on the rate of sorption in various clays. At temperatures of 10°C and 55°C[3] kaolinite and monmorillonite (bentonite) clay preparations had identical adsorption of diquat. This lack of an effect from temperature changes was anticipated from a process (clay-organocation) interaction that was primarily coulombic forces (Weber et al, 1965 in Hayes et al, 1972). However, adsorption of diquat to vermiculite was retarded at lower temperatures in accord with an endothermic adsorption process (Pick, 1973 in Hayes et al, 1972).

  In deeper lakes, a thermocline can form during summer months wherein there is a sharp boundary between the warmer surface water and cold deeper water. Thermoclines could increase diquat persistence in two ways. 1) As there is little exchange of water across the thermocline, there is less water volume to dilute the herbicide, particularly in lakes treated over a large percentage of their surface. 2) Any diquat that penetrates the thermocline encounters a colder environment where degradation by microbes is slowed.

  Laboratory studies, typically conducted at 20°C to 25°C (68°F to 77°F), may yield half-lives that are somewhat shorter than studies in ponds or lakes. In addition, the latitude of the lake, with varying temperature regimes, make comparisons difficult. For example, Reinert and Rogers (1987) estimate that the work done by Simisman and Chesters (1976) gives an overall aquatic half-life of 3.8 days at 25°C. They further estimated that the aerobic and anaerobic half-life of diquat in weed-free systems was 31.9 and 49.5 days at 25°C and QSAR models predict a biodegradation half-life of <15 days at 25°C. However, it is noteworthy that the original research by Simsiman and Chesters (1976) indicates that while aerobic degradation in weed-free systems is faster than anaerobic degradation, the system half-life under these conditions is very long (>180 days). The field dissipation half-life of diquat in water is often just a few days if the right conditions exist for rapid adsorption or degradation (Frank and Comes, 1967, Hiltibran et al, 1972, Fujie, 1988, Gilderhaus, 1967 and Serdar, 1997). Half-lives and dissipation of diquat on sediment in the field are often quite long. For example, concentration of diquat in sediment remained as high as 1.7 ppm c.e. in ponds that had been treated with diquat at 0.27 lbs/ acre 4 years earlier (Beasley, 1965 in Hamer, 1994); Compliance Services International (CSI) has estimated that the concentration of diquat in sediments from experimental ponds in Denver, Colorado to be more than 163 days with concentrations of diquat as high as 36 and 24 ppm at 85 and 160 days, respectively (Frank and Comes, 1967). Concentrations of diquat in sediment reached maximum concentrations of 10 ppm c.e. for every 1 ppm c.e. applied to the water within about 12 weeks of application and no significant dissipation occurred by the 24th week after application (Gilderhaus,1967).

- **PH**

Alkalinity and acidity appear to have no significant impact on the degradation of diquat. Within a very large range (pH 5 and pH 9) hydrolysis has no effect on the degradation of diquat (EPA, RED, 1995) (Table 3.2.). However, diquat may not be stable at very high pH (pH> 9.0). This does not mean that pH has no effect on the properties of diquat. For example, diquat is more effective in controlling duckweed at pH 4 to 6 than at pH 8 to 10 (Guse, 1961 in Yeo, 1967). Also, diquat is more toxic to bluegills, fathead minnows and grass carp at lower pH than at higher pH (Surber and Pickering, 1962, El-Deen &Rogers, 1992 and Toobey et al, 1980).

In most natural waters, pH values are typically 6 to 9 (Reid, 1961, Goldman and Horne, 1983), aside from unusual lakes such as bog lakes, alkaline lakes, or those subject to acid rain, few of which are found in Washington State. The higher values are generally found during spring and summer, when more vigorous algal growth use large amounts of dissolved CO_2, driving the pH toward alkalinity through the carbonate/bicarbonate cycle. The more eutrophic (nutrient rich) a lake is, the larger the chance of enhanced algal growth and a higher pH.

The pH also seems to have little or no impact on the adsorption of diquat into bentonite or muck at high pH (8.5) or low pH (3.5). Furthermore, this complete adsorption of diquat at both high and low pH was not affected by changes in temperature between 0°C and 50°C (Harris and Warren, 1964).

Applications of diquat to water in outdoor polyester basins caused a drop in pH. These microcosms contained water, sediment, macrophytes, phytoplankton, zooplankton and destruents. After 2 years of stabilization, these microcosms were oligotrophic and macrophyte-dominated. This effect occurred when there was one application of diquat at 1.0 ppm or the diquat concentrations was maintained at 0.3 ppm for 8 weeks. The pH of both treated plots showed a marked decrease in pH immediately after treatment. After the application of diquat at 1.0 ppm, the pH tended to rise again as diquat dissipated. The 8-week contaminations with 0.300 ppm diquat resulted in a permanent decline in the pH value of these microcosms. While these were the conclusions of Neugebaur-Buchler et al (1994), the only information that could be directly ascertained from the data was that after treatment, the pH dropped within 1 month from ~9.0 to ~8.75. Furthermore, in the post treatment period, the pH did not recover and remained 0.5 to one pH unit below the control (8.5) for at least 6 months after treatment. It is well known that pesticides like diquat, which damage primary producers, will cause an immediate decrease in the pH of aquatic systems. These results show that herbicidal impact on reduction in pH could serve as an easy to measure end point for damage to primary producers. It can be conjectured that some of the drop in pH immediately after treatment with diquat may be due an exchange of H^+ cations for diquat cations in some clay sediments. As the H^+ cations enter the water, it can be expected that a small decrease in pH should occur.

- **Aerobic state**

The amount of oxygen dissolved in the water can have an effect on diquat persistence since degradation is largely the result of the action of aerobic microflora, which require oxygen, although it is apparent that diquat can also be degraded anaerobically by some aquatic microbes. Dissolved oxygen (DO) levels are typically 6 ppm to 10 ppm in well-mixed natural water bodies, though levels outside of that range are not unknown. The colder a water body, the higher the saturation value, or the maximum amount of DO that it can hold.

DO primarily enters the water from the atmosphere and from the photosynthesis of algae and submerged plants. Dissolved oxygen is consumed by fish and microflora in the water column on the sediments, and zooplankton and bottom-dwelling organisms such as aquatic insects. Plants also consume limited amounts of oxygen in their "dark cycle" respiration at night. Decay of vegetation and other organic materials, primarily on the lake bottom, also consumes significant oxygen. If a thermocline forms, water circulation is impaired and the water below the thermocline will become anaerobic if all of the dissolved oxygen is consumed.

One of the aquatic studies of diquat degradation previously cited described an increased rate of degradation when diquat was applied more than once (4 treatments occurring 1 month apart). For example, the half-life of diquat after its first use was 1.6 and 2.3 days at Bivens Arm and the Shongaloo Fisheries (Florida), respectively. Subsequent treatments at these sites decreased the half-life to 0.72 to 0.81 and 0.76 to 1.4 days, respectively; or the rate of dissipation increased by about 2-fold if the site had experienced previous diquat treatment (Fujie, 1988). Since sediment concentrations of diquat did not change significantly after the 29th day following the first treatment, it is likely that dissipation in the water column due to subsequent treatments was caused in large part by microbial degradation. However, the author (Fujie, 1988) does not discuss this possibility. Other general works discuss the mutation, selection, recruitment and acclimation of microbes more suited to the degradation of a herbicide during the first treatment. After the initial treatment, degradation would be more rapid for subsequent treatments due to the presence of degrading species that can utilize diquat as a sole source of carbon and energy. This has been demonstrated by Weber and Coble (1968) for an unidentified mixture of soil bacteria which can degrade unbound diquat to carbon dioxide. Only a few studies were found specifically addressing oxygen depletion resulting from macrophyte or algae kills by diquat. Most studies indicated that treatment with diquat did not significantly alter the dissolved oxygen content or oxygen budget of the treated water body [Gravely Lake, Lake Steilacoom (Serdar, 1997), Ivy Farm Lake (Chichester, Surrey, England) (Newman and Way, 1966), Piedmont ponds in South Carolina (Langeland and Warner, 1986), the Kelowna Boat Basin (Okanogan Lake) (Water Investigations Branch, 1977)]. At most of these sites the dissolved oxygen content was observed to be at or near saturation during the time after treatment when the greatest amount of plant decay was occurring.

However, heavy treatments with diquat were seen to cause the dissolved oxygen content (DO) to drop to near zero in some cases. For example, Gangstad (1986) found that constantly negative oxygen budgets occurred in the Chickahominy Reservoir (Virginia) at sampling stations where there was heavy and moderate growth of macrophytes due to the cessation of macrophyte photosynthesis and the beginning of bacterial degradation. Restoration of normal DOs at six weeks was not due to the recovery of the macrophytes, but due to a gradual loss of the dead plant material and a dramatic increase in phytoplankton levels (Gangstad, 1986). Severe deoxygenation was seen on Oxton Lake (Nottinghamshire, England) after treatment with 0.5 ppm paraquat. At Oxton Lake the DO dropped from 10 ppm prior to treatment to 0 ppm eight days after treatment when the most extensive plant decay was occurring. The DO largely recovered at Oxton Lake by the 37th day after treatment, probably due to a bloom of the green algae *Volvox* (Newman and Way, 1966).

Laboratory mesocosms with thick matted stands of Eurasian watermilfoil and American waterweed had DOs of 12 to 20 ppm, which was higher than saturation, prior to treatment with diquat (1.5 ppm) or diquat plus endothall (0.75 ppm plus 2.5 ppm or 0.375 ppm plus 0.75 ppm, respectively). However, 2 days after treatment with diquat or diquat plus endothall, the DO dropped to <1.0 ppm throughout most of the water profile, and anoxic conditions persisted for 4 days in the diquat treated systems and 12 days in the combined herbicide treated systems. Further treatment with Cutrine® did not control algae but the DO decreased dramatically on occasions where considerable algal death and decomposition occurred. This oxygen deficiency had an extremely detrimental effect on aquatic biota and caused the death of guppies and oxygen stress in snails in all systems receiving some form of diquat treatment.

During control efforts in Wisconsin (1990), deoxygenation can occur due to rapid use of oxygen during the decomposition of plants or algae and the loss of photosynthesis following herbicide treatment. Dissolved oxygen depletion is more rapid with diquat and copper treatments than 2,4-D or endothall treatments because death and decomposition is much faster. Field studies in Wisconsin (Daniel, 1972) found that applications of Cutrine® plus diquat at concentrations of 2.2 plus 3.0 ppm cause a rapid decrease of the DO to concentrations <1.0 ppm for 4 to 8 days after treatments. Surface water DOs returned to normal within 12 days of application but DOs at a 30 to 90 cm depth took nearly 40 days to return to normal. These low DOs were extremely unhealthy for the system and all fish in these treated mesocosms died within one day (possibly from asphyxiation) after treatment with Cutrine® plus diquat.

Work conducted by Inabinet (1976) with 2,4-D and diquat applied at rates close to the maximum use rate on August 4-6 and August 27 at Lake Marion (North Carolina) resulted in decreases in DO concentration from, 3.0 to 6.0 ppm on August 24 to 0.9 to 2.0 ppm at the surface and 0.5 to 1.2 ppm at the bottom on August 20. Seven days after treatment with diquat (September 2) the DO continued to decrease with surface water concentrations of 1 to 2.77 ppm and bottom water concentrations at 0.3 to 0.6 ppm. These DOs continued to remain low through September 11th. However, by October 3, the dissolved oxygen concentrations had recovered significantly (2.8 to 3.8 ppm at the surface and 3.1 to 3.3 ppm at the bottom) and by November 5, the DO had returned to pre-treatment levels.

Although not discussed by Fujie (1988), the slow degradation of diquat in the Florida ponds can probably be attributed to a low initial microflora population, rather than a depleted population. The increase in degradation rate after subsequent treatments of diquat may be due to the selection of a population of microflora that is more effective in utilizing diquat as a carbon and energy source. This selection can affect the dominant microflora species present and the genetics of these microflora populations. A population that is genetically more fit to utilize diquat as a sole carbon and energy source would probably degrade diquat more rapidly.

The speed of restoration of oxygen in a natural lake would be dependent on water temperature, mixing throughout the water column, introduction of oxygenated water from elsewhere in the lake, and the contributions of algal photosynthesis. In the case of a poorly mixed lake or of a treated shoreline area having a heavy macrophyte kill, reoxygenation might be delayed and diquat persistence extended. The effect would be more pronounced in lakes with heavy macrophyte growth given a whole-lake treatment.

Although Reinert and Rogers (1987) citing Simsiman and Chesters (1976) indicate that aerobic microbial metabolism (DT50 = 31.9 days) is more rapid than anaerobic metabolism (DT = 49.5 days), it is unclear where these DT50 (time to 50% dissipation) values came from. The original work by Simsiman and Chesters (1976) indicates that while aerobic microbial metabolism is ~8 times more rapid than anaerobic microbial in weed-free systems, 82% to 94 % of the diquat in the system remains unchanged and bound to the sediment for at least 95 days after treatment. In mesocosms, which contained realistic amounts of water, sediment and aquatic weeds, the system DT50 was about 3.8 days. It is noteworthy that in most aquatic systems, diquat persistence in sediments may be extensive even though water concentrations may drop below detectable limits within a few days of treatment with the maximum use rate (Wauchope et al, 1992).

- **Trophic state**

The trophic state of a natural water body may exert an indirect influence on diquat persistence. Because eutrophic (high nutrient concentrations) and high-end mesotrophic lakes are likely to have a larger macrophyte population, they are more likely to be included in an aquatic weed control problem. Therefore, a larger population of microflora which may degrade diquat can be expected to be present and persistence would be expected to be shorter. Conversely, when a large pool of carbon is available from decaying plant and animal matter, diquat may not be utilized by microorganisms as readily as in lower-trophic state lakes. This appears to be confirmed by the observation that Lake Mendota, Minnesota (a eutrophic lake) degrades diquat at a much more rapid rate than Lake Tomahawk, [an oligotrophic lake (Simsiman and Chesters, 1976)]. Similar effects were not observed when comparing the rate of dissipation in Lake Steilacoom (an eutrophic lake) and Gravely lake (an oligomesotrophic lake). However, there were considerable differences in the manner in which Lake Steilacoom and Gravelly Lake were treated. Lake Steilacoom was treated as a whole lake treatment and Gravelly Lake was treated with spot treatments along the southern shore and in a northeast shore embayment; therefore, the two lakes cannot be compared for the effects of their trophic states on relative dissipation rate. However, mesotrophic and especially eutrophic water bodies usually have a higher population of algae that can substantially contribute to the restoration of DO following an aquatic plant kill from a diquat application, and can thus help speed degradation by aerobic microflora such as *Pseudomonas, fluorescens*, *Rhizobium* sp., *Aerobacter aerogenes* and *Lipomyces starkeyi* (Simsiman and Chesters, 1972 cite Calderbank, 1968 and Wright, 1971). Other species of microflora, which have potential to degrade diquat are those that are known to degrade paraquat like the bacteria, *Corynebacterium fascians*. and *Clostriidium pasteurianum* (Baldwin et al, 1966 in Wright, 1971) and several unidentified species (Weber and Coble, 1968) and a short gram-negative rod-shaped bacterium capable of degrading a photolysis product (picolinamide) of diquat (Orpin et al, 1972).

One possible negative effect of a eutrophic state on diquat persistence should be mentioned. As stated above, the high nutrient levels usually give rise to a dense population of algae and various macrophytes as well as phytoplankton and benthic organisms and a proportionately larger amount of decaying organisms can be expected in a eutrophic lake. The first stages of this decay are generally aerobic, which uses dissolved oxygen. If conditions occur such as poor water circulation, the formation of a thermocline, or a population crash of a dense species population, the bottom of the lake (and possibly shallower depths) can become anaerobic. The inhibiting effects of low DO on diquat-degrading microorganisms then may become a significant factor in the persistence of the compound.

- **Adsorption to sediment**

  Adsorption and uptake of diquat by aquatic macrophytes and algae is addressed in Section 4 of this document. In general, adsorption to montmorillonite (bentonite) clay particles has been observed to decrease the availability of diquat for uptake by both plants and mixed populations of microorganisms. Other clays which do not form a significant lattice structure and have cation exchange sites only on the surface and angles of the clay particles do not significantly inhibit the uptake or degradation of diquat even at sediment particle concentrations higher than necessary to completely bind diquat or paraquat (Weber and Coble, 1968; Weber and Scott, 1966; Weber et al, 1969 and Coats et al, 1966). It has been observed that if Eurasian watermilfoil or duckweed is planted after treatment with diquat rapid regrowth of these species occurs (Daniel, 1972 and Birmingham and Colman, 1983). Most authors have found that the removal of diquat from the water column by sediment is the most important dissipation mechanism (Simsiman and Chesters, 1976; Haven, 1969; Gilderhaus, 1975; Frank and Comes, 1967; Hiltibran et al, 1972 and Birmingham and Colman, 1983). Adsorption by a variety of sediments was found to be irreversible at sediment concentrations of ~250 ppm c.e. However, levels of diquat adsorbed on sediment, which are biologically safe, certainly vary based on the species present and the kind of sediment present. For example, other work has found that the maximum concentrations of diquat irreversibly adsorbed onto soil/sediment between 16.5 and 165 ppm (Birmingham and Colman, 1983) between 100 and 350 ppm (Coats et al, 1984 and Hiltibran et al, 1972) and 2,500 ppm (Corwin and Farmer, 1966). As the amount of clay minerals increased, the amount of irreversibly adsorbed diquat also increased. Furthermore, clay minerals like montmorillonite (bentonite) with a layered or lattice structure were able to irreversibly sorb (90%) more diquat than clay minerals like vermiculite (20%), mica, illite or kaolinite (20%) that did not have a layered or latticed structure. Irreversible adsorption of diquat tends to reduce phytotoxicity necessitating larger applications in order to achieve the desired results.

  In some cases, diquat has been observed to bind to plant tissue only to be released to bind to sediment after the plants have decayed. A portion of the diquat residue may bind immediately after application to the sediment (Simsiman and Chesters, 1976). However, it was observed by a number of authors that diquat may not adsorb significantly to sediment until the treated macrophytes have decayed and diquat has been released from the decaying plant humic material (Frank and Comes, 1967). This is an important observation since aquatic macrophytes are unable to degrade diquat to a significant degree.

  In a turbid water body with significant amounts of particulate sediment (seston) suspended in the water, there is a greater solid surface area for diquat adsorption. The seston concentration can be high enough to completely inactivate diquat in irrigation drainage waters where the concentration is as high as 0.5 to 0.8 ppm. To achieve such an inactivating effect, the amount of seston suspended in the water would be quite high (in the range of 100 ppm) with an adsorption coefficient (Kd) of 113,000 (Bowner, 1982). Similar data were obtained for adsorption of diquat by montmorillonite and kaolinite clay (Weber et al, 1965) and adsorption of diquat obtained from silty lake sediments in New Zealand (Graham, 1976 in Bowner, 1982) and for adsorption of the related bipyrdylium compound paraquat by montmorillonite (Knight and Denny, 1970 in Bowner, 1982). These adsorptions and deactivations of diquat or paraquat are primarily due to the strong adsorption capacity (SAC) of the adsorbent; the SAC is usually less than the CEC for a given sorbent with the SAC:CEC ranging from 0.38 for kaolinite and 0.76 for monmorillonite. As a more conservative estimate, Birmingham and Colman (1983) found that the strong adsorption capacity would typically be about 7% of the maximum adsorption. A number of other authors including Hiltibran et al (1972) and Haven et al (1969) indicated that sorption to the seston included sorption to clay and silt suspensions or algae.

Depending on the sediment type, Birmingham and Colman (1983) estimated that it would take ten applications of a soil/sand mixture containing 22.7% organic carbon and more than fifty applications of natural Buckhorn Lake sediment containing 56.5% organic carbon to saturate the strong adsorption capacity and allow diquat to reenter the water column from the sediment. Dyson and Takacs (2000) ran a number of computer models that indicate this estimate is excessively conservative. It was estimated that an application of diquat at 1 Kg/ha/yr would result in maximum sediment concentrations after 40 years of 59 ppm assuming that all of the applied diquat reaches the sediment with no interception by weeds, and no degradation in water or sediment. This concentration is much lower than the maximum SAC (~100 to 2,500 ppm). Also models were run at treatment rates of 4.4 Kg/ha and compared with data collected from tidal creeks (Nomini, Virginia); the model yielded a one year expected concentration that was higher (10 ppm) than the results obtained from an actual treatment (1.2 to 4.1 ppm).

Data from soil studies also indicate that diquat applied as a desiccant has a 75% loss by 184 to 240 days after application (Anderson and Earl, 1996 in Dyson and Takacs, 2000). Also, treatment for 11 years at 1 Kg/ha resulted in the top 15 cm of soil containing only 0.88 ppm diquat. Use of diquat in Denmark at 32 field sites for many years resulted in a concentration of <0.2 ppm up to about 1.2 ppm. Thus, indications are that diquat residues will not exceed the biological deactivation capacity (SAC), which should result in an adequate sediment quality and should not adversely impact plants that are rooted in sediment that has been exposed to diquat (Bewick et al, 1984 in Dyson and Takacs, 2000).

Diquat can be released from soils or sediment by adding lime or fertilizers to soil which contain NH_4^+, Ca^{2+}, Mg^{2+} or K^+ at fairly high concentrations (0.05 Normal). However, water itself will generally not displace diquat to any significant degree even in young vermiculite (Weed and Weber, 1969). Even if diquat was released, harm would likely be prevented because diquat in sediment solution would be degraded by soil microorganisms and be strongly adsorbed by unaffected sediment. Extraction of diquat from sediment with concentrated sulfuric acid is the only process known to result in a permanent shift in equilibrium. However, this extraction results in the total destruction of sediment and is not environmentally relevant. During more than 30 years of use in over 100 countries for every conceivable agronomic and aquatic practice and soil/sediment type, there has not been a single case of adsorbed diquat residues being reactivated and causing noticeable damage to the environment (Dyson and Takacs, 2000).

- **Transport and dilution**

The most important and obvious physical processes affecting diquat concentration is adsorption to sediment and seston, particularly that containing even small amounts of montmorillonite (bentonite clay). However, the amount of diquat lost from mixing of water can be substantial particularly in large, deep water bodies. Since diquat is extremely water soluble (700,000 ppm c.e.), it should be easily transported within water currents in a lake. Obviously, the larger the area of a lake that is treated, the more water current will be needed to dilute and disperse the herbicide, with the extreme case occurring in whole-lake treatments.

In lakes without significant inflow or outflow, most dilution of diquat-treated water will occur through vertical movement in the water column. Solar heating is not as important to water movement in these lakes as the effects of wind. While sunlight can heat the surface waters, the warmer water tends to stay at the surface and little vertical circulation occurs. Wind can induce mixing between water depths even at low velocities. Surface water driven against a shoreline is forced downward and mixes with lower depth water, diluting the pesticide concentration of the surface water and may carry it into contact with sediment-dwelling microflora. However, the only field study conducted with diquat to determine its short-term mixing in the water column indicates that treatment at 0.28 to 0.344 ppm did not result in significant vertical dispersion when thickening (sinking) agents like polyacrylamine or polycarboxylate polymer were used to aide in dispersion through the water column. In the first two hours after application, the concentration of diquat was higher in the top of the water column (0.101 to 0.232 ppm) than in the middle (0.027 to 0.062 ppm) or bottom (0.025 to 0.072 ppm). Furthermore, significant lateral dispersion did not occur during the first 168 hours after treatment. In that time less than 6% of the samples collected 200 feet from the edge of the treatment plot contained more than 0.010 ppm diquat in the water column (Langeland et al, 1994). Somewhat different observations were made at Bivens Arm and Shongaloo Fisheries in Florida. Within the first day, the concentration of diquat in the top, middle and bottom of the water column were similar (0.08 ppm) in treated areas of the pond, although they were quite different immediately after treatment when there was 0.75 ppm in top waters, 0.27 in middle waters and <0.004 in bottom waters. Lateral dispersion was also rapid with concentrations in both treated and untreated areas being the same within the first day (0.07 ppm) (Fujie, 1988). These data were of particular interest since concentrations of diquat in sediment remained low (≤0.1 ppm) in the top 5 cm of sediment on the day after treatment. These data show that thorough dispersion (lateral and vertical mixing) can occur within 24 hours.

In lakes treated over only a part of their surface, dilution is a significant mechanism for reducing diquat concentration in the treated areas. Dilution can occur from wind-driven water currents or water flow through the lake, both of which can give rise to vertical and horizontal mixing and dilution. Movement of water through the lake can result from inlet streams and rivers, storm runoff outlets, submerged springs, or diffuse surface runoff into the lake from the surrounding basin. Operation of dams, weirs or other controls on lake outlets will impact the magnitude of water movement and consequently, the dispersal of treated water. For example, Chautauqua Lake (New York) was treated at 3 to 4 lbs c.e./acre and the diquat had dissipated to below detection limits in less than one day at all depths. Similar observations were made at Gravelly Lake (Washington) where a treatment of the southern perimeter and a northeast embayment at 0.11 ppm c.e. resulted in a very rapid dissipation of diquat from the treated area and a rise in the concentration of diquat at untreated areas (Serdar, 1997). After two days, the concentration of diquat in the treated area dissipated to 0.0013 ppm and the concentrations in the untreated areas rapidly rose to levels similar to that found in the treated area. These data show that thorough lateral and vertical mixing can occur in less than 24 hours and that this mixing can result in complete dissipation of diquat in only a few days after application as a peripheral or spot treatment.

If a large portion of the lake is treated, diquat can be carried out of a lake and into outlet streams if water movement is rapid or if there are insufficient microflora to break the herbicide down quickly. In view of the potential impacts on river biota, including fish far from the treated lake, water mass movement and the specific water budget for a particular lake must be taken into consideration when applying diquat. In western Washington, rainfall events, particularly in the months preceding July and after mid-September, can rapidly dilute diquat residues in a treated lake due to stream inflow and surface runoff, and can also move treated water into outflow streams more rapidly than anticipated before degradation is completed. This possibility is borne out by a whole lake treatment of Lake Steilacoom. The lowest concentrations of diquat occur in areas that were untreated and had a significant dilution from an inlet stream (0.043 to 0.0571 ppm at the mouth of Ponce de Leon Creek and in an open area 300 feet from the shore where significant water flow did occur (0.0325 to 0.0777 ppm). The highest concentrations occurred in the outlet stream to Chambers Creek (0.0355 to 0.0912 ppm) and in an embayment where the water flow was restricted (0.0587 to 0.0870 ppm). Apparently, the flow of water within Lake Steilacoom and the rate of microbial degradation is similar to that described for a typical lake. As indicated here, whole lake treatments can persist for a significant period of time with complete dissipation for Lake Steilacoom taking longer than 12 days. Coves in Lake Findley (New York) that were treated at the maximum use rate (4 lbs/acre) dissipated diquat at a much slower rate than in peripheral treatments. It took more than four days to completely dissipate diquat applied to these coves while it took less than 1 day to completely dissipate diquat from lakes where only the shore line was treated.

Monitoring of Guntersville Reservoir, a 68,000 acre lake on the Tennessee River, offers a good illustration of the effect of dilution and flushing on residue levels (Rogers et al, 1992). A liquid formulation of diquat dibromide plus Komeen® was applied to 20 acres at two gallons/acre and five gallons/acre, respectively (0.4 plus 0.4 ppm, respectively). Water and sediment were monitored for residues for 29 days. It is not clear from the report exactly where sampling occurred, but it is believed that samples were taken from the diquat plus copper treatment sites in the Guntersville Reservoir and also from nine potable water treatment plants in the Guntersville Reservoir. Significant residues were found in sediments in the diquat/Komeen® treatment area. Concentrations of 0.67, 1.1, 1.6, 2.16, 1.5 and 1.67 ppm diquat were found 0.25, 0.875, 8.0, 15.0 and 29 days, respectively after treatment. No diquat was found in sediment samples taken prior to treatment or 2.0 days after treatment. Maximum residues of 0.029 ppm were found in water 0.875 days after treatment and similar concentrations were found in water 0.25 and 2.0 days after treatment. By 8 days after treatment, only 0.011ppm of diquat was detected in reservoir water; and at 15 and 29 days after application no residues (<0.050 ppm) were found in the diquat/Komeen® treatment area. The results are similar to those found by Frank and Comes (1967) and Grzenda et al (1966) for water concentrations and somewhat less than has been typically seen for sediment concentrations. However, the concentration of diquat on sediment at approximate 5- to 10-fold, the water application rate displays a similar pattern to the concentrations of diquat on sediment reported by Fujie (1988) or Gilderhaus (1967). The setback distance of the treatment area from the nine potable water treatment plants appears to be sufficient since no diquat (<0.05 ppm) was seen in water coming from these plants. It is not clear how far these plants were located from the treatment areas, but it is probably farther than the minimum 270 to 400 feet recommended in Waterborne (1995) or 1,440 feet recommended in Singh and Martin (1997).

- **Type of formulation**

Only the Reward® Landscape and Aquatic (37.35%) diquat dibromide (20% diquat cation = 2 lb c.e./gallon) liquid formulation is being used in Washington State. Although there are other formulations, none of these are likely to be used for the control of aquatic weeds and filamentous algae. The use of liquid formulations usually results in higher initial water residues than with granular formulations since the entire application is present immediately in the water column. Sediment concentrations can be expected to be lower with liquid formulations since the chemical is injected in the upper water column relatively far from the sediment surface, and must be carried to the sediment by water currents or dispersion.

Although granular formulations of diquat are currently not used for the control of aquatic weeds and filamentous algae, granular formulations of aquatic herbicides are discussed here since major competitors of diquat including Aquathol® (Potassium Endothall) Hydrothol® 191 (Endothall mono(N,N-dimethylamine salt), Navigate® (2,4-D BEE) and Aqua-Kleen® (2,4-D BEE) are marketed in a granular form. Granular formulation can be expected to give higher initial sediment concentrations and lower water concentrations. As granular endothall and 2,4-D BEE is released from the granules over time, sediment concentrations will likely persist, albeit at low levels, for a longer period than with a liquid formulation and water concentrations are likely to be very low or non-detectable. Since the bottom waters in deeper lakes and shoreline areas are frequently colder than surface and mid-water depths, the higher sediment concentrations that granular formulations may produce are more likely to persist for a longer period in colder water due to inhibition or slowing of microbial metabolism of the chemical.

Except in very shallow, littoral areas, herbicide in liquid formulations can be expected to have less direct impact on deep-water or sediment-dwelling organisms than comparable granular formulations because of generally lower sediment concentrations and shorter persistence resulting from use of the liquid form.

Because diquat is deactivated by adsorption onto the sediment, formulation of diquat in a granular form is not advisable since the released diquat in direct contact with the sediment would be immediately adsorbed and deactivated by the sediment rendering diquat ineffective in the control of aquatic weeds.

For a detailed discussion of the effects of granules of Aqua-Kleen®, Navigate®, Aqauthol® or Hydrothol® 191, please see Section 3 for 2,4-D and Endothall. The function of granular formulations is discussed in great detail in these sections.

Table 3.5: Diquat Persistence in Aquatic Systems

System	Formulation	Initial application rate	Half-life (DT_{50})	Time to disappearance[1]	Comments	Reference
Lab aerobic aquatic microcosm*	Diquat dibromide	n.r.[2]	System[6]: n.s.d.[2] but DT 50 >31 days	n.s.d.[3]	Sediment type not reported but believed to have significant amounts of montmorillonite clay since 95-99% of the applied ^{14}C was found as unchanged diquat in the sediment. 30-day study.	EPA RED, 1995
Lab anaerobic aquatic microcosm*	Diquat dibromide	n.r.[2]	system: n.s.d. but DT50 > 9 months CSI estimate half-life 122 months	n.s.d. 5% of applied diquat degraded to one unidentified degradate.	Sediment type not reported but believed to have significant amounts of montmorillonite clay since 89 to 100% of the applied ^{14}C was found as unchanged diquat in the sediment 9 months after application.	EPA RED, 1995
Lab aerobic weed-less flask study with 5:200 sediment:water ratio	Diquat dibromide	5 ppm c.e.[4]	system: DT50 = 31.9 days	n.r.[2]	5:200 sediment:water weed-less flask study with sediment containing 39.1% clay (monmorillonite predominates). CSI does not believe that the original data supports the conclusions of Reinert and Rogers.	Reinert & Rogers,, 1987 citing Simsiman and Chesters, 1976
Lab anaerobic weed-less flask study with 5:200 sediment:water ratio	Diquat dibromide	5 ppm c.e.[4]	system: DT50 = 41.5 days	n.r[2]	5:200 sediment:water weed-less flask study with sediment containing 39.1% clay (montmorillonite predominates). CSI does not believe that the original data supports the conclusions of Reinert and Rogers.	Reinert & Rogers,, 1987 citing Simsiman and Chesters, 1976

Table 3.5
Diquat Persistence in Aquatic Systems (Continued)

System	Formulation	Initial application rate	Half-life (DT_{50})	Time to disappearance[1]	Comments	Reference
Lake Mendota sediment (8L) and 60L of water with a heavy growth of Eurasian water milfoil and American waterweed.	Diquat	1.5 ppm c.e.[4]	Water: DT50 <2 days Sediment: n.d. System: DT50 = 3.8 days	Water at 22 days 1% of applied-^{14}C as diquat 32% of applied-^{14}C as water soluble degradation products Sediment at 22 days = 19% of applied ^{14}C as diquat Volatilized at 22 days as $^{14}CO_2$ 48% of applied ^{14}C System at 22 days 20% of applied^{14}C as diquat	Diquat initially adsorbed by primarily by aquatic weeds and secondarily by sediment. Degradation of diquat by microbes associated with decomposing weeds. Diquat was degraded rapidly to water soluble metabolites and volatile metabolites (probably CO_2). This rapid metabolism of diquat is accompanied by a drop in DO between the 2nd and 8th day. After the initial algal bloom on the 8th day, recurring algal growth occurred in the system.	Simsiman & Chesters, 1976 and Reinert and Rogers, 1987

Table 3.5
Diquat Persistence in Aquatic Systems (Continued)

System	Formulation	Initial application rate	Half-life (DT_{50})	Time to disappearance[1]	Comments	Reference
Lake Mendota sediment (8L) and 60L of water with a heavy growth of Eurasian water milfoil and American waterweed.	Diquat	1.5 ppm c.e.[4]	Water: DT50 <2 days Sediment: n.d. System: DT50 = 3.8 days	Water at 180 days 0% of applied ^{14}C as diquat 10% of applied ^{14}C as water soluble degradation products Sediment at 180 days 13% of applied ^{14}C as diquat Volatilized at 180 days as $^{14}CO_2$ 77% of applied ^{14}C System at 180 days 13% of applied-^{14}C as diquat	Diquat initially adsorbed by primarily by aquatic weeds and secondarily by sediment. Degradation of diquat by microbes associated with decomposing weeds. Diquat was degraded rapidly to water soluble metabolites and volatile metabolites (probably CO_2). Only small amounts of diquat desorbs after it ahs adsorbed to the montmorollinite clay fraction.	Simsiman & Chesters, 1976 and Reinert and Rogers, 1987
Aerobic weed-free Lake Mendota (water/sediment 200:5) system	Diquat	5 ppm c.e.	Water: DT50 = <2 days DT95 = ~7days Sediment: n.s.d.	Water up to 180 days: remaining applied ^{14}C exists as soluble metabolites (2.72 to 7.10%) Sediment up to 180 days: remaining applied ^{14}C exists as unchanged diquat (92 to 104%) Evolved CO_2 up to 180 days (0.46 to 1.425%) of applied ^{14}C)	Dissipation is primarily controlled by adsorption to sediments. Once adsorption has occurred, very little desorption occurs. Approximately 21 days is necessary before adsorption equilibrium is reached. Rate of degradation to CO_2 is more rapid in aerobic than anaerobic systems	Simsiman and Chesters, 1976

Table 3.5
Diquat Persistence in Aquatic Systems (Continued)

System	Formulation	Initial application rate	Half-life (DT_{50})	Time to disappearance[1]	Comments	Reference
Anaerobic weed-free Lake Mendota (water/sediment 200:5) system	Diquat	5 ppm c.e.	Water: DT50 = <1 days DT95 = ~ 2 days Sediment: n.s.d.	Water up to 95 days: remaining applied ^{14}C exists as soluble metabolites (0.61 to 1.45%) Sediment up to 95 days: remaining applied ^{14}C exists as unchanged diquat (94 to 112%) Evolved CO_2 up to 180 days (0.12 to 0.27% of applied ^{14}C)	Dissipation is primarily controlled by adsorption to sediments. Once adsorption has occurred, very little desorption occurs. Approximately 7days is necessary before adsorption equilibrium is reached. Rate of degration to CO_2 is slower in anaerobic than aerobic systems	Simsiman and Chesters, 1976

Table 3.5
Diquat Persistence in Aquatic Systems (Continued)

System	Formulation	Initial application rate	Half-life (DT_{50})	Time to disappearance[1]	Comments	Reference
Aerobic weed-free Lake Tomahawk (sediment 200:5) system	Diquat	5 ppm c.e.	Water DT50 = <2 days DT95 = 14 days Sediment: n.s.d.	Water up to 65 days: remaining applied ^{14}C exists as soluble metabolites (1.5 to 1.64%) Sediment up to 65 days: remaining applied ^{14}C exists as unchanged diquat (100 to 102%) Evolved CO_2 up to 65 days (0.0.02 to 0.04% of applied ^{14}C)	Dissipation is primarily controlled by adsorption to sediments. Once adsorption has occurred, very little desorption occurs. Approximately 14 to 21 days is necessary before adsorption equilibrium is reached. Slower dissipation in Tomahawk system is due to about half as much montmorillonite clay as in the Mendota system. Lake Mendota sediments are eutrophic and Tomahawk sediments are not. Therefore, more bacteria or at least a different species spectrum is in Mendota sediment than Tomahawk sediment. So, degradation to CO_2 is more rapid in aerobic than anaerobic systems.	Simsiman and Chesters, 1976

Table 3.5
Diquat Persistence in Aquatic Systems (Continued)

System	Formulation	Initial application rate	Half-life (DT_{50})	Time to disappearance[1]	Comments	Reference
Aerobic weed-free Lake Mendota sediment:water (20:5) system	Diquat	5 ppm c.e.	Water: n.r. Sediment: n.s.d.	Water up to 65 days: remaining applied ^{14}C exists as soluble metabolites (1.82%) Sediment up to 65 days: remaining applied ^{14}C exists as unchanged diquat (100%) Evolved CO_2 up to 65 days (3.57% of applied 14C)	Dissipation is primarily controlled by adsorption to sediments. Once adsorption has occurred, very little desorption occurs. More rapid degradation to CO_2 occurs in 20:5 (water:sediment system) than in 200:5 water sediment system). Reason was hypothesized to be due to a greater amount of suspended particulate matter in the 200 mL water system than in the 20 mL water system. Or perhaps due to the more interment contact between sediment and water when the water to sediment ratio is smaller.	Simsiman and Chesters, 1976
Lake Seminole field applications to control watermilfoil	Diquat salt (dibromide or dichloride)	n.r.	Water: DT50 <1 day Sediment: n.d. Watermilfoil: detected in plants at 7 days.	Traces after less than 1 day. A trace would be just above the limit of detection.	Diquat was not detected in significant concentrations in water or sediment. Diquat was removed rapidly by watermilfoil in Lake Seminole.	Daly et al, 1968.
Model aquatic ecosystem	Diquat dibromide	n.r.	system: DT50 >1000 days	n.r.	Half-life estimated based on data from laboratory and field. Long half-life due to tight soil sorption.	Wauchope et al, 1992

Table 3.5
Diquat Persistence in Aquatic Systems (Continued)

System	Formulation	Initial application rate	Half-life (DT_{50})	Time to disappearance[1]	Comments	Reference
Typical effects in aquatic ecosystems	Diquat dibromide	1.0 ppm	n.r.	Water: 8 to 11 days Sediment: 6 months	Rapidly removed from water. Persistent in sediment	Newbold, 1975
Treatment of dual purpose ponds in Colorado for control of dense aquatic weeds	Diquat salt (dibromide or dichloride)	0.62 ppm	Water: CSI calculated DT50 = 0.63 days Sediment: CSI calculated DT50 = 163 days	Water: 4 to 8 days 0.01 ppm c.e. at 4 days Sediment: >160 days 24 ppm c.e. at 160 days	Rapid dissipation from water may be due to adsorption by dense weed growths. Concentrations in sediment do not peak till 56 days after treatment when 37 ppm of diquat was detected. This concentration occurred at a late date after treatment due to the release of diquat from dead and dying aquatic weeds.	Frank & Comes, 1967
Treatment of Reservoirs in California with diquat plus copper to control mixed aquatic weeds	Diquat	0.1 ppm diquat plus 0.3 ppm copper. Three treatments per season in mid May, Mid to Late July and Late August	Water: DT50 <1 day	Water: typically 1 day	Rapid dissipation of diquat was attributed to rapid adsorption by aquatic weeds, soil and detritus. Reservoirs with shortest diquat half-life had more planktonic organisms than reservoirs with longer diquat half-lives	Gangstad, 1986 and Stanley and Ganstad, 1987
Mesocosms in Wisconsin containing 3-inch layer of silt loam soil, 3-foot layer of water and 70 lbs of *Elodea* per pond.	Diquat	1.0 ppm c.e.	Water: DT50 = ~3 days Sediment: n.d.	Water: 10 to 21 days ~0.14 ppm c.e. at 10 days Sediment: >24 weeks 6.5 to 10 ppm c.e. at 24 weeks	Diquat continues to accumulate in sediment for 12 to 24 weeks and obtains a concentration of 6.5 to 10.1 ppm c.e in 24 weeks	Gilderhaus, 1967

Table 3.5
Diquat Persistence in Aquatic Systems (Continued)

System	Formulation	Initial application rate	Half-life (DT_{50})	Time to disappearance[1]	Comments	Reference
Mesocosms in Wisconsin containing 3-inch layer of silt loam soil, 3-foot layer of water and 70 lbs of *Elodea* per pond.	Diquat	3.0 ppm c.e.	Water: DT50 = ~4 days Sediment: n.d.	Water: 21 to 42 days 0.07 to 0.12 ppm at 21 days Sediment: >24 weeks 20 to 50 ppm c.e. at 24 weeks	Diquat continues to accumulate in sediment for 12 to 24 weeks and obtains a concentration of 20.2 to 50.5 ppm c.e in 24 weeks	Gilderhaus, 1967
Mesocosms in Wisconsin containing 3-inch layer of silt loam soil, 3-foot layer of water and 70 lbs of *Elodea* per pond.	Diquat	2 or 3 times at 1.0 ppm c.e. 8 weeks apart	Water: no sample Sediment: n.d.	Water: no sample Sediment: >24 weeks 54 to 72 ppm c.e. at 24-weeks	Diquat continues to accumulate in sediment for 24 weeks and obtains a concentration of 54.1 to 71.5 ppm c.e in 24 weeks	Gilderhaus, 1967
Mesocosms in Wisconsin containing 3-inch layer of silt loam soil, 3-foot layer of water and 70 lbs of *Elodea* per pond.	Diquat	2 times at 3.0 ppm c.e. 8 weeks apart	Water: no sample Sediment: n.d.	Water: no sample Sediment: >24 weeks 36 ppm c.e. at 24 weeks.	Diquat continues to accumulate in sediment for 24 weeks and obtains a concentration of 36.0 to ppm c.e in 24 weeks	Gilderhaus, 1967

Table 3.5
Diquat Persistence in Aquatic Systems (Continued)

System	Formulation	Initial application rate	Half-life (DT_{50})	Time to disappearance[1]	Comments	Reference
Ponds in Anderson, SC with thick stands of *Nitella*, heavy suspended sediment and bottoms of finely divide organic matter and silts	Diquat dibromide	2.5 ppm c.e.	Water: DT50 = ~0.9 days Sediment: no sample	Water: 7 to 14 days 0.01 ppm c.e. at 7 days	Very short half-life may be due to adsorption by thick stands of *Nitella* spp. but authors believe that higher turbidity due to suspended sediment was the main reason for the short half-life of diquat in water. Temperature was not believed to significantly impact the rate of degradation in these ponds. I t was not entirely understood why the persistence in this pond was so short since there is no reason to believe that deposits of organic matter in this pond should influence the persistence of diquat in water.	Grzenda et al, 1966
Ponds near Anderson, SC with negligible standing crops of vascular, very little suspended sediment and sandy clay sediment.	Diquat dibromide	2.5 ppm c.e.	Water: DT50 = ~3.0 days Sediment: no sample	Water: 27 to 30 days 0.01 ppm c.e. at 27 days	Longer half-lives in these ponds may be due to low densities of aquatic plants and low concentrations of suspended sediment. Temperature was not believed to significantly impact the rate of degradation in these ponds. I t was not entirely understood why the persistence in this pond was so long since sandy clay sediment should adsorb diquat more readily then finely divided organic matter.	Grzenda et al, 1966

Table 3.5
Diquat Persistence in Aquatic Systems (Continued)

System	Formulation	Initial application rate	Half-life (DT_{50})	Time to disappearance[1]	Comments	Reference
Nomini Creek, Virginia. Tributary to the Potomac River. Tidal stream with 4 foot depth Heavy stands of Eurasian water milfoil.	Diquat dibromide	0.35 ppm c.e.	Water: n.d. at any time. Sediment: n.s.d.	Water: <1 day Sediment: >356 days 4.07 ppm c.e. at Station 1 1.19 ppm c.e. at Station 2	Maximum concentration in sediment at both stations occurs at 36 days which is when diquat will typically be released from the humic matter of dead and decaying plants. Residues bound to sediments are not bioaccumulated by shellfish. Low levels of diquat in water attributed to rapid adsorption by silt clays in suspension or by bottom mud and the diluting effects of tidal currents. However, CSI believes that the heavy stands of Eurasian watermilfoil may also have contributed by adsorbing diquat from the water column.	Haven, 1969
Laboratory studies using clean tap water	Diquat	1.0 ppm c.e.	Water: DT50 = ~7 days	Water: >28 days 0.07 ppm c.e. at 28 days	System contained no sediment and was not sterile.	Hiltibran, 1972
Laboratory studies using aged tap water	Diquat	1.0 ppm c.e.	Water: DT50 = ~7 days	Water: >65 days 0.03 ppm at 65 days	System contained no sediment and was not sterile.	Hiltibran, 1972
Laboratory studies using lake water (Miller Pond)	Diquat	1.0 ppm c.e.	Water: DT50 <7 days	Water: >65 days 0.01 ppm c.e at 65 days	System contained no sediment and was not sterile.	Hiltibran, 1972
Laboratory studies using tap water = fish	Diquat	1.0 ppm c.e.	Water: DT50 = ~7 days	Water: >58-<65 days 0.01 ppm c.e. at 58 days	System contained no sediment and was not sterile. Fish Loading not specified	Hiltibran, 1972

Table 3.5
Diquat Persistence in Aquatic Systems (Continued)

System	Formulation	Initial application rate	Half-life (DT_{50})	Time to disappearance[1]	Comments	Reference
Laboratory studies using clean tap water	Diquat	5.0 ppm c.e.	Water: DT50 = ~14 days	Water: >65 days 0.34 ppm c.e at 65 days	System contained no sediment and was not sterile.	Hiltibran, 1972
Laboratory studies using aged tap water	Diquat	5.0 ppm c.e.	Water: DT50 = ~28 days	Water: >65 days 1.31 ppm c.e. at 65 days	System contained no sediment and was not sterile.	Hiltibran, 1972
Laboratory studies using lake water (Miller Pond)	Diquat	5.0 ppm c.e.	Water: DT50 >65 days	Water: >65 days 2.71 ppm c.e. at 65 days	System contained no sediment and was not sterile.	Hiltibran, 1972
Laboratory studies using tap water = fish	Diquat	5.0 ppm c.e.	Water: DT50 = ~14days	Water: >65 days 0.25 ppm c.e at 65 days	System contained no sediment and was not sterile. Fish Loading not specified	Hiltibran, 1972
Whole pond treatments applied in May to a small pond in Monticello, Illinois	Diquat	1.0 ppm c.e.	Water: DT50 <24 hours	Water: <24 hours	Rapid dissipation may have been due to the presence of a heavy bloom of phytoplankton at the time of application.	Hiltibran, 1972
Whole pond treatments applied in June to a small pond in Monticello, Illinois	Diquat	1.0 ppm c.e.	Water: DT50 <24 hours	Water: <24 hours	Rapid dissipation may have been due to the presence of a heavy bloom of phytoplankton at the time of application.	Hiltibran, 1972
Fall applications to enclosed areas in Miller pond	Diquat	0.5 ppm c.e.	Water: DT50 <24 to <48 hours	Water: 48 to 120 hours Up to 0.03 ppm c.e at 96 hours	Plastic sheets enclosed treated areas of Miller Pond. Water was extremely turbid at time of treatment. Turbidity probably due to phytoplankton bloom. Turbidity was believed to be responsible for short half-life	Hiltibran, 1972

Table 3.5
Diquat Persistence in Aquatic Systems (Continued)

System	Formulation	Initial application rate	Half-life (DT_{50})	Time to disappearance[1]	Comments	Reference
Fall applications to enclosed areas in Miller pond	Diquat	1.0 ppm c.e.	Water: DT50 >24 to <48 hours	Water: <24 to <144 hours. Up to 0.07 ppm c.e. at 120 hours	Plastic sheets enclosed treated areas of Miller Pond. Water was extremely turbid at time of treatment. Turbidity probably due to phytoplankton bloom. Turbidity was believed to be responsible for short half-life	Hiltibran, 1972
Fall applications to enclosed areas in Miller pond	Diquat	5.0 ppm c.e.	Water: DT50 <24 to <96 hours	72 to 120 hours up to 1.29 ppm c.e at 96 hours	Plastic sheets enclosed treated areas of Miller Pond. Water was extremely turbid at time of treatment. Turbidity probably due to phytoplankton bloom. Turbidity was believed to be responsible for short half-life	Hiltibran, 1972
Spring applications to enclosed areas in Miller pond	Diquat	0.5 ppm c.e.	Water: DT50 = >4 to 6 days	Water: >13 days 0.21 ppm c.e.	Aluminum sheets enclosed treated areas of Miller Pond. Longer half-life and time to complete dissipation was longer than in the experiment conducted in the fall probably due to low turbidity even though aquatic plants were present	Hiltibran, 1972

Table 3.5
Diquat Persistence in Aquatic Systems (Continued)

System	Formulation	Initial application rate	Half-life (DT_{50})	Time to disappearance[1]	Comments	Reference
Spring applications to enclosed areas in Miller pond	Diquat	1.0 ppm c.e.	Water: DT50 > 13 days	Water: >13 days 1.75 ppm c.e.	Aluminum sheets enclosed treated areas of Miller Pond.. Longer half-life and time to complete dissipation was longer than in the experiment conducted in the fall probably due to low turbidity even though aquatic plants were present. Water concentrations of diquat did not vary for 13 days of the study.	Hiltibran, 1972
Spring applications to enclosed areas in Miller pond	Diquat	5.0 ppm c.e.	Water: Dt50 = ~8 days	Water: >8 days 2.4 ppm c.e.	Aluminum sheets enclosed treated areas of Miller Pond. Longer half-life and time to complete dissipation was longer than in the experiment conducted in the fall probably due to low turbidity even though aquatic plants were present	Hiltibran, 1972
Whole pond treatments of Smith Pond in the Piedmont Plateau, SC	Diquat	1.0 ppm c.e.	Water: DT <6 hours DT95 = 17 hours	Water: 33 hours <0.003 ppm c.e. at 33 hours	Rapid dissipation can be attributed to dense algae infestations by *Pithophora oedogoniana* and dense infestations of the vascular plant *Wolffia columbiana* as well as rapid adsorption to high clay particulate content and photolysis	Langeland and Warner, 1986

Table 3.5
Diquat Persistence in Aquatic Systems (Continued)

System	Formulation	Initial application rate	Half-life (DT_{50})	Time to disappearance[1]	Comments	Reference
Whole pond treatments of Rogers Ponds in the Piedmont Plateau, SC	Diquat	1.0 ppm c.e.	Water: DT50 <6 hours DT95 = 17 hours	Water: 33 hours <0.003 ppm c.e. at 33 hours	Rapid dissipation can be attributed to dense algae infestations by *Pithophora oedogoniana* and dense infestations of the vascular plant *Wolffia columbiana* as well as rapid adsorption to high clay particulate content and photolysis	Langeland and Warner, 1986
Whole pond treatments of Jenks Pond in the Piedmont Plateau, SC	Diquat	1.0 ppm c.e.	Water: DT50 <6 hours DT95 = 20 hours	<0.003 ppm c.e. at 30 hours	Rapid dissipation can be attributed to dense algae infestations by *Pithophora oedogoniana* and dense infestations of the vascuara plant *Wolffia columbiana* as well as rapid adsorption to high clay particulate content and photolysis	Langeland and Warner, 1986
Eutrophic Orange Lake, FL with polymer added to aid in sinking of test substance	Diquat	0.28 ppm c.e. plus no polymer	Water: DT50 = 25 hours	81% of all samples taken 61 meters from treated plot edges contained <0.00001 ppm c.e. 94% of samples taken 0 meters from treated plot edges contained l<0.050 ppm c.e.	Plot was 1.6 meters deep. Concentrations were: Top 0.101 Middle: 0.054 Bottom 0.072 ppm c.e. Rapid dissipation indicates that the disappearance of diquat is due to factors other than dilution. Surface covered extensively with Hydrilla.	Langeland et al, 1994

Table 3.5
Diquat Persistence in Aquatic Systems (Continued)

System	Formulation	Initial application rate	Half-life (DT_{50})	Time to disappearance[1]	Comments	Reference
Eutrophic Orange Lake, FL with polymer added to aid in sinking of test substance	Diquat	0.34 ppm c.e. plus1% v/v 30% polyacrylamine polymer	Water: DT50 = 25 hours	81% of all samples taken at 61 meters from treated plot edges contained <0.00001 ppm c.e. 94% of samples taken 0 meters from treated plot edges contained l<0.050 ppm c.e.	Plot was 1.3 meters deep. Concentrations were: Top 0.232 Middle: 0.62 Bottom 0.025 ppm c.e. Rapid dissipation indicates that the disappearance of diquat is due to factors other than dilution. Surface covered extensively with Hydrilla.	Langeland et al, 1994
Eutrophic Orange Lake, FL with polymer added to aid in sinking of test substance	Diquat	0.28 ppm c.e. plus 1% v/v 30% polycarboxylate	Water: DT50 = 39 hours	83% of all samples taken 61 meters from treated plot edges contained <0.00001 ppm c.e. 91% of samples taken 0 meters from treated plot edges contained l<0.050 ppm c.e.	Plot was 1.7 meters deep. Concentrations were: Top 0.156 Middle: 0.027 Bottom 0.03 ppm c.e. Rapid dissipation indicates that the disappearance of diquat is due to factors other than dilution. Surface covered extensively with Hydrilla.	Langeland et al, 1994
Six sites in Guntersville Reservoir (AL & TN)	Diquat plus Komeen®	0.4 ppm c.e. diquat + 0.4 ppm Cu Komeen®	Water: DT50 =3.18 days Sediment: DT50 3.85 days	Water: 8 day to 15 days Up to 0.006 ppm c.e. at 8 days Sediment: 15 to >29 days Up to 1.5 ppm at 15 days of 1.62 ppm at 29 days	15 to 33% of surface covered with aquatic plants. Adsorption and microbial degradation were considered to be the major fate processes affecting diquat persistence	Rogers et al, 1992

Table 3.5
Diquat Persistence in Aquatic Systems (Continued)

System	Formulation	Initial application rate	Half-life (DT_{50})	Time to disappearance[1]	Comments	Reference
Reservoir treatments	Diquat	0.125 ppm c.e.	Water: DT95 = <4 days	Water: 4 days ≤0.06 ppm c.e. 8 days <0.02 ppm c.e. 12 days <0.009 ppm c.e.	Concentrations of diquat in 0.125 ppm treatment plots were greater than in the 0.25 ppm c.e treatment plots probably due to lower weed densities	Yeo, 1967
Reservoir treatments	Diquat	0.250 ppm c.e.	Water: DT95 = 0.5 hours	Water: 4 days ≤0.06 ppm c.e. 8 days <0.02 ppm c.e. 12 days <0.009 ppm c.e.	Heavy weed infestation (duckweed) and extremely shallow water caused rapid dissipation of diquat. Concentrations of diquat recurred after death of plants	Yeo, 1967
Reservoir treatments	Diquat	0.500ppm c.e.	Water: DT95= ~ 8 days	Water: 4 days ≤0.6 ppm c.e. 8 days <0.02 ppm c.e. 12 days <0.009 ppm c.e.	Concentration varied from 0.250 ppm to 0.05 to 0.075 ppm c.e. immediately after application. Differences were thought to be due to high turbidity in reservoirs where low concentrations of diquat were found.	Yeo, 1967
Reservoir treatments	Diquat	1.000 ppm c.e.	Water: DT95 < 4 days	Water: 4 days ≤0.06 ppm c.e. 8 days <0.02 ppm c.e. 12 days <0.009 ppm c.e.	Shortest half-life of less than 0.5 hour occurs in reservoir with extremely turbid water. Author suggests that short half-life is due to adsorption onto suspended clay particles.	Yeo, 1967
Pool treatments to 4x4x2 ft deep with 2 inches of clay loam + 180 gallons of water	Diquat	0.500 ppm c.e.	n.r.	8 days <0.04 ppm c.e. 12 days <0.018 ppm c.e.	Hardness had no impact on dissipation rate.	Yeo, 1967

Table 3.5
Diquat Persistence in Aquatic Systems (Continued)

System	Formulation	Initial application rate	Half-life (DT_{50})	Time to disappearance[1]	Comments	Reference
Pool treatments to 4x4x2 ft deep with 2 inches of clay loam + 180 gallons of water	Diquat	1.000 ppm c.e.	n.r.	8 days <0.04 ppm c.e. 12 days <0.018 ppm c.e.	Hardness had no impact on dissipation rate.	Yeo, 1967
Pool treatments to 4x4x2 ft deep with 2 inches of clay loam + 180 gallons of water	Diquat	4.0 ppm c.e.	n.r.	12 days 0.600 to 0.900 ppm c.e.	Hardness had no impact on dissipation rate.	Yeo, 1967
Lake Steilacoom; Whole lake (320 acres) treatment.	Diquat dibromide	4lbs c.e./acre (0.134 ppm c.e.)	Water: DT50 = 3.4 days	≥12 days 0.007 ppm c.e	Mouth of Ponce de Leon Creek. Since water flow is causing dilution the diquat concentration is somewhat lower for the 1st three days 0.057 to 0.043 ppm c.e. than in the embayment (0.087 to 0.059 ppm c.e.).	Serdar, 1997
Lake Steilacoom; Whole lake (320 acres) treatment.	Diquat dibromide	4lbs c.e./acre (0.134 ppm c.e.)	Water: DT50 = 2.1 days	≥12 days 0.007 ppm c.e	Totally enclosed shallow bay in treated area. Poorly flushed . Therefore, for 1st three days, concentrations range from 0.087 to 0.059 ppm c.e.	Serdar, 1997
Lake Steilacoom; Whole lake (320 acres) treatment.	Diquat dibromide	4lbs c.e./acre (0.134 ppm c.e.)	Water: DT50 = 3.2 days	≥12 days 0.007 ppm c.e	300 feet offshore in treated area of north basin. Since it is in an open area of the lake concentrations are some what lower for first three days (0.047 to 0.033 ppm c.e. than in the embayment (0.087 to 0.059 ppm c.e.	Serdar, 1997
Lake Steilacoom; Whole lake (320 acres) treatment.	Diquat dibromide	4lbs c.e./acre (0.134 ppm c.e.)	Water: DT50 = 2.6 days	≥12 days 0.007 ppm c.e	Concentration of diquat appears to concentrate in the outflow to Chambers Creek, just below the dam for the first three day (0.091 ppm to 0.036 ppm c.e.	Serdar, 1997

Table 3.5
Diquat Persistence in Aquatic Systems (Continued)

System	Formulation	Initial application rate	Half-life (DT_{50})	Time to disappearance[1]	Comments	Reference
Spot treatment of Gravelly Lake	Diquat dibromide	4lbs c.e./acre (0.11 ppm c.e.)	n.r.	6 days 0.0006 to 0.0016 ppm c.e at 1 to 3 days	No inlet or outlet. Sampling in a non-treated area on SW shoreline. 550 feet from nearest treatment area	Serdar, 1997
Spot treatment of Gravelly Lake	Diquat dibromide	4lbs c.e./acre (0.11 ppm c.e.)	n.r.	>12 days 0.0010 to 0.0005 at 1 to 12 days	No inlet or outlet. Sampling in a non-treated area on NE shoreline. 00 feet from nearest treatment area	Serdar, 1997
Spot treatment of Gravelly Lake	Diquat dibromide	4lbs c.e./acre (0.11 ppm c.e.)	n.r.	6 days 0.0028 to 0.0009 ppm c.e. at 1 to 3 days	8 acre treated along southern shoreline. Very rapid dissipation.	Serdar, 1997
Treated aquarium with no sediment, no plants and no animals	Diquat	1.44 ppm c.e.	n.r.	n.s.d. in 143 hours	No apparent degradation in demineralized tap water	Shaw and Hopke, 1975
Model ecosystem with water sediment, plants and animals from Chautauqua Lake, NY	Diquat	1.50 ppm c.e.	Water: DT50 = ~22 hours Sediment: n.s.d.	Water: >195.5 hours 0.03 ppm c.e. a t 96.5 hours and 0.015 ppm c.e. at 120.5 hours Sediment: >195.5 hours 52 ppm c.e. at 195.5 hours plants 0.04 to 0.15 ppm at 195.5 hours	Diquat appears to be primarily removed from water column by the sediment. Concentration in plants after 195.5 days of exposure does not seem to be much higher than concentration in water.	Shaw and Hopke, 1975

Table 3.5
Diquat Persistence in Aquatic Systems (Continued)

System	Formulation	Initial application rate	Half-life (DT_{50})	Time to disappearance[1]	Comments	Reference
Kelowna Yacht Club boat basin on Lake Okanogan. 3.7 acres; 7-8 feet deep	Diquat dibromide	0.75 ppm c.e. diquat plus 0.75 ppm c.e. paraquat	Water: DT50 = ~12 hours Sediment: n.r.	Water: 60 hours 0.044 to 0.071 ppm c.e. at 36 hours Sediment: >2 weeks n.d. to 0.54 ppm c.e. at 2 weeks	Taken up rapidly by plants (Eurasian watermilfoil). Probably released by plants at two weeks and then adsorbed expensively to soil. Diquat not detected in sediment for 1st two weeks after application. Water quality problems due to a significant rise in ammonia and orthophosphate and a drop in the DO after treatment	Water Investigations Branch, 1977
Kin Beach, Lake Okanogan Two 30 x 45 foot plots	Diquat dibromide	1.0 ppm c.e. diquat plus 1.0 ppm c.e. paraquat	Water: DT50 <43 hours	Water; diquat n.d. at 43 hours	No comments	Water Investigations Branch, 1977

Table 3.5
Diquat Persistence in Aquatic Systems (Continued)

System	Formulation	Initial application rate	Half-life (DT_{50})	Time to disappearance[1]	Comments	Reference
Reservoir Model by EXAMS II	Diquat dibromide	4 lbs/acre spot treatment	Water: DT50 >74 days to infinity Plants DT50 >25 days to infinity	Water: 24 hours if treatment area 70 X 200 feet 48 hours if treatment area 70 x600 feet 48 to 120 hours if treatment are 70 x1000 feet Time to 0.02 ppm c.e. = 4-8 hours if treatment area 70 x 200 Time to 0.02 ppm c.e. = 4-8 hours if treatment area 70 x 200 to 70 X 600 feet Time to 0.02 ppm c.e. = 8 hours if treatment area 70 x 1000 feet.	Setback distance up to 270 to 400 feet. Important dissipation processes, are adsorption by soil and adsorption by plant matter. Binding is irreversible to clay minerals and Koc in peat may be as high as 54,000 L/Kg. Insignificant processes are hydrolysis, volatilization and oxidation, photolysis and microbial decay may be important in some situations.	Waterborne, 1995

Table 3.5
Diquat Persistence in Aquatic Systems (Continued)

System	Formulation	Initial application rate	Half-life (DT_{50})	Time to disappearance[1]	Comments	Reference
River Model by EXAMS II	Diquat dibromide	4 lbs a.e./acre spot treatment not more than 200 feet long x15 feet wide x 2 feet deep	Water: DT50 >74 days to infinity Plants DT50>25 days to infinity	Water: Under low flow conditions (2.4-3.5 inches /minute) with sandy sediment– 24 hours to reach <0.02 ppm c.e. Under high flow conditions (2.4-3.5 inches /minute) with sandy sediment– 4 hours to reach <0.02 ppm c.e.	Setback distance under low flow and sand sediment is 1200 feet. Setback distance under high flow with sand sediment is 1400 feet Worst case setback distance may be as high as 1,600 feet.	Waterborne, 1995
Findley Lake, NY 10 acre cove	Diquat dibromide	4 lbs a.e./acre	Water: DT50 = 1 to 2 days	Water: 4 to 8 days With a concentration of ~0.02 ppm at 4 days at all depths between 6 and 42 inches	Cove treatments are more persistent than peripheral treatments. *Myriophyllum* spp. was controlled within 10 days of application.	Sewell, 1969
Chautauqua Lake, NY Perimeter treatment	Diquat dibromide	4 lbs a.e./acre	Water: DT50 <1 days	Water: <1 day With a concentration of ~0.3 to ~0.7 ppm zero days.	Cove treatments are more persistent than peripheral treatments. *Myriophyllum* spp. was controlled within 10 days of application. Immediately after application, concentrations were significantly higher at the bottom of the water (0.67 ppm c.e.) than at the top of the water column (~0.3 ppm c.e.).	Sewell, 1969

Table 3.5
Diquat Persistence in Aquatic Systems (Continued)

System	Formulation	Initial application rate	Half-life (DT_{50})	Time to disappearance[1]	Comments	Reference
Chautauqua Lake, NY Perimeter treatment	Diquat dibromide	3 lbs a.e./acre	Water: DT50 <1 days	Water: <1 day With a concentration of ~0.15 to ~0.9 ppm zero days.	Cove treatments are more persistent than peripheral treatments. *Myriophyllum* spp. was controlled within 10 days of application. Immediately after application, concentrations were significantly higher at the top of the water (~0.9 ppm c.e.) than at the bottom of the water column (0.15 ppm c.e.).	Sewell, 1969
Various Pools	Diquat dibromide	0.3 lbs c.e./acre	n.r.	Sediment: >4 years 1.7 ppm	Diquat is very persistent in the bottom mud of pools and ponds.	Beasley et al, 1965 in Funderburk and Bozarth, 1967

[1] Detection limit in is typically: water = 0.004 ppm, sediment = 0.01 ppm
[2] n.r. = not reported
[3] n.s.d. = no significant degradation
[4] c.e. = cation equivalent
[5] DT_{95} = time to disappearance of 95% of initial residues
[6] system = soil and sediment residues are not distinguished in the reference
* = EPA guideline registration study

3.6 MICROBIAL DEGRADATION

Summary: *In the field, diquat probably does not primarily dissipate from the water column or sediment due to the action of aquatic microbes. As discussed above, after diquat has been adsorbed to sediment it is in a bio-inactive form and is not readily degraded by most aquatic microbes. However, in mesocosm studies containing a heavy growth of Eurasian watermilfoil and American waterweed, a system DT50 of 3.8 days occurs due to the action of aquatic microbes. After 22 days, only 20% of the applied* ^{14}*C-diquat remains unchanged and the bulk of that diquat (19%) remains adsorbed to the sediment. The remainder of the applied* ^{14}*C-diquat has been converted to unidentified water-soluble metabolites and 48% of the applied* ^{14}*C-diquat has been converted to volatile metabolites (probably* CO_2*). After 180 days of degradation in this mesocosm, 77% of the applied* ^{14}*C-diquat has been converted to volatiles,10% of the applied* ^{14}*C-diquat remains as water-soluble degradates and 13% of the applied* ^{14}*C diquat is bound unchanged to the sediment. The type of sediment present may be critical to the availability of* ^{14}*C- diquat or complete degradation to* $^{14}CO_2$*. If diquat is bound to monmorillonite at concentrations that do not exceed the CEC of this clay, mixed populations of microbes grown in a nutrient solution will not degrade diquat to* $^{14}CO_2$*. However, even if diquat is bound to kaolinite at concentrations that do not exceed the CEC of kaolinite, mixed populations of microbes grown in nutrient solution will readily degrade* ^{14}*C-diquat to* CO_2*, and the rate of degradation will be as fast or faster than the rate of degradation in nutrient medium without kaolinite. Other observed degradations of diquat in nutrient solution involve the photolytic degradation of diquat to picolinamide with subsequent degradation of picolinamide by a short gram-negative rod-shaped bacterium to products that can potentially enter the Krebb's cycle and form* CO_2 *as the final product. However, this final mineralization has not been observed in the degradation of picolinamide. Since photolysis has minimal impact on the degradation of diquat, the primary mode of diquat degradation is the action of microflora - bacteria and fungi - that are found in soil, water and sediment. A number of genera of these microorganisms have been identified by researchers and appear to be widespread in the soil and sediment, though not necessarily at high enough population levels to play a part in initially reducing diquat residues. Only Simsiman and Chesters (1976) and Weber and Coble (1968) have shown conclusively that microbes in aqueous solutions can extensively degrade diquat within a reasonably short time frame. However, these systems where significant degradation by mixed populations of soil microbes occurred were set up in an ideal manner to encourage rapid degradation of diquat. Although the work of Fujie (1988) and Frank and Comes (1967) are indicative of possible degradation of diquat by microbes in field situations and Dyson and Takacs (2000) indicate that degradation by soil microorganisms is an important factor in preventing environmental damage from the release of soil/sediment bound diquat, it has not been proven that microbial degradation of diquat is a significant factor preventing the build up of diquat in the sediment*

Some microflora can probably utilize diquat as their sole source of carbon. While other microflora can metabolize diquat, it is not clear if they can utilize it as their sole carbon source. Microbes which have been identified as capable of degrading diquat include the bacteria *Pseudomonas fluorescens*, *Rhizobium* spp., *Aerobacter aerogenes* and the yeast *Lipomyces starkyi* (Calderbank, 1968 in Simsiman et al, 1976 and Funderburk, 1969 in Wright, 1971). Other microbes that may be able to degrade diquat include unidentified microbes (Simsiman and Chesters, 1976; Weber and Coble, 1968), a gram-negative rod-shaped bacterium capable of degrading the diquat photolysis product (piconaolamide) (Oprin et al, 1972) and bacteria capable of utilizing paraquat (a dipyridyl compound related to diquat) including *Corynebacterium fascians* and *Clostridium pasteurianum* (Wright, 1971*)*. While these species are known to degrade diquat in aqueous media, it is not clear if they are capable of utilizing diquat as a sole carbon source. Calderbank, (1968 in Simsiman et al, 1976) have shown that at least three metabolites are produced from the degradation of diquat by *Pseudomonas fluorescens*, these metabolites were not identified. However, *Psedomonas fluorescens* has good potential to degrade bioavailable diquat since it can tolerate concentrations of diquat up to 2,500 ppm. Oprin et al (1972) has shown that a gram-negative rod-shaped bacterium can degrade picolinamide, which is a photolysis product of diquat, to a number of products including picolinate, 6-hydroxypicolinate, 2,5-dihydroxypyridine, malemate, maleate and fumarate. The last two metabolites are Krebb’s cycle intermediates, which are known to be further degraded to carbon dioxide. Simsiman and Chesters (1976) and Weber and Coble (1968), have shown that diquat that is bioavailable (bound but not irreversibly to sediment) can be extensively degraded to water-soluble metabolites, CO_2 and possibly other volatile degradates. It is not clear if the discussed microbes can utilize diquat as a sole carbon source but it may be possible since these microbes are known to metabolize the diquat photoproduct (picolinomide) to Krebb's cycle intermediates and carbon dioxide.

Two laboratory studies have presented evidence of the dependence of Diquat degradation on the presence of a microbial population.

In their laboratory microcosm studies of diquat degradation in Lake Mendota water and sediment, Simsiman and Chesters (1976) concluded that a rapid degradation of diquat to water soluble and volatile degradates was possible in aquatic ecosystems that included extensive growths of aquatic weeds, particularly Eurasian watermilfoil and American waterweed. From this data, Reinert and Rogers (1987) calculated a diquat aquatic systems half-life of 3.8 days. After 22 days, only 20% of the applied ^{14}C-diquat remained unchanged and most of this (19%) appeared to be bound to the sediment. Further degradation of sediment associated diquat was observed 180 days after application of ^{14}C-diquat, with no diquat remaining in the water and only 13% of the applied ^{14}C-diquat remaining bound to the sediment. This indicates that even diquat that is presumably irreversibly bound to the montmorillonite clay fraction of the sediment may be slowly released and degraded primarily to volatile components (77%).

Further work by Simsiman and Chesters (1976) indicates that diquat in weed-free aquatic systems may rapidly bind to the sediment and once bound to sediment may not readily degrade. It was estimated that the time to equilibrium in the adsorption process might be as short as 7 days in anaerobic systems and 14 to 21 days in aerobic systems. Once this adsorption occurred, very little degradation of diquat occurred with more than 90% of the applied diquat remaining bound to the sediment and for more than 180 days. Although degradation was seen to occur in these weed-free incubation systems, it was very slow. Although Reinert and Rogers (1983) estimated the half-life of diquat to be 31.9 days in aerobic systems and 49.5 days in anaerobic systems, it is unclear how this calculation was made, and the original paper by Simsiman and Chesters (1976) indicates a much longer half-life. In QSAR modeling experiments done by Reinert and Rogers, the biodegradation half-life was determined to be < 15 days. However, Wauchope et al (1992) determined that typical half-lives for diquat in most field situations would be greater than 1,000 days due to sediment binding.

Data from the field is unclear concerning the persistence of diquat in sediment. For example: Fujie (1988) indicates that diquat is rapidly dissipated with water half-lives ranging from 0.72 to 2.3 days and that accumulation (typically <1.0 ppm c.e.) is not extensive on sandy clay loam or sand sediments even after four applications closely spaced in time. Gilderhaus found that while water half-lives of diquat are generally short (~3 days) for environmentally reasonable application rates of 1 ppm, diquat accumulated on sediment to concentrations as high as 71.5 ppm c.e. when applied 3 times within a 56 day period. Frank and Comes (1967) found diquat half-lives of 0.8 days in the water column and over 163 days in the sediment with the concentration of diquat on sediment continuing to rise from the date of application through 56 days post application when diquat sediment concentrations reached levels of 36.8 ppm c.e. Haven (1969) found that diquat applied to a tidal basin dissipated so rapidly that it was never detected in the water column (<0.01 ppm) but was seen in tidal mud at concentrations ranging from 1.19 to 4.07 ppm even after 356 days of dissipation were allowed to pass. Various other authors have seen diquat persist in the environment with estimated half-lives or measurable persistence in the water column ranging from 0.9 to 1.6 days (Grzenda et al, 1966, Yeo 1967, and Langeland and Warner, 1986). Many possible mechanisms for dissipation were offered including photolysis, irreversible adsorption to the sediment, adsorption to the seston (suspended sediment and algae), and adsorption by aquatic macrophytes with subsequent release and adsorption to the sediment. However, no author seriously advanced the proposition that microbes play a major role in the degradation of diquat. Furthermore, only one field study has yielded data indicative of a significant contribution from microbes in the degradation of diquat, and this is only assumed for Bivens Arm and Shongaloo Fisheries (Florida) since the rate of dissipation increases when more than one application occurs in a short time frame and accumulation on sediment typically does not occur (Fujie, 1988).

The field data is to some degree verified by the effects of bacteria in laboratory studies conducted on Lake Mendota sediment. Simsiman and Chester (1976) aerobically incubated diquat (1.5 ppm c.e.) in a mesocosm containing 60L of water, 8L of sediment and a heavy growth of Eurasian watermilfoil and American waterweed for 180 days. The plants and the sediment rapidly removed diquat. Over 50 % of the applied ^{14}C-diqaut was removed from the aqueous phase within 2 days of application. By the 4th day, diquat was being removed more rapidly than the total ^{14}C-label which indicates that after 4 days, metabolism of diquat by microbes had become an important factor in the dissipation of diquat. This follows the general observation that a lag-phase occurs before a large enough population of microbes has accumulated to substantially participate in the degradation of an aquatically applied pesticide. However, relative to the field situation, this data may be artifactual since the experiment was set up in an ideal manner so that degradation by aquatic microbes would occur.

In many cases diquat may not be irreversibly bound to the sediment. For example, salt concentrations ranging from 0.05 to 2.0 of NH_4Cl, $ALCl_3$, $CaCl_2$, $MgCl_2$, KCl or $BaCl_2$ have been observed to desorb 15% of the strongly adsorbed component of diquat from montmorillonite clay and 70% to 90% of the SAC of diquat from vermiculite, or kaolinite (Reinert, 1989, Weber and Weed, 1969, Weber and Weed, 1968). However, it is unlikely that diquat is bioavailable at concentrations that would typically be found on sediments and soils. Furthermore, even these high salt solutions will not typically desorb diquat from clay minerals when diquat is still many times above the concentrations that will typically be found on sediments (Summers, 1980 and Malquori and Radelli, 1966 in Dyson and Takacs, 2000). Furthermore, even when diquat is sorbed onto clay at concentrations equal to the CEC (~Strong Adsorption Capacity), water generally will not release diquat adsorbed to montmorillonite (bentonite) and less than 2% of the applied diquat will be released from vermiculite. Because of these factors and the fact that diquat sorbs irreversibly to most sediments at concentrations of 250 to 2,500 ppm (Hiltibran et al, 1972 and Corwin and Farmer, 1966), it seems unlikely that diquat will be bioavailable or biodegradable (transformable) when sorbed to solids (Weber, 1972 in Simsiman et al, 1976). This may not be true for sandy soils which have relatively small amounts of clay; Tucker et al (1967) found that >80% of adsorbed diquat can be desorbed, and it has been found in the field that soils of this type do not strongly adsorb diquat (Fujie, 1988).

The degradation of diquat is much more efficient under aerobic conditions than under anaerobic conditions. It has been shown in weed-free laboratory systems that the amount of diquat metabolized to water soluble degradates and carbon dioxide is much higher under aerobic conditions than anaerobic conditions after 95 days of degradation. For example, the amount of

CO_2 produced after 95 days was 1.75% of the applied ^{14}C label in aerobic system and only 0.27% in anaerobic systems. Furthermore, the amount of water-soluble metabolites was 7.1% of the applied ^{14}C-label under aerobic conditions and 1.26% under anaerobic conditions. However, that portion of diquat adsorbed to the sediment is unlikely to desorb and therefore, also unlikely to be bioavailable or transformed to other products (Simsiman and Chesters, 1976). General data indicate that diquat adsorbs readily to most mineral and organic surfaces including ogano-clay complexes, peat, muck and organic soils as well humic substance, higher plants, lignin and cellulose. Even small amounts of montmorillonite clay incorporated into soil or sediment will make it unlikely that diquat will significantly degrade under most field applications scenarios (Simsiman et al, 1976).

There are seven degradates of diquat identified in Shearer and Halter (1980) and another five bacterial degradates of the photolysis product picolinamide (Oprin et al, 1972). No information on the persistence, ecological fate including mobility or biological effects of these compounds was located, although two of them are Krebb's cycle intermediates which are non-toxic and not likely to persist or impact the environment adversely (Wisconsin, 1980). These metabolites include TOPPS, picolinamide, picolinic acid, 3,4-dihydro-8-hydroxy-2H-pyrido (1,2-a)pyrazin-1,6-dione, 3,4- dihydro-2H-pyrido(1,2a)pyrain-1,6-dione, diquat monopyridone, diquat dipyridone, 6-hydroxypicolinate, 5-dihydroxypyridine, maleamate, maleate and fumarate. However, their toxicity, environmental fate and adsorption/desorption characteristics are unknown.

It is clear from the above examples and from numerous other diquat fate studies that diquat will be degraded rather slowly once it has adsorbed to sediment. However, in situations where sorption to sediment is low, degradation may occur more slowly after the first application of diquat than after subsequent applications. Furthermore, while the primary mode of dissipation for diquat is adsorption to sediment, seston (suspended sediment) and aquatic macrophytes adsorption, diquat may not sorb to high levels on sandy and sandy clay loam sediments. Under these conditions of poor adsorption, microbial degradation may play an important role in degradation of diquat and keep the sediment concentrations of diquat at low levels on the sediment.

3.7 MOBILITY

Summary: *Most of the data reviewed dealt with sorption to soil; there are a few privately produced studies dealing with sorption of diquat to sediments. Diquat exhibits variable adsorption and desorption to soil depending on individual soil parameters. In most soils, adsorption can be considered to be very high. Soil/water partition coefficients have been observed to vary from 15 L/Kg for sand sediment, 36-42 L/Kg for sand soil and ~2000 to ~10,000 L/Kg for sandy loam, sandy clay loam and loam soils. Sorption has also been seen to be very high for peat soil (17,000 L/Kg) and various sand/gravel sediment (30 to 1000 L/Kg), sand (1,000 to 10,000 L/Kg) and silty clay loam 10,000 to 60,000 L/Kg). Generally, if the soils/water distribution coefficient is greater than about five, a pesticide is generally considered to be immobile. Diquat is known to adsorb strongly to organo-clays, peat, muck, organic soils, humic substances, higher plants, liginin and cellulose. However, the greatest adsorption occurs on soils and sediments that have large amounts of lattice forming (expanding) clays like montmorillonite or bentonite. Less adsorption occurs on non-lattice forming (partially expanding and non-expanding clays) like, vermiculite, mica, illite and kaolinite. In situations where typical use rates of diquat are used (2.2 to 4.4 Kg c.e./ha = 0.375 to 0.75 ppm c.e.), diquat will be immobile after adsorption to sediment or soil surfaces. Leaching of diquat is likely to be negligible because of the strong adsorptive capacity of soils and sediments. pH does not appear to effect the*

bioavailability of diquat. However, heavy liming may cause desorption of diquat from soils to a degree that more sensitive species of plants may be adversely impacted. Also, use of high levels of fertilizers containing, Ca^{2+}, Mg^{2+}, NH_4^{2+}, K^+ *may increase the desorption of diquat from sediment by cation exchange with the sorbed diquat .*

Soil parameters interact so that it is difficult to pinpoint a single cause for high or low sorption in soil studies. Nevertheless, it is likely that diquat will bind to a moderate degree to sediments with high cation exchange capacity particularly if that high CEC is due to the presence of expandable clays like montmorollinite or bentonite. Partially expandable clays and non-expandable clays like vermiculite, mica, illite or kaolinite also strongly adsorb diquat but to a somewhat lesser degree than the expandable clays. Even soils containing high levels of organic matter may strongly adsorb diquat because these soils usually contain a high level of CEC. Salt solutions containing high amounts of exchangeable cations can desorb diquat from non-expandable clay sorbents or soils containing considerable organic matte. However, water will not desorb significant amounts of diquat from these sorbents, but may desorb diquat from sandy sediment/soils containing low amounts of clay or organic matter. Many lake bottoms have fluffy, light (flocculent) sediments and considerable amounts of suspended sediments rather than a solid surface, particularly in more eutrophic lakes with a large amount of decaying organic material on the bottom. The much larger amount of particle surface in these flocculent sediments greatly increases the likelihood of diquat adsorption compared with firm-surfaced sediment providing that these flocculent and suspended sediments have a high CEC. Turbidity caused by suspended sediments and suspended phytoplankton are known to be a major factor in removing diquat from a treated water column.

Mobility of diquat within the water column may also be affected by the sorption of diquat to aquatic macrophytes. It has been estimated that up to 70% of applied diquat could be temporarily adsorbed to aquatic macrophytes with ultimate adsorption and inactivation on the sediment occurring after the plants have sunk to the bottom of the water body, decayed and released that portion of diquat that has not degraded in close proximity to the sediment. Results from a single adsorption/degradation study indicate that the unidentified metabolites of diquat are either volatile or extremely water-soluble and do not readily adsorb to the sediment. The volatile component may consist of carbon dioxide and although the water-soluble metabolites have not been identified, there are at least three of them and they maybe Krebb's cycle intermediates.

Overall, evidence indicates that diquat at typical environmental concentrations bind strongly and irreversibly to most soils and sediments. This would normally raise concerns of potential ground water contamination. However, the binding is so tight that significant desorption is unlikely and in those sediments where diquat does not bioaccumulate, unbound diquat is either degraded or diluted and removed by hydraulic processes common in water bodies which have a slowly moving mass of water. These processes include both lateral and vertical dispersion as well as advection from the general flow of the water body. It is unlikely that significant amounts of diquat will be desorbed from the sediment due to an equilibrium shift in a short enough time period to adversely impact the use of water that has been treated with diquat. It has been conjectured in a number of review articles that such an equilibrium shift could cause an adverse impact on a water body and its resident biota. However, during more than 30 years of use in over 100 countries for every conceivable agronomic and aquatic practice and soil/sediment type, there has not been a single reported case of adsorbed diquat residues being released from the sediment or otherwise reactivated.

When diquat is applied to soil, no significant potential exists for the chemical to be carried down into the soil with water movement from rain and irrigation. Pesticides exhibit a wide range of leaching potential, from those that adsorb strongly to soil particles and are not released before they break down, to those that do not adsorb significantly (or adsorb, then desorb) and will travel considerable distances down through the soil, sometimes as far as the ground water table. The sorption of various chemicals to soil is affected in a number of ways by soil parameters such as organic matter, clay content and type, and pH. However, except in the case of purified sand, diquat has not leached below the top 0.5 to 1.0 inch of soil in sandy loam and Norfolk sand even after the application of 10 acre•inches of water. The main factor governing sorption and leaching of diquat in sand and sediment appears to be the cation exchange capacity and the amount of expandable clays (like montmorillonite or bentonite) found in the sediment (Coats et al, 1966)

Controlled laboratory "batch equilibrium" studies are designed to measure the adsorptive properties of pesticides to four representative soils (EPA RED, 1995). There are currently no comparable test guidelines specifically for sediment but the adsorptive properties of sediments have been reported by Ritter (2000) and Waterborne (1995). The results for these tests on a number of soils and sediments as well as estimates from modeling work are presented in Table 3.2.7. Only adsorption coefficients (K_{ads}) have been reported. Desorption coefficients (K_{des}) were not available, and it is not entirely clear what K_{des} values mean since after diquat is sorbed to soil or sediment at concentrations that typically range from 250 to 2,500 ppm, desorption does not readily occur in water solutions. Although, solutions with high concentrations (0.05 to 1.0 Normal) of $BaCl_2$, $CaCl_2$, $MgCl_2$, NH_4Cl or KCl may desorb diquat from partially expanding and nonexpanding clays (like vermiculite, mica, illite or kaolinite) when the diquat levels exceed the strong adsorption capacity (SAC) of the sorbent (Orwin and Farmer, 1966, Hiltibran, 1972, Coats et al, 1966, Summers, 1980 and Malquori and Radelli, 1966 in Dyson and Takacs, 2000). The soil partition coefficients Kd_{ads} and Kd_{des} are measures of the potential for adsorption to soil and for desorption from that soil, respectively, and are calculated as the diquat concentration in soil divided by the concentration in water at equilibrium in a soil/water system with a single diquat starting concentration in the water. Waterborne (1995) reported Kd values for several relevant sediments in Table 3.2.7. The Freundlich K_{ads} are another way of calculating leaching potentials, but use the results of a series of tests with different starting concentrations. The parameters are particular to the specific soil being tested, and soils are chosen to represent typical agricultural soil types. To calculate Kd_{ads} (and Freundlich K_{ads}), biologically active soil plus water containing radiolabeled diquat are put in a sealed vial and shaken slowly for several hours until an adsorption equilibrium is reached (no more diquat can be adsorbed by the soil). The amount of diquat in the water and soil is determined by measuring the radioactivity in each. If K_{des} values are also to be determined, the water is then removed, replaced with fresh water, and the vial shaken again to allow the diquat to desorb from the soil back into the water. From measurements then taken, the Kd_{des} is calculated in the same manner as Kd_{ads}. However, in the studies cited here, the amount of desorption is usually indicated by the percent of diquat that is desorbed from the soil/sediment. Taken together, the adsorption and desorption parameters indicate how well diquat is adsorbed to and released from that typical soil and hence, will give a measure of leaching potential.

Although there is some disagreement as to exact classification values, generally Kd_{ads} and Freundlich K_{ads} values greater than 5 are characteristic of compounds that are considered to be not appreciably mobile, values from about 1 to 5 indicate a potential for greater mobility, while values less than 1 denote considerable mobility potential. In a similar manner, high Kd_{des} and Freundlich K_{des} values or a low percentage of desorbed pesticide indicate that an adsorbed compound will remain bound to soil and resist being carried downward.

Kd and Freundlich K values are composite values measuring adsorption caused by any of several soil characteristics such as clays, aluminum content, cation exchange capacity (CEC), and organic carbon. Koc values represent an attempt to separate out the role of organic carbon in soil adsorption from the other factors. Because organic carbon plays a significant role in the soil adsorption of many pesticides, Koc values are often used to predict pesticide mobility. But since Koc depends on two variables (Kd and carbon content), it must be used with caution. In the case of diquat, Koc may be almost irrelevant since the controlling factor in the adsorption of diquat is probably the cation exchange capacity of the soil/sediment and the amount of expandable clays present in the soil/sediment.

Koc values are calculated by dividing Kd and Freundlich K values by the decimal percent of organic matter or organic carbon in a soil [(e.g. for a peat soil (Waterborne, 1995)], Freundlich Koc_{ads} is calculated as 17,000/0.31= 54,000. Koc values may give an idea of the importance of organic carbon in a soil or sediment in adsorbing a chemical. Koc values are generally numerically higher than Kd or Freundlich K values. A higher value indicates organic carbon is more influential in trapping a pesticide. For instance, in Table 3.7., a number of authors give Koc values for diquat that range from 205 L/Kg to 7,9000,000 (Waterborne, 1995, Wauchope et al, 1992, and Ritter et al, 2000). The lower values were estimates based on a QSAR model (Hunter et al, 1984 in Rienert and Rogers, 1987) or generated by estimates from microcosms where the concentration of diquat in water and soil were measured after an equilibrium had been established (Simsiman and Chesters , 1976 in Reinert and Rogers, 1986). Unfortunately, the percent organic matter was not available for most of these studies. Kd_{ads} were reported for a variety of soils and they range from 15 L/Kg for a sand sediment to 10,000 to 60,000 for a Louisiana silty clay loam sediment and ~2000 to ~10,000 L/Kg for terrestrial sandy loam, terrestrial sandy clay loam and terrestrial loam soils. It has been reported by Wauchope et al (1992) that the Koc on a typical soil is 1,000,000 L/Kg and Ritter has reported that the K_{ads} for typical sediment in Southeastern canals is ~5,000. It is clear from this data that diquat is likely to be immobile and biologically unavailable when it is sorbed to a typical soil or sediment.

It is emphasized that all of the "K" parameters discussed above are specific to a particular soil or sediment, and to the initial concentration of a chemical applied to the soil or to a sediment/water system. A Freundlich K for a particular soil is a single value calculated using the adsorption or desorption results from all of the initial concentrations used in an experiment, but a Kd is calculated separately from the result of each initial concentration. Unless specified otherwise, Kd and Freundlich K parameters reported in published literature are for adsorption; measurement of desorption values is rare. Where K values are given without the soil type and chemical concentration being specified, care should be exercised in using those values for evaluation of leaching potential.

3.7.1 Soil and Sediment

The adsorption constants (Kd or Freundlich K) located during this review deal with both soils and sediment. It is not clear, in the case of sediments, what kind of water body the sediment was obtained from. However, the location by state and the sediment/soil series type are specified. Soil mobility data are directly relevant to the expected behavior of diquat over-sprayed on shoreline vegetation and to some extent indicates what may happen if a lake level drops, exposing shoreline sediment to drying, soon after treatment. Soil data can also be reasonably extrapolated to predict to some extent the adsorption of diquat on pond and lake sediments. Sediment will usually have a higher organic material content than soils, except for muck soils, and therefore, soil tests of higher organic content soils could be used as a guide to anticipate the potential for diquat adsorption to higher-organic matter sediments. Unfortunately, the percent organic matter and CEC are often not indicated in studies where soil/water adsorption coefficients were calculated.

The EPA RED (1995) discusses mobility studies that were conducted according to EPA laboratory guidelines to determine the adsorption constants in three representative soils and a sand sediment. Each of the four soils was mixed with water to form a slurry, then ^{14}C-diquat acid was added to make several unspecified initial concentrations. After agitation for period of time long enough to obtain equilibrium, Freundlich K values were calculated for each set of concentrations for a given soil. Table 3.2.7 gives the results. In all of the soils, K_{ads} were very fairly high (15 to 10,000) indicating that diquat was strongly adsorbed to the soils and that it is unlikely that it will be readily desorbed back into solution. The soils used for this study did not have their organic carbon content or cation exchange capacity specified. Diquat sorption by soils has often been reported to be related to the cation exchange capacity of the soil (Corwin and Farmer, 1975; Hiltibran et al, 1972, and Weber et al, 1965 in Reinert and Rogers, 1987). However, sixteen soils and sediments with CEC and clay that varied considerably were seen to not desorb diquat until the concentration exceeded 250 to 2,500 ppm (Corwin and Farmer, 1975; Hiltibran et al, 1972 and Coats et al, 1966). One may conclude that while soil organic carbon played relatively little part in adsorbing the diquat, it clung tenaciously to the diquat that was adsorbed. A number of review articles have indicated that diquat sorbs readily to most solid surfaces including peat, muck and organic soils as well as organo-clay complexes and humic substances, higher plants, lignin and cellulose. It is also generally accepted that once adsorption of diquat has occurred, desorption is unlikely when concentrations of diquat on sediment are typical of environmental concentrations (Reinert and Rogers, 1987; Reinert, 1989; Simsiman et al, 1972; Dyson and Takacs, 2000 and Funderburk, 1969).

Waterborne (1995) reported K_{ads} values for a variety of sediments found in California, Louisiana, Florida, Wisconsin and New York. The percent organic carbon or organic matter was not reported. However, for California sand/gravel, New York sand/gravel and Wisconsin sand/gravel, the K_{ads} was 30 to 1,000; for Florida sands the K_{ads} was 1,000 to 10,000; and for the Louisiana silty clay loams the K_{ads} was 10,000 to 60,000. These findings indicate that diquat was strongly adsorbed on all of the sediments studied. It is unlikely that organic matter played a significant role in the sorption of diquat and the most important factor contributing to the sorption of diquat was probably the CEC and the amount of expandable clays like montmorillonite and bentonite. Diquat was adsorbed firmly by these sediments and is likely to be retained under typical treatment conditions. It has been noted by a number of authors that the main contributor to diquat adsorption is the CEC and the level of clay minerals found in the sediment or soil. It is apparent that other factors such as pH and temperature do not contribute significantly to the sorption of diquat (Weber and Best, 1972; Weber and Weed, 1974; Weber et al, 1965 in Hayes et al, 1975).

Soil constituents such as clay minerals can cause increased adsorption in some soils (Simsiman and Chesters, 1972; Corwin and Farmer, 1975 and Hiltibran et al, 1972), and adsorption of diquat on pure clays is also very high (Weber et al, 1965 in Reinert and Rogers, 1987) although it may not be as high in clay loam soils. Montmorillonite clay and kaolinite clay have been observed to have K_{ads} of ~700 to ~3,000 and 21 to 57 L/Kg, respectively. However, sandy clay loam soil and silty clay loam sediment have been observed to have extremely high K_{ads} (~10,000 to 60,000 L/Kg), which would lead to the conclusion that components other than CEC and clay mineral content can contribute substantially to the sorption of diquat.

Another method of determining if a herbicide is adsorbed and not easily leached is by applying the pesticide to the top of a soil column packed with environmentally relevant soils. When soil columns are packed with a sandy loam or a Norfolk sand treated with 16 lbs/acre ^{14}C-diquat and eluted with 10 acre•inches of water, diquat was immobile and did not leach in the soil column to depths of greater than 0.5 to 1.0 inches. However, in purified sand which presumably had a low

CEC, 75% of the applied radiation leached through three inches of sand and the remainder was evenly distributed throughout the soil column (Coats et al, 1966).

Attempts were made by Reinert and Rogers (1987) to estimate the Koc of diquat using data from a paper by Simsiman and Chesters that was primarily designed to determine the degradation and adsorption of diquat in weed-free microcosms. These Koc_{ads} values appeared to be fairly low (205 to 457 L/Kg) which would indicate that diquat may be mobile under certain circumstances. These values were similar to the QSAR estimate Koc_{ads} of 691 L/Kg (Hunter et al, 1984 in Reinert and Rogers, 1987). However, based on the data from real sediments and soils conducted in a manner compliant with U.S. EPA guidelines, these model Koc_{ads} values were discounted and are probably incorrect. General Koc_{ads} values used in SCS/ARS/CES and EXAMS II models range 32,000 to 7,900,000 L/Kg and these values are believed to have greater validity in light of the general conclusions that diquat adsorbs tenaciously to most sediment types and does not readily desorb in the presence of water unless that water contains very high concentrations of inorganic cations. For example, heavy liming at 20 tons/acre causes paraquat to become readily bioavailable and was more phytotoxic lower application of lime (1.6 tons/acre). Presumably, soils treated with fertilizers containing high concentrations of Ca^{2+}, Mg^{2+}, Fe^{2+} or K^{+} may also release paraquat at concentrations that could be phytotoxic. Since diquat is a similar compound but desorbs somewhat more readily, heavy liming or fertilization may cause a phytotoxic release of diquat from soil treated at higher use rates (Weber and Best, 1972 in Weber and Weed, 1974).

Simsiman and Chester (1976) aerobically incubated diquat (1.5 ppm c.e.) in a mesocosm containing 60L of water, 8 L of sediment and a heavy growth of Eurasian watermilfoil and American waterweed for 180 days. Diquat was rapidly removed by the plants and the sediment. Over 50 % of the applied ^{14}C-diqaut was removed from the aqueous phase within 2 days of application. By the 4th day diquat was being removed more rapidly than the total-^{14}C, which indicate that after 4 days metabolism by microbes had become an important factor in dissipation. This is not surprising since it has been observed that aquatic macrophytes extensively adsorb diquat at concentrations as high as 40 to 100 ppm (wet weight) or higher (Rogers et al, 1992; Coats et al, 1966; Way et al, 1971 and Birmingham and Colman, 1983) and that some of the pondweed species can adsorb up to 70% of applied diquat from aquarium water (Hiltibran, 1972). Furthermore, more than 90% of applied diquat can be adsorbed onto sediment under aerobic conditions in just a few days (EPA RED, 1995 and Simsiman and Chesters, 1976). The microbial metabolism of diquat has been discussed extensively in previous paragraphs but it is apparent that an aquatic system with extensive growth of microbes can degrade diquat under aerobic conditions with a half-life of only 3.8 days. The 80% of the applied diquat was metabolized to volatiles (48% possibly CO2) and water-soluble metabolites (32%) within 22 days after application; and 19% of the diquat remained bound to the sediment. Other experiments in weed-free systems indicated that diquat does not readily degrade, more than 90% of the diquat adsorbs to the sediment within just a few days, and that diquat remains adsorbed and unchanged on the sediment for up to 180 days (Simisman and Chesters, 1976 and EPA RED, 1995). Additional work in the field indicates that diquat adsorbed to sediment is not biologically available to most microbes and that unchanged diquat may remain bound to the sediment for more than a year (Haven, 1969; Beasley, 1966 in Hamer, 1994; Frank and Comes, 1967; Gilderhaus, 1967 and Fujie, 1988). Greater detail on the persistence of diquat in sediment has been discussed in previous sections.

Adsorption of diquat to suspended sediment and algae (seston) has been observed to be an important factor in removal of diquat from the water column. Usually a high level of seston increases the turbidity in the water. Ponds in Anderson (SC), Millers Pond (WI) and various reservoirs had a high turbidity and under these conditions diquat was removed rapidly from the water. Bowmer (1982) has indicated that high levels of seston (100 ppm with a $K_{ads} = 113{,}000$)

were sufficient to completely inactivate a 0.5 to 0.8 ppm application of diquat. However, it was suggested that once the strong adsorption capacity of this seston had been saturated by diquat at 8 ppm, control of aquatic weeds may be possible.

No studies that directly address the sediment mobility of diquat in the field are available. However, Fujie (1988) found that diquat sorbed to sand and sandy loam sediment at concentrations not higher than 1.2 ppm c.e. in ponds located in Florida. More typical concentrations found on the 0 to 5.0 cm sediment segment are 0.5 to 1.0 ppm c.e. and the concentration of diquat found in the 5 to 10 cm sediment segment is usually less than <0.01 ppm c.e. Therefore, he found that residues of diquat were relatively immobile, remaining in the upper sediment profile, though minor detections of diquat were made at the lower depth. Fujie (1988) further found that multiple treatments at the maximum use rate did not increase the concentration typically found in sediment. No explanation of this observations was given. However, it was stated that "once adsorbed, the residue is bound with little or no vertical movement through the sediment layer or desorption back into the water column." This would tend to indicate that most diquat would not desorb from most other sediments types since the sediments in these ponds are sands and sandy clay loams with only a low capacity to sorb diquat. Other field sites include ponds in Colorado, Wisconsin and tidal basins (Nomini Creek, Virginia). Diquat was found at these sites to be fairly high in the sediment with concentrations typically reaching levels that are ten or more times higher than the initial concentration applied to water (0.35 to 3.0 ppm). The concentrations on sediment were observed to not decrease or to decrease only gradually after complete adsorption had occurred between 36 and 56 days after application for the duration of the experiment (160 to 356 days). The dissipation of diquat from sediment has been discussed in greater detail earlier in this DOument. As diquat is slowly released from the sediment over a long period of time it is likely to be rapidly dissipated by readsorption to sediment, degradation by microbes, or dissipation by dispersion or advection. It is not clear in all cases, but the depth of the sampled sediment appears to be fairly shallow and in some cases may be only about 1.0 inch (Haven, 1969; Gilderhaus, 1967 and Frank and Comes (1967). These sediment samples were very difficult to sample and often consisted of extensively decomposed organic detritus and mud.

In the EPA RED (1995), no monitoring of wells, surface water or ground water was mentioned. However, EPA has found that diquat binds nearly irreversibly to the soil. It does not appear to be biologically available to aquatic organisms in surface water. Therefore, EPA does not require a surface water advisory. Also, since diquat binds nearly irreversibly to soil/sediment, it will not leach into the ground water. Diquat was not monitored in ground water between 1988 and 1995 in the state of Washington (Ecology, 1996) and it is not believed to be of great concern as a potential ground water contaminant.

It is possible, although unlikely, that wells in a real-world situation drilled very close to a pond or lake shore could draw water directly from a water body so rapidly that any diquat present in the lake would not have a chance to be diluted, broken down, or adsorbed to soil/sediment or aquatic weeds. Much would depend on the treatment rate of diquat in the water body, the soil type and porosity, the depth of the ground water table, the frequency and volume of pumping, and other hydrological parameters. Under a worst case scenario of a sand/gravel sediment with little or no clay or silt, that diquat would not adsorb and be inactivated and could, with a very heavy and continuous pumping of water, contaminate the ground water. However, there have been no credible reports of ground water contamination when diquat is used according to the label.

Many lake bottoms have fluffy, light (flocculent) sediments rather than a solid surface, particularly more eutrophic lakes with large amounts of decaying organic material on the bottom. The much larger amount of particle surface in these flocculent sediments greatly increases the likelihood of diquat adsorption compared with firm-surfaced sediment. However, organic matter itself may participate in only a minor way in the sorption of diquat. Diquat is most likely to sorb onto silty clay loams or other soil/sediments which contain high percentages of expandable clays. However, most review articles contain statements similar to that found in the EPA RED (1995) and indicate that diquat adsorbs tenaciously to most solid surfaces including peat, muck and organic soils as well as organo-clay complexes and humic substances, higher plants, lignin and cellulose. Birmingham and Colman (1983) and Hiltibran et al (1972) indicate that diquat may be adsorbed and removed from the water column by various species of algae and aquatic macrophytes.

3.7.2 Ground Water and Surface Water

Over the many years of its use as a terrestrial herbicide and aquatic herbicide, diquat has never been and observed to be desorbed and reactivated (Tyson and Takacs, 2000). Furthermore, the EPA RED (1995) has not reported any cases of ground water contamination from the proper or improper use of diquat. Because it is considered unlikely to contaminate ground water, Washington State has not monitored ground water for diquat or paraquat between 1988 - 1995 (Ecology, 1996; USGS, 1996). Nevertheless, the most likely routes for contamination are spills during mixing of application solutions at wellheads, illegal dumping, surface water runoff from treated fields, and movement down through the soils from heavily treated agricultural land. With the exception of contamination by spills or illegal dumping, none these routes of ground water contamination is likely since diquat tenaciously adheres to soil and sediment and is unlikely to be released from soil except under the conditions of very heavy liming (Weber and Best, 1972 in Weber and Weed, 1974) or heavy use of fertilizers containing high concentrations of cations like NH_4^+, Ca^{2+}, Mg^{2+}, and perhaps Fe^{2+} $Zn^{2,}$ or Cu^{2+} (Simsiman et al, 1976). With respect to ground water movement, the difference between terrestrial and aquatic weed control uses of diquat is that lakes provide, in essence, an isolated incubator in which diquat can be slowly released from the sediment and dissipated through slow microbial degradation or horizontal/vertical dispersion and advection by the slow flow of the water body (Ritter, 2000 and Waterborne, 1995).

The data reviewed in this DOument indicates that diquat is immobile in all environmentally relevant soils. In general, the higher the CEC and expandable clay content the more likely diquat is to adsorb to soil particles and hence the less mobile it will be. Temperature and pH have little or no effect on diquat adsorption (Weber and Best, 1972 in Weber and Weed, 1974 and Weber et al, 1965). However, while the time to soil sorption equilibrium is faster under anaerobic conditions (2 days) than under aerobic conditions (14 to 21 days), the ultimate adsorption (96% to 99%) of diquat is not affected by anaerobic or aerobic conditions (Simsiman and Chesters, 1976). Diquat has a very long persistence in aquatic systems and usually has fairly high concentrations of diquat (~0.5 to ~70 ppm) at the sediment surface, as a result of extensive sorption. Fairly high levels of diquat may persist on sediment for at least 180 days, and concentrations of diquat have been as high as 1.19 to 4.07 ppm c.e. for up to 356 days (Haven, 1969) and concentrations of diquat as high as 1.7 ppm have been seen on sediment for more than 4 years after treatment of a water body at 0.3 lbs/acre (Beasley et al., 1965 in Hamer, 1994). Even these high levels of diquat are tenaciously bound. Therefore, very little diquat would be available for movement either downward through the sediment or laterally into the soil surrounding the lake through subsurface water movement. Such movement would cause further dilution of the pesticide through continuing low-level adsorption to the soil that it moves through. A massive application to a small water body could result in movements of higher concentrations of diquat into surrounding soil/sediment before it had time to degrade.

No data were located relating to mobility of diquat degradates. The major initial diquat degradate due to photolysis is TOPPS and degradates due to the metabolic action of microbes are either water-soluble or volatile (possibly carbon dioxide). Although it seems unlikely, it is not clear if these water soluble metabolites have any toxicological significance and their likelihood of leaching is currently unknown (Simsiman and Chesters, 1976 and Simsiman et al, 1976).

Since diquat is essentially immobile in various soil substrates, the leaching potential of diquat, and its potential impact on ground water when used for aquatic plant control is significantly reduced due to its irreversible sorption to sediment and soil. Although very unlikely, equilibrium shifts that could release diquat from the bound state and cause it to be reactivated and possibly leach cannot be ruled out particularly if extensive liming or fertilization of treated soils occurs (Simisman et al, 1976). However, while a release of bound diquat cannot be ruled out, Dyson and Takacs (2000) have noted that due to irreversible adsorption of diquat at concentrations of at least 250 ppm on soil/sediment, it is unlikely that bound diquat would ever be reactivated. In further support of this observation, it was noted that diquat has been used for more than 30 years in over 100 countries for every conceivable agronomic and aquatic practice and soil/sediment type, without a single case of adsorbed diquat residues being released or reactivated.

There has been very little work done on the impact of diquat on surface waters, but after treatment of the Guntersville Reservoir (TN) with 0.4 ppm diquat and 0.4 ppm Komeen®, diquat was not detected at the six water treatment plants that service this reservoir (Rogers et al, 1992). It has been noted from modeling experiments that diquat can be used without exceeding the MCLG (0.02 ppm c.e.) at 2 gallons formulation per acre providing that a setback distance of 400 feet for reservoirs and 1,600 feet for rivers is observed. If this setback distance was not attainable, it would be necessary to wait 8 hours before using water from a treated reservoir or 48 hours before using water from a treated river (Waterborne, 1995). Somewhat different values were reported by Singh and Williams (2000), who reported that a 400 foot setback distance was necessary in large reservoirs with a treatment area of 0.32 acres but a 1,400 foot setback distance was necessary for large reservoirs with a treatment area of 50.2 acres. Singh and Williams also recommended water use restrictions of 1 and 20 hours, respectively if these setback distances cannot be observed during the treatment of reservoirs. Furthermore, Singh and Williams (2000) recommended that an upstream setback distance of 200 to 400 feet be observed for canals based on longitudinal and horizontal dispersions of 149.6 and 0.05 feet^2/sec or 3.0 and 0.0 feet^2/sec, respectively.

Table 3.7: Diquat Acid Adsorption/Desorption Constants

Soil/sediment type	**% organic carbon**	**CEC (meq/g)**	**% Clay**	**% Silt**	**% Sand**	**% Gravel**	**Kd_{ads}^{1} (L/Kg)**	**Koc_{ads} (L/Kg)**	**Freundlich K_{ads} (L/Kg)**	**Reference**
Sand sediment	n.r.^{2}	n.r.^{2}	n.r.^{2}	n.r.^{2}	n.r.^{2}	n.r.^{2}	n.r.^{2}	n.r.^{2}	15	EPA, RED, 1995
Sandy soil	n.r.^{2}	n.r.^{2}	n.r.^{2}	n.r.^{2}	n.r.^{2}	n.r.^{2}	n.r.^{2}	n.r.^{2}	36	EPA, RED, 1995
Sandy soil	n.r.^{2}	n.r.^{2}	n.r.^{2}	n.r.^{2}	n.r.^{2}	n.r.^{2}	n.r.^{2}	n.r.^{2}	42	EPA, RED, 1995
Sandy loam. Sandy clay loam soil	n.r.^{2}	n.r.^{2}	n.r.^{2}	n.r.^{2}	n.r.^{2}	n.r.^{2}	n.r.^{2}	n.r.^{2}	1,882-10,740	EPA, RED, 1995
Peat soil	31*	n.r.^{1}	n.r.^{2}	n.r.^{2}	n.r.^{2}	n.r.^{2}	17,000	54,000**	n.r.^{2}	Waterborne, 1995
California Sand gravel	1.0	n.r.^{2}	0	1.4	46.7	51.9	30-1,000	3x10^3 to 1x10^4**	n.r.^{2}	Waterborne, 1995
Louisiana Silty clay loam	2.0	n.r.^{2}	35.5	51.0	13.5	12	10,000-60,000	5 x10^4 to 3x10^6**	n.r.^{2}	Waterborne, 1995
Florida Sand	2.0	n.r.^{2}	0.0	0.0	88.0	12.0	1,000-10,000	5 x10^4 to 5x10^5**	n.r.^{2}	Waterborne, 1995
Wisconsin Sand gravel	1.0.	n.r.^{2}	0.0	1.0	52.0	47.0	30-1,000	3x10^3 to 1x10^4**	n.r.^{2}	Waterborne, 1995
New York Sand gravel	1.0	n.r.^{2}	N/A	N/A	N/A	N/A	30-1,000	3x10^3 to 1x10^4**	n.r.^{2}	Waterborne, 1995
Various unspecified Soil/Sediments	n.r.^{2}	n.r.^{2}	n.r.^{2}	n.r.^{2}	n.r.^{2}	n.r.^{2}	1,200-92,000 (average 27,000)	n.r.^{2}	n.r.^{2}	Ferguson et al, 1994 in Waterborne, 1995
Montmorillonite Clay	100	0.847	n.r.^{2}	n.r.^{2}	n.r.^{2}	n.r.^{2}	708-2,863	n.r.^{2}	n.r.^{2}	Weber et al, 1965 in Reinert and Rogers, 1987

Table 3.7: Diquat Acid Adsorption/Desorption Constants (continued)

Soil/sediment type	% organic carbon	CEC (meq/g)	% Clay	% Silt	% Sand	% Gravel	Kd_{ads}[1] (L/Kg)	Koc_{ads} (L/Kg)	Freundlich K_{ads} (L/Kg)	Reference
Kaolinite Clay	100	0.051	n.r.[2]	n.r.[2]	n.r.[2]	n.r.[2]	21-57	n.r.[2]	n.r.[2]	Weber et al, 1965 in Reinert and Rogers, 1987
Lake Mendota Sediment	8.4	0.24	39.1	57.5	3.3	n.r.[2]	n.r.[2]	205-457	n.r.[2]	Simsiman and Chesters, 1976 in Reinert and Rogers, 1987
QSAR value based on structure and physical characteristics of diquat	n.r.[2]	n.r.[2]	n.r.[2]	n.r.[2]	n.r.[2]	n.r.[2]	n.r.[2]	691	n.r.[2]	Hunter et al, 1984 in Reinert and Rogers, 1987
SCS/ARS/CES estimate	n.r.[2]	n.r.[2]	n.r.[2]	n.r.[2]	n.r.[2]	n.r.[2]	n.r.[2]	1×10^6	n.r.[2]	Wauchope et al 1992
Typical Values for sediments/soils	n.r.[2]	n.r.[2]	n.r.[2]	n.r.[2]	n.r.[2]	n.r.[2]	n.r.[2]	3.2×10^4 7.9×10^6	n.r.[2]	Ritter et al, 2000
Typical partition coefficients in Southeastern Canal Sediments	n.r.[2]	n.r.[2]	n.r.[2]	n.r.[2]	n.r.[2]	n.r.[2]	5,000	n.r.[2]	n.r.[2]	Ritter et al, 2000
Suspended New Zealand Sediment	<1.0	0.058-0.094 SAC	1-10	n.r.[2]	n.r.[2]	n.r.[2]	113,000	n.r.[2]	n.r.[2]	Bowmer, 1982

* % organic carbon calculated by this reviewer from Freundlich K and Koc values presented by Waterborne (1995) using the equation % organic carbon = $(K_d / K_{oc}) \times 100$.

**K_{oc} values calculated by this reviewer using the equation $K_{oc} = (K_d / \text{\% organic carbon}) \times 100$. See text.

[1] ads = adsorption, des = desorption

[2] n.r. = data not reported

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Diquat

Volume 4, Section 4

ENVIRONMENTAL EFFECTS

209 PAGES

TABLE OF CONTENTS

4.0 ENVIRONMENTAL EFFECTS ASSESSMENT – DIQUAT

Executive Summary: The information contained in this report was compiled from studies submitted to EPA by the sponsor, data found on various EPA web-sites, and the open literature on the toxicity of diquat dibromide salt and diquat cation. Data collected included acute toxicity for the standard test species of microbes (algae and protists), plants, fish, free-swimming invertebrates and benthic (sediment-dwelling) invertebrates. Chronic toxicity data consists of early life-stage studies for fish and amphibians and life-cycle studies for free swimming invertebrates. No chronic toxicity data was collected for algae, plants or sediment organisms. Although chronic data was in short supply, since the half-life of diquat is very short (<1 to ~4 days), chronic risk assessments were not believed to be necessary to determine safety of diquat for the biota. However, acute to chronic toxicity ratios based on the LC50/MATC and the LC50/NOEC were calculated to determine these ratios and their variability. An estimate of the chronic Maximum Acceptable Toxic Concentration (MATC) was made based on the acute/chronic toxicity ratio for animal species, which have both acute and chronic data available. Additional data were collected on species other than the standard test species to supplement the data further. A risk assessment was conducted based on the procedures outlined in Urban and Cook (1986) and the procedures outlined in Campbell et al (2000). Urban and Cook state that if acute risk quotients (RQs) are less than 0.1 and chronic risk quotients are less than 1.0, the biota should be safe from the toxic effects of the tested pesticide with assurance that 95% of the tested biota will be protected. These values are respectively termed the acute and chronic level of concern. The acute RQ is defined as the short-term expected environmental concentration (EEC) divided by the acute EC50; and the chronic RQ is defined as the long-term EEC divided by the chronic NOEC or MATC.

Campbell et al (2000) did not use safety factors as described in Urban and Cook (1986) but felt that if the EEC is not higher than the LC50 in more than 90% of the cases that safety to the biota should be sufficient. Judgement must be used to determine the best approach based on past experience and the need to provide extra protection to threatened, endangered or sensitive species, and species that are key elements in the food-web.

Acute risk quotients are defined as the four day geometric mean of the Expected Environmental Effects Concentration (EEC) divided by the concentration of the herbicide that will cause mortality in 50% of animals exposed in a standardized acute toxicity test (EC50 or LC50). These values are calculated from the most typical initial concentration of diquat (0.224 to 0.37 ppm c.e.); half-life is considered to be an important element in calculating the EEC. For diquat, the water dissipation half-life ranges from <1.0 to about 4 days in most aquatic ecosystems. For EPA's worst case scenario [an impounded farm pond 200-cm deep treated at the maximum use rate (4.48 kg c.e./ha)] the half-life can be considered to be 0.75 days (Shaw et al, 1995). The most typical concentration (EEC) at zero time for diquat is considered to be 0.224 ppm c.e. However, the short term EEC for a typical exposure of 4 days with a half-life of 0.75 days would be the geometric mean of the concentrations occurring during this exposure period. These values have been reported by Shaw et al (1995) to be 0.0801 ppm c.e., but were calculated here to be 0.059 ppm c.e. These values were validated by Fujie (1988) with concentrations averaging less than 0.060 ppm c.e. for all sampling times of 1 day or greater. Chronic Risk Quotients are defined as the 28-day geometric mean for the Expected Environmental effects concentration (EEC) divided by the no observed effect concentration or maximum allowable toxic concentration (NOEC or MATC) for animals after exposure in a standardized chronic toxicity test These values are calculated from typical day-0 concentrations of diquat (0.224 ppm c.e.) and a typical half-life of 0.75 days. The long-term EEC for diquat is estimated to be 0.0086 mg/L based on the

geometric mean of the concentrations experienced during a 28-day exposure to diquat. Therefore, the chronic EEC is estimated as being 0.0086 ppm c.e These values have also been verified by Fujie (1988) who found mean concentration averaging less than 0.009 ppm c.e. for all sampling times of 5 days or greater. Similar observations have been made by (Sewell, 1969), Frank and Comes (1967), Langeland and Warner, 1986 and Serdar (1997) for lakes and ponds in New York, Colorado, North Carolina, and Washington, respectively.

Diquat products affect various species of algae and protists. Various species of algae and protozoans found in the water column are affected by concentrations of diquat ranging from 0.30 ppm to 30 ppm c.e. Similar treatments have induced algal blooms of green algae in the laboratory (Melendez et al, 1993 and Pratt et al, 1990) as well as Chara *(Johnson, 1962),* Mougeotia *(Cook, 1977) and other algae (Inabinet, 1976) in the field. It has been observed in laboratory microcosms that species richness of protozoan populations is adversely affected by diquat concentrations of >0.3 ppm c.e. and that higher concentrations of diquat produced progressively greater lack of diversity in exposed protist populations. These changes in microbial population and function can persist for periods longer than three weeks in absence of sediment, but if sediment is present, these disruptions in microbial population structure and function do not appear to last longer than about 1-week (Pratt, 1990).*

*Although efficacy is claimed on the label for only two genera of algae (*Spirogyra *spp. and* Pithophora *spp.), diquat is sufficiently toxic to be considered a high risk herbicide to most species of algae (Peterson et al, 1997). When applied at concentrations close to the maximum use rate in two feet of water (0.75 ppm c.e.), there is 100% inhibition of growth of all of the freshwater blue-green alga, green and diatom species tested (Peterson et al, 1997 and Peterson et al, 1994). An exception may be marine green algae and diatoms with EC50s that are quite high (6.48 to 4.3 ppm c.e. for marine green algae and 3.24 ppm c.e. for marine diatoms) (Walsh, 1972). Under the risk assessment scheme proposed by Peterson et al (1994), all of the species tested at ~0.75 ppm are at very high risk with more than 50% inhibition of growth and a risk quotient (EEC/EC50 >1.0) for diquat. If one factor in the dissipation of diquat for a typical pond treated at 0.224 ppm c.e., most species of algae will not be adversely impacted by the 4-day geometric mean EEC of diquat. With the exception of* Lyngbya wollei *and* Peridinium cinctum*, the algae species tested did not yield risk quotients that were greater than the level of concern for aquatic plants (1.0) in typical standard tests for algal growth on exposure to diquat. Confirming the data presented in this risk assessment, the Water Investigations Branch (1977 in Shearer and Halter, 1980) found that mixtures of 0.5 ppm diquat plus 0.5 ppm paraquat did not reduce the standing crop of phytoplankton; but stimulated growth 3 weeks after treatment of a British Columbia boat marina. Similar lack of effects or transitory effects was observed for a variety of phytoplankton, and other algae including* Anabaena *spp.*, Staurastra *spp. and* Vaucheria dicotoma *(Butler, 1977, Daniel, 1972, Tatum and Blackburn, 1962 and Fish, 1966 in Shearer and Halter, 1980). There is some evidence that there can be algal blooms with Chara (Johnson, 1962) and* Mougeotia *(Cook, 1977) and other algae (Inabinet, 1976) probably due to releases of nitrogen and phosphate when a water body is treated with diquat for control of* Egeria densa *and other macrophytes and filamentous algae.* Chara *spp.*, which *is usually not a problem prior to the removal of macrophytes, may flourish and become the dominant species and it cannot be controlled with typical treatments of 0.25 ppm diquat Some authors have reported that duckweed (Hulbert, 1987) and Eurasian watermilfoil (Water Investigation Branch, 1977 in Shearer and Halter, 1980) were inadequately controlled by standard applications of diquat because control was achieved for only 1 and 2 months, respectively. There may also be shifts in dominant species to those which are less susceptible to diquat, including* Chara *spp.,* Mougeotia *spp.,* Nymphaea *spp and*

Nuphar *spp. The levels of diquat necessary to control* Myriophyllum spicatum *will also kill most other common aquatic plants.*

Diquat is toxic to indicator species of aquatic macrophytes at low concentrations. The representative species in the laboratory are Lemna minor *and* Spirodela polyrhza *and the toxicities (EC50s) of diquat are typically 0.001 to 0.01 ppm c.e. for Lemna minor and 0.00075 to 0.01 ppm c.e. for* Spirodela polyrhyza. *Since use rates may be as high as 0.75 ppm c.e., these macrophytes would be controlled under typical field situations Results from field and semi-field studies indicate that Eurasian watermilfoil (*Myriophyllum spicatum*), coontail (*Ceratophyllum demersum*), American waterweed (*Elodea canadensis*), Sago pondweed (*Potamogeton pectinatus*), curlyleaf pondweed (*P. crispus*), Richardson's pondweed (*P. richardsonii*), duckweed (Lemna minor),* Callitriche *spp., Hydrilla (*Hydrilla verticillata*), waterlettuce, (*Pistia stratiotes*), waterhyacinth (*Eichhornia crassipes*),* Salvinia molesta *and Filamentous algae (*Mougeotia *spp. and* Zygnema *spp.) are controlled by diquat at concentrations between 0.25 and 0.5 ppm. All of these species are "controlled" by diquat. However, the Reward® LA label does not recommend the use of diquat for the control of Richardson's pondweed and no efficacy is claimed for the control of* Callitriche *spp.,* Mougeotia *and* Zygnema *spp. Eurasian watermilfoil, curlyleaf pondweed and duckweed are controlled by diquat, but use of diquat in the northwest appears to control Eurasian watermilfoil for only about 2 months, curlyleaf pondweed for only about 1.5 months and duckweed for only about 1 month (Johnson, 1962, Hulbert, 1987, and Water Investigation Branch, 1977 in Shearer and Halter, 1980).*

Diquat has a toxicity to algae that is high enough so that they may be harmed by the maximum use rate of 0.75 ppm c.e. The only tested green algae which may be at risk are Selenastrum capricornutum *(EC50 0.019 to 0.492 ppm),* Chlorella vulgaris *(LC50 = 0.395 to >2.94 ppm c.e) and* Scenedesmus quadricauda *(EC50 = 0.026-0.047 ppm c.e.) However, if the four day averaged EEC (0.059 ppm c.e.) for EPA's worst treatment scenario of 0.224 ppm c.e in a farm pond, only* Selenastrum capricornutum *may be at risk. There is some evidence that unicellular green algae* (Oocystis, Characium, Ankistrodesmus, Cosmarium and Staurastrum*) may become the dominant algae species when the treatment levels are 0.3 to 10 ppm c.e.; but this data is from laboratory microcosms which may or may not reflect their likelihood of domination in a field situation. Species of blue-green algae are sensitive enough to be controlled by the maximum use rate of 0.75 ppm c.e. The EC50 for* Anabaena flos-aquae *varies from 0.042 ppm to 0.138 ppm c.e. and the tested species of* Oscillatoria *spp. and* Pseudoanabaena *spp yield EC50s in the range of 0.063 ppm to 0.082 ppm. However, while these high doses may provide temporary control and put these blue-green algae species at risk (RQ = >17), it is likely that the 4-day average EEC for EPA's worst case scenario would not place blue-green algae in a high risk category (RQ = 0.059 ppm/0.082ppm = 0.71). This does not mean that no species of blue-green algae would be at risk since* Anabaena inaequalis *has an EC50 at less than 24 hours that is approximately 0.005 ppm c.e. Diatoms are typically similar to blue-green algae in their toxicity response to diquat, yielding LC50s that range around 0.1ppm for* Navicula pelliculosa, Cyclotella meneghiana *and* Nitzchia *spp. As discussed above, blue-green algae are only temporarily reduced in number and often participate in algal blooms. Since concentrations of diquat at 0.3 to 10 ppm apparently decrease the species richness of protists in a laboratory microcosm, it is apparent that diquat affects the population structure of microbes (Melendez et al, 1993) in absence of sediment; but if sediment is present, this effect is only transitory and typically lasts less than a week (Pratt et al, 1990).*

Although the risk assessment scheme promoted by Peterson et al (1994) indicates a high risk for all aquatic algae and plants at the maximum use rate, use of EPA's worst case scenario allows *for a four-day average EEC to be considered. When this four-day average EEC (0.059 ppm c.e.) is considered, the risk quotient is usually less than the level of concern of 1.0 for algae although most macrophytes appear to be at risk. Notable exceptions in the algae are the diatom* Ochromonas danica *(EC50 = 0.022 ppm c.e) and the pyrophyte* Peridinium cinctum *(EC50 = 0.001 ppm c.e.) which will yield risk quotients that are higher than the level of concern. For other species the risk is less clear since a range of EC50s is available, some at which are higher than the EEC, some of which are lower than the EEC and some of which are not significantly different from the EEC. Eurasian watermilfoil, (EC50 = 0.046 ppm c.e.), duckweed EC50 <0.02 ppm c.e) Elodea canadensis (EC50 = 0.023 ppm c.e.), floating leaf pondweed (EC50 = 0.0072 ppm) and water fern (EC50= 0.023 ppm c.e.) are clearly macrophyte species that are at high risk when exposed to diquat (RQ = >1.0). However, these plants should not be considered in the general risk assessment since the label specifies them as targets for diquat control.*

- ***Summary of Diquat Effects on Aquatic Animals***

  *For fish, the standard bioassay time is 96 hours. Diquat has a high laboratory acute toxicity to fish (96 hour LC50 = 0.54 to 156 ppm c.e. for striped bass sac-fry and adult mosquito fish, respectively). If the EPA's typical worst case scenario of 0.224 ppm is used, no significant mortality should occur since the lowest defined LC50 is much greater than the EEC of 0.224 ppm c.e. However, if the 4-day geometric mean of 0.059 ppm c.e. is used as the EEC, virtually no fish are affected at this concentration and the risk quotient is less than or approximately equal to the level of concern of 0.1 (RQ = 0.11 = 0.059 ppm/0.54 ppm) Only one test in 78 reviewed exceeds the level of concern (and in this case just barely). Striped bass are known to be particularly sensitive to many pesticides especially their neonates. Other species that are known to be particularly sensitive to pesticides include the walleye (*Stizostedion vitreu*) (lowest LC50 = 0.75 ppm c.e), smallmouth bass (*Micropterus dolomieu*) (lowest LC50 = 1.5 ppm c.e.) and the largemouth bass (*Micropterus salmoides*)(lowest LC50 = >1.62 ppm). In all of these cases, the risk quotient is lower than the level of concern of 0.10. Even the salmonids, which are of special concern as a game fish, aesthetically, and as representatives of an endangered group, are not particularly sensitive to diquat; the lowest LC50s are 6.1, 17.77, 20.5 and 30 ppm c.e. for rainbow trout fingerlings, brown trout fingerlings, and Coho salmon fingerlings respectively.*

  Limited field data with sentinel organisms (caged fish) and net capture population surveys indicate that diquat lacks acute environmental toxicity to fish and amphibians when applied at labeled rates (Bain and Boltz, 1992; Olaleye et al, 1993; Johnson, 1962; Cooke, 1977, and Gilderhaus, 1967).

  Coho salmon smolts may be affected by concentrations of diquat that are similar to the field rate. Shearer and Halter (1980 citing Lorz et al, 1979) indicate that salmon exposed to 5.0 to 20 ppm c.e diquat for 144 hours experienced 15 to 87% mortality in a subsequent saltwater challenge test Lorz (1979) found that exposure of Coho salmon to 0.5 to 3.0 ppm diquat did not produce subsequent mortality in a saltwater challenge test and that these concentrations did not affect the level of gill ATP-ase. However, all dosages of diquat down to 0.5 ppm interfered with the ability of Coho smolts to migrate downstream and that this failure to migrate was dosage dependent. The highest concentration of diquat (3.0 ppm) produced histopathological effects on the eyes, kidney, gills, and liver Similar experiments with Chinook salmon at concentrations ranging from 0.125 ppm to 0.5 ppm c.e. produced no mortality in subsequent seawater challenge tests and all exposed fish were similar to controls in their ability to osmoregulate in seawater (Merrill, 1997). But, the interference of Coho *smolts' ability to migrate downstream indicates that diquat may interfere with the parr to smolt metamorphosis in andromonous fish species.*

In general, fish and shellfish do not bioaccumulate diquat. When bluegill were exposed to diquat at concentrations of ~1.0 ppm c.e. for 14 days, the maximum measured concentration in whole fish was 1.30 ppm c.e. upon removal of the fish from diquat exposure, 50 % of the residues were eliminated from the fish in three days (Hammer, et al, 1987). In field studies to measure the bioconcentration potential of diquat in catfish and Talapia *spp., no bioconcentration of diquat was seen in edible catfish tissue and only 0.3 ppm diquat was found in the edible tissue of Talapia and this depurated to 0.1 ppm within three days of application (Fujie, 1987). In channel catfish exposed to 0.005 to 0.020 ppm, diquat did not bioaccumulate in any of the tissues. Most of the residue in white muscle, skin and kidney was unchanged diquat while the residues in liver and bile were primarily unidentified metabolites (Schultz et al, 1995).*

For invertebrates the standard bioassay times vary from species to species. For most species, the standard bioassay time is 96 hours. However, for Daphnia*, the standard bioassay time is 48 hours. In all cases the standard bioassay time is given for both general toxicity purposes and in order to standardize the risk assessment Diquat is not highly toxic to most invertebrates. For example, 48-hour LC50s range from 0.12 ppm c.e for the amphipod* (Hyalella azteca) *to >100 ppm c.e. for damselfly larvae, dragonfly larvae* (Enallagma *spp. and* Libellula *spp.), and bloodworm larvae (Tendipedinae); the 96-hour LC50 ranges from 0.058 ppm c.e. for the amphipod* (Hyalella azteca) *to 141 ppm c.e. for the Eastern oyster* (Crassostrea virginica)*. Formal assessment indicates low risk to all species except for the amphipod (Hyalella) (RQ = 0.025), water fleas* (Daphnia *spp. – lowest 48 hour LC50 = 0.324 ppm c.e.) and possibly pocket shrimp* (Mysidopsis bahia *– 96-hourLC50 = 0.42 ppm c.e.) and apple snail* (Pomacea paluosa *– LC50 = 0.34 ppm c.e.) To conduct a standard risk assessment, the EPA's worst case scenario, where the application rate is 0.224 ppm c.e. and the 48-hour and 96-hour average EEC is 0.10 ppm c.e and 0.059 ppm c.e., respectively, were used. In these cases the Risk Quotient is greater than the level of concern at 0.1 for the amphipod (RQ = 0.059 ppm/0.048 ppm = 1.2), water flea (0.10 ppm/0.162 ppm c.e. = 0.62), pocket shrimp (RQ = 0.059 ppm c.e./0.422 ppm c.e. = 0.14 and the apple snail (RQ = 0.059 ppm c.e./0.34 ppm c.e. = 0.17). Current guidance indicates that treating diquat as a restricted use herbicide should allow for an additional level of safety where the risk quotient level of concern can be raised to 0.5. This approach would make the use of diquat acceptable when pocket shrimp or apple snails are likely to be impacted The pocket shrimp is an estuarine species and may not be affected; one can reasonably expect that extensive dilution of water containing diquat will occur before it reaches an estuary containing pocket shrimp. The risk quotients for the amphipod and water flea greatly exceed the level of concern, so these species may be at risk in exposures to diquat. In support of these observations, field work with the amphipod* (Hyalella azteca) *and various cladoerans indicates that treatment will significantly reduce the number of* Hyalella azteca *when they are exposed to standard applications of diquat in the field (Wilson, 1967) Hilsenhoff (1966) found that reduction in habitat due to elimination of* Elodea canadensis *may be the reason for reduction in numbers of* Hyalella azteca *in the field (Hilsenhoff, 1966) rather than direct toxicity of diquat. Therefore, the inclusion of artificial substrate like conservation webbing may prevent these population reductions (Berry et al, 1975). Field work with cladoerans* (Daphnia *spp.) at concentrations slightly higher than the maximum concentration that may be encountered in the field (1.0 ppm) has reduced population numbers and shifted the population peak to a time after diquat has degraded and is no longer present at toxic concentrations (Gilderhaus,* *1967). Another cladoeran* (Simocephalus vetulus) *was adversely impacted at concentrations that are slightly higher than the maximum that may be encountered in the field. This species disappeared for 64 days after treatment with 1.0 ppm diquat (Shearer and Halter, 1980 cite Newbold, 1975) No other species of invertebrate were found to be affected by exposure to diquat in the field (Gilderhaus, 1967, Berry et al, 1975, Hilsenhoff, 1966 and Bond, 1967).*

The chronic toxicity of diquat has not been extensively evaluated in studies that would currently fulfill EPA study guidelines. However, a couple of early life-stage studies and one life-cycle study have been conducted with this test substance. Although considerable additional work has been done with killifish and amphibians, it is not readily interpretable since the lowest concentration used in these studies in addition to causing developmental abnormalities also caused all the test organisms to die, or the study was insufficient in duration to be considered as a chronic exposure test. In fish the MATC was >1.0, 0.2 and <1.4 ppm c.e. in channel catfish (Ictalurus punctatus)*, fathead minnow* (Pimephales promelas) *and rainbow trout* (Onchorhynchus mykiss)*, respectively The LC50s for chronic tests were >0.65 and 2.9 ppm for fathead minnow and rainbow trout, but was not determined for channel catfish. The tests on channel catfish and fathead minnow measured reduction in weight as compared to the controls and the test on rainbow trout measured survival. The length of these tests ranged from 21 days to ~34 days. Since the field dissipation half-life is short, it is unlikely that fish will encounter concentrations of diquat high enough to affect them chronically. In the worst case scenario typically used by the EPA, it is estimated that a 0.224 ppm application of diquat would dissipate to a time-weighted average of 0.012, 0.0087 and 0.0071 ppm c.e. in 21, 28 and 35 days, respectively. These levels of diquat were validated in field studies conducted by Fujie (1988), who found that diquat applied at rates similar to that of EPA worst case scenario dissipated to an average of <0.007 ppm c.e. in approximately seven days. Similar finding were seen by Frank and Comes (1967), Seawall (1969), Gilderhaus (1967), Rogers et al (1992) and Sedor (1997) who applied diquat at concentrations up to or slightly higher than the current maximum use rate to lakes or ponds in Florida, Colorado, New York, Wisconsin, Alabama and Washington, respectively. Since none of these expected concentrations are as high as the estimated MATCs it is not likely that chronic effects on fish would be observed in the field. There is not enough data to predict the MATC for other species. However, the acute to chronic ratio for fathead minnow is 38 (LC50/MATC = 7.6ppm c.e./0.2 ppm c.e.). The most sensitive species of fish (striped bass) would have a predicted MATC of 0.014 ppm c.e (0.54 ppm c.e./38) and this value is much greater than the average EEC predicted 28 days after treatment of a water body (EEC = 0.0087 ppm c.e). Therefore, even the most sensitive species of fish is unlikely to be adversely impacted by chronic exposure to diquat.*

The experimental chronic toxicity (MATC) is 0.045 ppm c.e. for Daphnia magna. *Under EPA's worst case scenario (an initial application of 0.224 ppm c.e.), the concentration of diquat would dissipate to 0.012, 0.0087 and 0.0071 ppm c.e. in 21, 28 or 35 days. Therefore, it is unlikely that this species would be exposed to concentrations that would impact its growth and reproduction (RQ = EEC/MATC = 0.012 ppm c.e./0.045 ppm = 0.45 in 21 days). There is not enough data to predict the MATC for other species. However, the acute to chronic ratio for* Daphnia magna *is 19 (LC50/MATC = 0.87 ppm c.e./0.045 ppm c.e.)(it is noteworthy that this ratio is similar to that generated for fathead minnow). The most sensitive species of invertebrate* (Hyalella azteca) *would have a predicted MATC of 0.0071 or 0.0025 ppm c.e. (0.14 ppm/19 or 0.048 ppm c.e./19) based on the 48-hour LC50 or 96-hour LC50, respectively These MATC values are high enough to yield a chronic risk quotient for* Hyalella azteca that is *higher than the chronic level of concern of 1.0 (RQ = 0.0087 ppm* *c.e./0.003 ppm c.e. =2.9) Therefore, the most sensitive invertebrate species tested is likely to be adversely impacted from chronic exposure to diquat. Of the tested species, only* Hyalella azteca *appears to be adversely impacted by chronic exposure to diquat. The predicted MATCs for the apple snail and pocket shrimp, 0.018 ppm c.e. (0.343 ppm c.e./19) and 0.022 ppm c.e. (0.422 ppm c.e./19), are high enough that the level of concern for the chronic risk quotient will not be exceeded and therefore, these two relatively sensitive species should be safe when exposed chronically to diquat . As discussed above, field studies have been conducted to show that* Hyalella azteca *and certain cladoerans are adversely impacted by exposure to diquat, but it is unclear if these field effects are due solely to acute toxicity, or if there is a chronic component. The fact that juvenile daphnia appear to be unable to mature to the adult form when exposed to 1.0 ppm in a static system containing sediment and that the effects of exposure of* Simocephalus vetulus *in the field to 1.0 ppm seem to last longer than two months presents good evidence for chronic effect.*

There have been arguments made that the presence of sediment reduces the toxicity of diquat by binding it tightly and making it biologically unavailable (Simsiman et al, 1976). It is apparent that large amounts diquat (250 ppm c.e.) bound to the sediment are undetectable in the water column (Hiltibran, 1972). It has also been shown that these high sediment concentrations are not biologically available to plants growing in contaminated sediment (Coats et al, 1967 and Daniel, 1972) Similarly, it is apparent that the presence of sediment can reduce the toxicity of diquat to the more sensitive benthic organisms. For example, in absence of sediment the 96-hour LC50 to Hyalella azteca *is 0.048 ppm. However, if sediment is added to the system, this 96-hour LC50 rises to 6.8 ppm and thus might spare this very sensitive species from both the acute and chronic effects of diquat.*

Conclusion: Except for the inability of Coho smolts to migrate downstream at concentrations within label rates, diquat will generally have no significant acute or chronic impact on fish when applied rates recommended on the label are used. However, striped bass sac-fry, smallmouth bass sac-fry, and similarly sensitive species may be at risk after exposure to diquat. It is also believed, that except for amphipods like* Hyalella azteca *and cladoerans like* Daphnia *spp. and* Simocephalus *spp., diquat should not have significant impact on aquatic invertebrates typically present in treated water bodies. There appears to be virtually no tendency for diquat to bioaccumulate in fish or invertebrates when exposed from contact with water treated with diquat. While crop plants do not appear to accumulate diquat in their tissues after irrigation with water that has been treated with diquat, aquatic plants susceptible to diquat may extensively accumulate diquat, which is released and bound rapidly to the sediment after the susceptible treated plants die. Furthermore, the concentrations listed on the label (0.5 to 2 gallons formulation per acre) will control the aquatic macrophytes listed on the label including, but not limited to, Eurasian watermilfoil* (Myriophyllum spicatum*), Pondweeds (*Potamogeton spp.*), waterlettuce (*Pistia stratiotes*), waterhyacinth (*Eichhornia crassipes*), duckweed (*Lemna *spp.), American waterweed (*Elodea canadensis*),* Salvinia molesta *and coontail (*Ceratophyllum demersum*). Concentrations in the water column of 0.25 ppm are generally effective in the control of waterweeds and some species of filamentous algae. However, treatment of both Lake Steilacoom and Gravelly Lake at concentrations of ~0.15 ppm appears to be effective in controlling Elodea spp. and pondweeds. Some concern has been expressed that species that have not been pests in the past may become dominant after treatment with diquat due to their resistance to diquat, lack of competition with other weeds and algae or lack of long-term effectiveness. These species may include* Chara *spp.* Mougeotia *spp.,* Nuphar *spp. and* Nymphaea *spp.* species. *Also Richardson's pondweed

(**Potamogeton richardsonii*****) may not be effectively controlled and duckweed, curlyleaf pondweed, and Eurasian watermilfoil may not be controlled for sufficient periods of time for this relatively expensive aquatic herbicide to be of maximum utility. Diquat should not be used to control species of aquatic weeds that are not specified on the label, except as provided by FIFRA Section 2(ee). Diquat has limited effectiveness on some species of filamentous algae, but is not generally used to control most algal species. Although laboratory data indicates that diquat may be toxic to the more sensitive species of crustacean invertebrates (*****Hyalella, azteca***** and cladoerans), it is believed that other species of crustacean invertebrate will be substituted by fish as part of their diet so that there should be minimal impact to fish that typically use these species of invertebrates for food.***

4.01 Objective

The purpose of this section is to update the environmental toxicity data and to use this data to assess the potential risks to wildlife and the environment from using diquat products. When wildlife is discussed, the organisms referred to include aquatic plants and animals, terrestrial plants and animals, and microorganisms including algae, bacteria, and fungi.

4.0.2. Study Approach

4.0.2.1 Information Compilation

In order to collect appropriate information regarding wildlife toxicology, several sources of information were used. As a primary and definitive source of data, reports submitted to the EPA Environmental Effects Branch by the registrant Zeneca® Agricultural Products to support the registrations and re-registration of diquat products were used. These submittals are considered to be definitive sources on the wildlife toxicology of diquat because the tests are conducted in an agreed upon design with agreed upon organisms. These organisms are considered to be good representatives or good surrogates for plants and animals that are highly sensitive. Other sources of acute and chronic toxicity data include literature searches with the Dialog Online Database for refereed journal articles and compilations of data in the form of literature reviews (Shearer & Halter, 1980; Ecology, 1982, 1989, 1991/1992; and EPA RED, 1995). Such literature reviews are a good source of information for older data. Similar compilations of EPA data were also searched such as EPA's Brian Database (1999) and EPA's ECOTOX Database (1999). These are online databases for retrieval of data submitted to support registration (Brian Database, 1999) and data from referred journals used as supplemental material to be used for risk assessment and evaluation (ECOTOX Database, 1999).

The US EPA and Washington's Department of the Ecology (Ecology) use these data for the following evaluations:

- To establish acute toxicity levels of active ingredients to test organisms.
- To compare toxicity information with measured or estimated pesticide residues in the environment in order to assess potential impacts to fish and wildlife.
- To provide data which determine the need for precautionary label statements and permit requirements in order to minimize potential adverse effects to wildlife and aquatic organisms.

- To indicate the need for further laboratory and field studies to support regulatory decisions.

If an adverse impact is noted in the basic data, additional studies are conducted and evaluated to determine the effects of the product on sensitive species and sensitive stages of those species. These studies typically take the form of long term chronic, early life stage, reproductive effects and life-cycle effects. These studies take into account the toxicity of the product and compare that toxicity with expected environmental concentrations. If an adverse impact is noted at levels consistent with environmental concentrations, further “field” or laboratory work is necessary to evaluate the acute and chronic effects on different organisms.

4.0.2.2 Risk Assessment Methodology

Risk assessment is conducted in a manner similar to that described in EPA (1982), Brooks (1973 in Ebasco (1993), Ecology (1980, 1989 and 1991/1992) and in Urban and Cook (1986). For assessment of acute risk, the LC50 is determined for a variety of organisms within a class (fish, aquatic invertebrates, algae, other aquatic and terrestrial plants, birds and mammals). The LC50 is the concentration at which 50% mortality is seen; the LD50 is the “oral” or “dermal” dose at which 50% mortality is seen. The relative toxicity of these values is determined in two ways: 1) The EPA has certain specific descriptive classifications for inter-chemical comparisons only and these classifications do not reflect actual environmental concentrations or hazards to the test species. For an example of these classifications please see Table 1; 2) The Acute LC50 or LD50 is compared with the Expected Environmental Concentration or Expected Environmental Dose (EEC or EED). The Acute Risk Quotient (ARQ) is determined by dividing the Expected Environmental Concentration (4-day geometric mean or other appropriate evaluation of the EEC or EED) by the laboratory measured acute toxicity (4-day LC50, LD50). The ARQ is not based on values obtained for a single species but is based on the most sensitive environmentally relevant species in a specific segment of the biota; e.g. algae, other microbes, macrophytes, fish, free-swimming aquatic invertebrates, or benthic organisms. If the ARQ is <0.1, the evaluated pesticide is generally considered to be safe to that segment of the biota for exposures of short duration. A short duration is generally defined as four or five days. However, many investigators believe that this is an excessively cautious approach and that if 90% of the species will not be affected at the 50% level by concentrations that are found in 90% of the treated water bodies, the product should be judged to be safe to aquatic organisms. Nevertheless, if the 10% of those species affected include endangered local species such as salmonids then that assumption is not dependable.

Similar calculations are used for an assessment of chronic risk. However, chronic risk is based on an exposure period of 7 or more days. Seven days exposure is considered to be a short-term chronic risk. Typically 21 to 90 days exposure is considered to be a long-term chronic risk. Short-term chronic risk involves the exposure of sac-fry to the toxic substance and long-term chronic risk involves the exposure of newly fertilized egg through free swimming and actively growing fry. For invertebrates, the chronic life-cycle test involves exposure of newborns through 21 to 28 days when the maximum number of F1 newborns will have been deposited. Dividing the 28-day EEC by No Observed Effect Concentration (NOEC) or the Maximum Acceptable Toxic Concentration (MATC) determines the Chronic Risk Quotient. The CRQ is not based on the values obtained for a single species but is based on the most sensitive environmentally relevant species in a specific segment of the biota; e.g. algae, other microbes, macrophytes, fish, free-swimming aquatic invertebrates, or benthic organisms. If the CRQ is <1.0, the evaluated pesticide is generally considered to be safe to that segment of the biota for exposures of chronic duration.

To determine how well acute toxicity can predict chronic toxicity, an acute (LC50)/ (chronic MATC) was evaluated for species that had both values available. This ratio was taken regardless of the quality of the data and then the quality was evaluated. If an individual ratio was an extreme outlier, it was discarded for the purposes of assessing the acute/chronic toxicity ratio. If extensive chronic data was not available, the acute to chronic ratio was used to estimate the chronic toxicity for species where the test had not been conducted.

4.1 DIQUAT

*Summary: Two registered products containing diquat are used for controlling aquatic weeds and algae in the state of Washington. Several products are registered for use in Washington including Reward® Landscape and Aquatic Herbicide, Zeneca® Diquat Herbicide, Misty® Weedtrol VF, Aqua Trim® II Vegetation Control and Sea-Cide®. However, only Reward® Landscape and Aquatic Herbicide is labeled for use in public waterways. Reward ® LA is used for the control of Eurasian watermilfoil (*Myriophyllum spicatum*), most species of pondweeds (*Potamogeton *spp.), duckweed (*Lemna *spp.), American waterweed (*Elodea canadensis*), coontail (*Ceratophyllum demersum*), certain species of filamentous algae (*Spirogyra *spp. and *Pithophora *spp.) and certain other macrophytes listed in the label. In order to write a label and determine if these products are safe to the biota, organisms with an extensive history of use in pesticide testing are evaluated by the registrant for their response to acute and chronic exposure. The most sensitive, easily culturable species are selected for testing. The most sensitive stages of any organism are usually those when rapid growth is occurring or the time of reproduction or shortly thereafter when eggs or newborn offspring are present. The most likely exposure route should also be selected to most closely mimic environmental reality.*

There are currently eight aquatic use registered formulations of diquat in the United States. While registering all eight formulations, Washington State is only likely to permit the use of Reward® Landscape and Aquatic Herbicide for use in public waterways. It is the diquat dibromide containing 20% diquat cation equivalence (c.e.). This review directly addresses only this formulation labeled for aquatic use in public waterways and registered for this use by the Washington State Department of Ecology and Washington State Department of Agriculture as of July, 2000. The toxicity of the diquat cation in the diquat dibromide formulation, will be addressed to support the risk assessment since this is the principal form that diquat dibromide is converted to prior to its effect on aquatic weeds and wildlife (EPA RED, 1995). When diquat was originally registered for aquatic use, it became quickly apparent that when adsorbed by fish or wildlife it was eliminated largely unchanged from the organism, although the presence of large amounts of unidentified metabolites in the liver and bile indicates that the liver can metabolize diquat to other presumably less toxic metabolites (Schulz et al, 1995 and Shearer & Halter, 1980). Since the diquat dibromide rapidly dissociates to diquat cation, diquat cation is believed to be the intoxicating agent. A high degree of water hardness decreases the toxicity of diquat cation by as much as ten-fold. Therefore, the toxicity of diquat in soft water is emphasized for the purposes of risk assessment.

The diquat products currently registered for aquatic use in Washington State are as follows:

Reward® Landscape and Aquatic Herbicide -- A liquid product containing 37.3% diquat dibromide (20% diquat cation). Distributed by Syngenta Agricultural Products. Labeled for application in ponds, lakes, reservoirs, marshes, bayous, drainage ditches, canals, streams, rivers, and other slow-moving or quiescent bodies of water (public waterways) for control of aquatic weeds.

Syngenta® Diquat Herbicide -- A liquid product containing 37.3% diquat dibromide (20% diquat cation). Distributed by Syngenta Agricultural Products. Labeled for application to noncrop or non-planted areas on farms including fence lines, farmyards, farm buildings, fuel storage areas, barrier strips, equipment areas, and non-flooded portions of ponds, lakes and drainage ditches on farms. Application to public waterways is not mentioned.

Misty® Weed Trol VF -- A liquid product containing 4.35% diquat dibromide (1.7% diquat cation). Distributed by Amrep/MPL. Labeled for application to ponds, lakes, and drainage ditches where there is little or no out-flow of water, which is totally under of the control of the product's users. Application to public waterways is not mentioned.

Aqua Trim II® Vegetation Control -- A liquid product containing 1.85% diquat dibromide (0.70% diquat cation). Distributed by Check-Mark, Division of DM Resources. Labeled for application to ponds, lakes, and drainage ditches where there is little or no out-flow of water, which is totally under of the control of the product's users. Application to public waterways is not mentioned.

Weedtrine® D Aquatic Herbicide -- A liquid product containing 8.53% diquat dibromide (3.4% diquat cation). Distributed by Laporte Water Technologies. Labeled for application to ponds, lakes, and drainage ditches where there is little or no out-flow of water, which is totally under of the control of the product's users. Application to public waterways is not mentioned.

Quick Kill® Non-Selective Vegetation Killer -- A liquid product containing 1.85% diquat dibromide (0.7% diquat cation). Distributed by Share Corporation. Labeled for application to ponds, lakes, and drainage ditches where there is little or no out-flow of water, which is totally under of the control of the product's users. Application to public waterways is not mentioned.

Eliminator® -- A liquid product containing 1.85% diquat dibromide (0.7% diquat cation). Distributed by Total Solutions. Labeled for application to ponds, lakes, and drainage ditches where there is little or no out-flow of water, which is totally under of the control of the product's users. Application to public waterways is not mentioned.

Sea-Cide® -- A liquid product. Distributed by Drummond American Corporation. No details of product are available.

Although there is a wide range of products available for use, only Reward® Landscape and Aquatic Herbicide appear to be labeled for use in public waterways.

Reward® LA (37.3% diquat dibromide) has a federal aquatic registration and is distributed by Syngenta Agricultural Products. This liquid formulation contains 37.3% diquat dibromide (20% diquat cation). This product has been used in Washington to control American waterweed (*Elodea canadensis)* and pondweeds (*Potamogeton* spp.) except Richardson's pondweed in Lake Steilacoom and Gravelly Lake. It is also labeled for the control of bladderwort (*Urticularia* spp.), coontail (*Ceratophyllum demersum*), naiad (*Najas* spp.), watermilfoil (*Myriophyllum* spp.), Hydrilla (*Hydrilla verticillata*), waterlettuce (*Pistia Stratiotes*), waterhyacinth (*Eichhornia crassipes*), frog's bit (*Limnobium spongia*), Salvinia (*Salvinia* spp.), duckweed (*Lemna* spp.), cattails (*Typha* spp.), and some species of filamentous algae (*Spirogyra* spp. and *Pithophora* spp.)

The other diquat products are labeled for a similar, though not identical, spectrum of weeds. Changes in the labeling of these products may allow for them to be used for the control of aquatic vegetation in public waterways.

4.1.1 Evaluated Organisms and Sensitive Stages (EPA, 1982)

In order to develop the most sensitive risk assessment possible, appropriate species and appropriate life stages must be chosen within each class of organisms. The classes of organisms of interest are microorganisms (bacteria, fungi and algae), macrophytes, fish, aquatic invertebrates, sediment organisms (includes several classes), terrestrial plants, birds, mammals and terrestrial invertebrates (includes several classes). The life stages that are tested are selected for high sensitivity and ease of manipulation. Each class of organism is broken down into appropriate species as indicated in Table 2.

- **Microbes --** Very little work has been conducted on microbes, particularly aquatic bacteria and fungi. Recent work primarily deals with the effects of diquat on various species of algae with some early work (before 1975) conducted to determine the effects of diquat on bacterial species.

- **Algae --** There are four standard species that are typically evaluated in algal toxicity tests. They are *Anabaena flos-aquae* (freshwater blue-green algae), *Selenastrum capricornutum* (freshwater green algae), *Navicula pelliculosa* (freshwater diatom) and *Skeletonema costatum* (marine diatom). These have been selected as the standard species because there is an extensive database on the effects of many pesticides on their growth rate. Additional algal species including *Chlorella vulgaris* (green algae), *Dunaliella tertiolecta* (marine green algae), *Chloroccocum spp*. (marine green algae), *Lyngbya wollei* (blue-green algae), *Microcystis aeruginosa* (unicellular blue-green algae), *Anabaena inaequalis* (filamentous N-fixing blue-green algae), *Oscillatoria* spp. *(*filamentous blue-green algae,), *Pseudoanabaena* spp. (filamentous blue-green algae*), Euglena gracilis* (Euglenophyte), *Cytotella menghiana* (diatom), *Nitzchia spp. (*diatom*), Cryptomonas ovata* (diatom), *Ochromonas danica* (diatom), *Phaeodactylum tricornutum* (marine diatom), *Isochrysis galbana* (marine diatom) and *Chryptomonas ozolini* (diatom) have also been tested with Reward® LA (diquat dibromide products). The endpoint of interest in algal studies is a 50% reduction in log-phase growth after five days of exposure to a static solution (EC50). Field studies normally measure the amount of chlorophyll a or use cell counts at the site as an indicator of population size.

- **Aquatic macrophytes --** For macrophytes, one species (*Lemna minor* or duckweed) is typically used in the laboratory. Duckweed is a standard species with an extensive database on the effects of many pesticides on its growth rate. Results from the standard test species were reported to EPA but it was not the only macrophyte for which data could be found. Data was found for *Lemna minor* (Common duckweed), *Spirodela polyrhiza* (Giant duckweed), *Eichhornia crassipes* (waterhyacinth). *Pistia stratiotes* (waterlettuce), *Potamogeton natans* (floating leaf pondweed), *Elodea canadensis* (American waterweed), *Myriophyllum spicatum* (Eurasian watermilfoil), *Wolfiella floridana* and *Azolla caroliniana* (Azolla). The endpoint of interest in macrophyte studies is a 50% reduction in growth after 7 to14 days of exposure to a static solution containing plants at a very sensitive period in the growth cycle**.** The field studies utilized whatever species were available in whatever growth stage they were in at the time and measured the percent reduction in lake or pond coverage as an endpoint. See Table 2 for a species listing and a summary of the available data**.**

- **Fish toxicity**

  - **Acute toxicity:** The standard species tested in the laboratory include *Onchorhynchus mykiss* (rainbow trout), *Lepomis macrochirus* (bluegill sunfish), *Pimephales promelas* (fathead minnow), *Cyprinodon variegatus* (sheepshead minnow), and *Ictalurus punctatus* (channel catfish). Rainbow trout, bluegill sunfish, fathead minnow, channel catfish and sheepshead minnow were tested with diquat dibromide. Many of the other species tested with diquat would also be found to be acceptable for risk assessment purposes by the EPA. These would include but not be limited to yellow perch (*Perca flavescens*), black bullhead (*Ictalurus melas*), brook trout (*Salvelinus fontinalis*), and Coho salmon (*Onchorhynchus kisutch*). Rainbow trout, brook trout, and Coho salmon are representatives of a cold water species (salmonids) requirement, and the warm water species requirement is fulfilled by tests with bluegill sunfish, fathead minnow, channel catfish, black bullhead (catfish). The marine and estuarine species requirement is fulfilled by tests with the sheepshead minnow. The standard acute LC50 test is run with juvenile fish of a uniform age-class or size, which would typically fall in the class of fingerling (<0.5 grams). These acute toxicity tests are not typically run with smolts, eggs and sac-fry, but in some cases acute toxicity information is provided for these stages. The test is typically run for 96 hours although some of the LC50s may be based on 24, 48 or 120-hour mortality data. The measured endpoint is mortality. The species selected are considered to be representative of a broad sensitivity range and ecological, economic and aesthetic relevance. Other species may also be tested. Those of particular interest based on ecological relevance and/or sensitivity to diquat are striped bass (*Morone saxatilis*), largemouth bass (*Micropterus salmoides*), smallmouth bass (*M. dolomieu*), walleye (*Stizostedion vitreu*), and various salmonids (*Onchorhynchus* spp. and *Salmo* spp.).

  - **Chronic toxicity:** The standard species tested for chronic toxicity are fathead minnows, rainbow trout, and sheepshead minnow, which represent a warm freshwater species, a cold freshwater species, and warm estuarine species. Consensus opinion is that rainbow trout are the most sensitive species in this group. Chronic toxicity tests can be run in the sac-fry stage for at least seven days (standard time period, 28 days). In addition to mortality, the endpoints are growth and sub-lethal behavioral effects. Another study design is the early life-stage test where the endpoints are percent hatch, time to first and last (95%) hatch, swim-up or first-feed, growth and sub-lethal behavioral effects. The effective concentration is the lowest NOEC value obtained for the most sensitive endpoint. In summary reports that are obtained from agencies or registrants, the most sensitive endpoint is often not expressed. In some reporting formats, the effective concentration may be termed the No Observed Effect Concentration (NOEC), Lowest Observed Effect Concentration (LOEC) or the Maximum Allowable Toxic Concentration (MATC), which is the geometric mean of the NOEC and the LOEC. This is a very sensitive test and it often may yield an unacceptably high CRQ when the ARQ indicates a high degree of safety for the more sensitive species in the biota. When endpoints other than survival are used, the MATC is considered to be the concentration of interest for performing risk assessment. For diquat, only the test with fathead minnow and channel catfish was considered adequate for risk assessment purposes. The chronic test with rainbow trout was not considered adequate because a NOEC was not seen for a test where survival was the final end point.

- **Aquatic invertebrates**

  - **Acute aquatic invertebrate toxicity:** For aquatic invertebrates, the standard species tested for acute toxicity include *Daphnia magna* (water flea), *Ceriodaphnia dubia* (water flea), *Mysidopsis bahia* (pocket shrimp) and *Crassostrea virginica* (eastern oyster), which represent two warm freshwater species and two warm estuarine species. Consensus opinion is that the eastern oyster test is usually the most sensitive. Only three of these species have been tested with diquat; *Daphnia magna*, pocket shrimp and the eastern oyster. The endpoints for these tests are immobility for the arthropod species and shell growth for the eastern oyster. The endpoint is expressed as the 48- or 96-hour EC50 or LC50 for the three arthropods, and EC50 (dosage causing 50% decrease in shell growth in 96 hours) for the oyster. A number of other non-standard species and non-standard methods were tested with the diquat and are listed along with a summary of the data in Table 2

  - **Life-Cycle invertebrate toxicity:** Life-cycle invertebrate toxicity studies are typically done with *Daphnia magna* (daphnia), *Ceriodaphnia dubia* (daphnia) and *Mysidopsis bahia* (mysid shrimp). However, only a very limited database is available for diquat products and their chronic effects on invertebrates. The only species of invertebrate that has been tested with diquat is *Daphnia magna*. Since these are the only life-cycle tests presented, the database may be insufficient to support life-cycle safety of diquat on invertebrates. These tests are usually run for 21 days with *Daphnia magna*, 7 days with *Ceriodaphnia dubia* and 28 days with mysid shrimp. The parent generation is selected from a group of animals less than 24 hours old. The endpoints are immobility, reduction in number of live newborns produced per female, and growth of the parent daphnids or mysids during the test. The endpoint is expressed as the most sensitive EC50 in reference to immobility, reduction in neonate production and reduction in growth and the NOEC or MATC in reference to the most sensitive endpoint. The advantage of using the mysid shrimp, as a test organism is that, since it shows sexual dimorphism, certain endorine disruptive effects could be determined in the parental generation as it matures. However, these endorine effects cannot be interpreted or correlated with similar effects on vertebrates since it is unlikely that the effects of steroid sex hormones like estrogen or testosterone determine sex in this species. However, this species was not tested for diquat's effect on its life-cycle.

- **Sediment organisms**

  - **Sediment organism acute toxicity:** There are major disagreements among scientists as to how sediment organism studies should be conducted. The disagreements are so great that many researchers believe daphnia studies make good surrogates for sediment organism studies. The main problem with sediment organism studies is that these organisms require sediment with a specific particle size in order to function properly in a physiological sense. However, in acute tests the sediment is often eliminated from the study because it adsorbs the toxicant and interferes with analytical chemistry when the sediment phase must be extracted. Most short-term (acute) 96-hour sediment organism studies are conducted without sediment present. There is a need for these tests since there is no reason to assume that sediment organisms will respond in a manner similar to other aquatic invertebrates. These sediment organism acute toxicity studies are conducted in a similar manner as acute tests with other invertebrates except that the age at initial exposure and the exposure period is specific to each species. These specific characteristics are listed in Table 2. However, since diquat bound to sediments is generally considered to be biologically unavailable to sediment organisms, these sediment toxicity studies may not have great utility for testing the effects of diquat on benthic species.

4.1.2 Exposure Routes

Regardless of the organism, aquatic exposure to diquat can take several routes. These include adsorption from the water column, consumption of water or organisms while eating, contact with plants or sediments that have been treated with the test substance, or eating the granules. More detail for exposure routes is given below:

- Aquatic Algae: Exposure is through adsorption from the water column.
- Fish and Aquatic Invertebrates: Fish and aquatic invertebrates can be exposed to diquat by:
  - Adsorption through the "skin" or cuticle.
  - Adsorption through the gills.
  - Adsorption through the gut from the consumption of other animals or plant and algal material.
  - Adsorption through the gut after eating the formulated pesticide granules found at the bottom of the water body. This is not relevant to the diquat case since the formulation of diquat typically used is a liquid.
  - Detritovoirs can be exposed through eating detritus found in the sediment or catching the detritus from upper regions as it floats past.

For terrestrial organisms, exposure routes can be: 1) contact with treated water, 2) consumption of treated water, 3) organisms that have been in treated water, or 4) consumption of the pesticide granules if they have access to them; again not relevant to diquat liquid formulations.

4.2 ENVIRONMENTAL TOXICITY REVIEW: EFFECTS OF THE PHYSICAL AND CHEMICAL ENVIRONMENT ON HABITAT

There is no evidence that prior exposure to diquat will enhance the degradation of diquat by microbial organisms. Sites exposed to diquat products may degrade diquat when it is on the surface of plants or in the water column. However, after diquat has been adsorbed to the soil, it is often unavailable to further degradation (Dyson and Takacs, 2000). Once adsorbed to the sediment, diquat may be stable and not degrade further for several years. For example, simulated impoundments of Lake Mendota water treated with diquat had concentrations at 92% of the highest concentrations seen in this sediment for 180 days after application under aerobic conditions (Simsiman and Chesters, 1976). Similar stability in Lake Mendota sediment under anaerobic conditions for 95 days after application was also noted. Frank and Comes (1967) similarly found that sediment in the bottom of a pond in Colorado continued to adsorb diquat for 56 days after application, eventually reaching a concentration of 37 ppm c.e. and this diquat concentration was still present at a concentration of 24 ppm for at least 160 days after application. However, typical sediment concentrations are much less than this even after multiple applications. E.G. Beasley (1966 in Hammer, 1994) found 1.7 ppm in pools 4 years after treatment with 0.3 kg/ha diquat; Gilderhaus (1967) found 10 ppm c.e. diquat after treatment of ponds with 1.0 ppm diquat but higher application rates (3.0 ppm) or multiple applications could result in up to 72 ppm diquat adsorbed to the to the sediment after 24 weeks; Newbold (1975 in Hammer, 1994) found 0.6 ppm c.e. diquat six months after treating ponds with 1.0 ppm c.e. In a well controlled experiment, four maximum use rate applications applied at one-month intervals, were made to two ponds in Florida. Sediment concentrations found 180 days after the last application were quite low ranging from <0.01 ppm to 0.44 ppm (Fujie, 1988). This study was especially interesting because sediment concentrations did not appear to build after multiple applications. It is unclear if control measures as stringent as these were carried out in any of the other cited experiments.

Due largely to the adsorption of diquat by sediment and plants, the concentration of diquat in the water column rapidly dissipates. Diquat dissipates in water to levels below the detectable limit of ~0.01 ppm in 4 to 12 days depending on the sediment type and application rate (Coates et al, 1964; Frank and Comes, 1967; Grzenda et al, 1966; Yeo, 1967 and Fujie, 1988). Concentrations of diquat in water when applied at not greater than the maximum use rate, have been observed to drop below levels that are typically considered to be harmful to fish in less than four days. For example, applications of the maximum use rate to coves of Finley Lake, New York resulted in concentrations of 0.08 ppm c.e. one day after application and 0.024 ppm c.e. four days after application (Sewell, 1969). In a similar controlled experiment, using ponds in Florida, Fujie found concentrations averaging only 0.060 ppm c.e. after one-day of dissipation and 0.020 ppm c.e. after three days of dissipation. Maximum rates applied to almost the entire area of Lake Steilacoom, Washington resulted in concentrations of 0.071 ppm c.e. after one day of dissipation and 0.045 ppm c.e. after three days of dissipation. Similar observations were made for ponds in Colorado that were treated with 0.62 ppm c.e. where the day one1 concentration was 0.49 ppm and the day four concentration was 0.01 ppm c.e (Frank and Comes, 1967). In cases where the treatment rates were somewhat higher than the currently recommended rates, concentrations in the water column were proportionally higher. For example, when ponds in South Carolina were treated with diquat at concentrations up to 2.5 ppm c.e., the diquat concentration dissipated to 0.81 to 1.25 ppm c.e. in one day and 0.01 to 2.89 ppm c.e. in six to seven days.

Bioconcentration in plants occurs to a very high degree. This bioconcentration by plants reaches up to 2,430 ppm for the diatom *Navicula pelliculosa* and 600 to 1,310 ppm for Hydrilla (*Hydrilla verticillata*) (Cassidy and Rogers, 1989) and Eurasian watermilfoil (*Myriophyllum spicatum*) (Birmingham and Colman, 1983), respectively. Other susceptible species of algae and aquatic vascular plants may accumulate diquat to similar levels. Bioconcentration in animals has not been seen with diquat. Whole body bioconcentration factors have been seen to be less than one for bluegill sunfish (Hammer et al, 1987), channel catfish and *Tilapia* (Fujie, 1987 and Schultz, 1995). However, the BCF was somewhat greater than 1.0 in most species of aquatic invertebrates with mayfly nymphs at BCF = 32 (Hammer et al, 1987)*, Daphnia magna* at BCF = 83 (Hammer, 1987), crayfish at BCF = ~1.0 (Fujie, 1987), clams at BCF = ~10 (Fujie, 1987) and Pacific oysters at BCF = ~10 (Thompson, 1987). Furthermore, the diquat that is adsorbed by these species is rapidly dissipated with a half-life that is typically less than three days; this would lead to 90% or better elimination of diquat in less than 10 days after exposure to diquat.

4.2.1 Potential Soil and Sediment Interactions

4.2.1.1 Impact of Various Soils (Sediment/Substrate) Composition

Summary: Due to its extremely high soil/water distribution coefficient, diquat adsorbs well to most soils. Furthermore, this adsorption is highest on soils with a high clay and silt content (Kd = 10,000 to 60,000) and lowest on sediments with a high sand and gravel content (Kd = 30 to 1000). These very high soil/water distribution coefficients can lead to a rapid disappearance of diquat from the water column and very high concentrations of diquat in the sediment. Therefore, *the concentration of diquat in sediment ranges from < 1.0 ppm c.e. (Fujie, 1988) to more than 37 ppm c.e. (Frank and Comes, 1967). However, in most cases concentrations of diquat in sediment from treated ponds was far less. Increases in sediment concentrations will often continue for up to a month after application of diquat to the water body. This continued increase in sediment concentrations of diquat is probably due to the initial adsorption of diquat by plants and subsequent release of diquat after the plants have died and sunk to the bottom of the water body. Although the concentration of diquat in sediment can be high and persistent, most soils appear to be able to adsorb diquat concentrations as high or higher than 250 ppm c.e. without release of diquat from the sediment (Hiltbran et al, 1972). In more than 30 years of diquat use to control aquatic weeds, there has never been a recorded release of bound diquat at concentrations that exceed the levels of aquatic organisms to tolerate it (Hiltibran et al, 1972 and Dyson and Takacs, 2000).*

Diquat residues do not persist in the water column for any extensive period of time. The time of dissipation from the water column to levels below the MCLG (maximum concentration level goal) is typically 4 to 12 days. However, persistence in sediment can be very long with concentrations of diquat continuing to accumulate for more than a month in some cases and then persisting in an unchanged state for 180 days or more (Frank and Comes, 1967; Simsiman and Chesters, 1976, and Rogers et al, 1992). Although diquat has a tendency to accumulate in some sediments over time, conservative estimates indicate that 10 to 50 applications would be necessary before the sorptive capacity of diquat would be overcome and diquat would be released back into the water column. However, more realistic estimates indicate that it would take hundreds of applications before the sorptive capacity of diquat was overcome and enough diquat could be released back into the water column to potentially cause damage to the biota (Dyson and Takacs, 2000; Shaw et al, 1995). Metabolism may occur on the surface of plants degrading diquat into a variety of metabolites. Degradation of diquat does not appear to occur in plants and the diquat released from plants after they have died and sunk to the bottom is readily adsorbed to the sediment where it may be retained unchanged for a long period of time (Frank and Comes, 1967; Simsiman and Chesters 1976)

Diquat adsorbs readily to most soil/sediments. Typical ranges of adsorption coefficients (Kd) on sediments/soils is 30 to 1,000 for sand-gravel, 1,000 to 10,000 for sand, and 10,000 to 60,000 ppm for silty clay loams (Ritter et al, 1995). In general, a compound with a Kd value of less than 5, and particularly with Kd values less than 1.0 are considered highly mobile in soil (U.S. EPA, 1986a in Ebasco, 1989). Therefore, under normal conditions, the ability of diquat to contaminate ground water is extremely unlikely. There have been no recent credible reports of ground water contamination from the use of diquat to control aquatic weeds which is expected based on the extremely high adsorption coefficients that are seen experimentally. However, because of its tendency to accumulate on the upper layer of sediments previously exposed to diquat under aquatic conditions, diquat is susceptible to erosional processes and can be transported to water bodies during runoff. More details on the nature of soil mobility and its ecological meaning are discussed in Section 3.

4.2.1.2 Potential for Increased Erosion and Re-suspension of Soils and Sediments from Plant Removal

Summary: Since these products are not generally applied terrestrially, classical erosion effects typically do not occur. However, removal of plants from irrigation canal situations may result in erosive processes occurring to a limited extent.

During aquatic weed control, diquat products are applied directly to water and not to the terrestrial environment. Therefore, classic erosion, in a strict sense, generally does not occur from this use. That is, soil and humic material is not dislodged by wind and water and washed into the waterway due to the removal of plants from the adjacent terrestrial environment. Removal of plants from non-flowing water systems may allow for the re-suspension of sediment from the bottom of a lake or pond due to wind mixing of the water, interactions with benthic organisms and direct interfering effects of human beings with the hydrosoil during periods of either work or recreation.

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The only likelihood of classical erosion occurring is if ponds treated early in the season evaporate or are drawn down. Under such conditions, the previously submerged banks and possibly bottom of the lake will temporarily become terrestrial environment subject to classical wind and water erosion. Erosion in these areas would initially be high due to lack of plant cover. However, dead aquatic vegetation, if not yet broken down by natural decay processes, would function like a mulch to help reduce erosion until the area is re-vegetated with terrestrial plants or the area is re-flooded with water. A worst case scenario could occur if the area does not re-vegetate before the dead vegetation completely decomposes and exposes the underlying soil/sediment.

Without the presence of plant species providing soil stability, physical characteristics of the soil/sediment are the primary factors affecting soil erosivity. The two most important soil characteristics affecting water-influence are infiltration capacity and structural stability. Soil texture, organic content and clay content (i.e. swelling clays also influence infiltration capacity (Brady 1974 in Ebasco, 1993), structural stability depends on the ability of soil/sediment aggregates to withstand breakup caused by physical bombardment of water and wind. This depends on many factors, including both biological (mechanical, binding action of microorganisms, cementing action of the intermediate products of microbial synthesis and decay, and cementing action of the more resistant stable humus components) and the organic/inorganic component interaction that provides bridging between organic matter and soil/clays (Brady, 1974 in Ebasco, 1993).

The Soil Conservation Service (USDA, 1978a in Ebasco, 1993) has developed simplified erodibility factors (K) based purely on soil texture of different topsoil and subsoil regimes. These K factors can be used as approximate erosivity estimates. The K values listed in Table 3 are used in predicting rainfall erosion losses with the universal soil loss equation (USDA, 1978b in Ebasco, 1993) and may be used as relative indicators of erosivity across different soil texture classifications.

The loss of soil by wind erosion involves detachment and transport mechanisms. Detachment results from abrasion by both wind and entrained particles. Transport may cause soil particles to travel along the land surface by saltation or to travel parallel to or upward from the land surface by suspension. Soil moisture is the primary factor in determining erosion by wind. Other soil characteristics include mechanical stability of dry soil clods and aggregates, presence of a stable soil crust and bulk density and size of erodible soil fractions (Brady, 1974 in Ebasco, 1993). Once detached, finer-grained particles are most likely to move in the wind and rain. Water erosion has a tendency to have a greater effect on sandy loam than on sediments (particularly silt loam) since water effectively conglomerates and seals the finer soil/sediments. Greater amounts of diquat or other pesticides are more likely to wash out of sandy loam than silt loam sediments. Coats et al (1966) found that leaching of diquat was much greater from columns containing sand than sandy loams and that 10 acre-inches of water leached less than 5% of an applied 14C-label below the top 0.5 inches of sandy loam soils. Details were only given with an experiment done with paraquat. However, similar results were claimed for diquat. Various soil types retained diquat at different levels in adsorption experiments and while it is unclear what the exact adsorption coefficients were, it is clear that bentonite clay retained more diquat than kaolinite clay; and that kaolinite clay retained more diquat than sandy loam soil. It has been reported that montmorillonite clays have even higher capacities to adsorb diquat than bentonite clays (Simsiman et al , 1976). Nevertheless, because of its tendency to accumulate on the upper layer of soil/sediment, diquat is susceptible to erosional processes and can be transported to water bodies during runoff.

Strictly aquatic herbicides like diquat are not commonly used to treat canals or ditch banks. However, diquat may be used to control weeds growing in noncrop areas which, under a liberal view, could be interpreted as allowing for the control of riparian weeds on the banks and top berms of ditchbanks. In the case of canals treated with diquat, erosion is unlikely to be a problem. Generally speaking, the major aquatic weed problems in irrigation canals are emergent or riparian weeds growing on the banks of drainage canals. There are times during the season when these riparian weeds may become a problem. These herbicides are used to control emergent macrophytes. It is unclear from the label which species may be controlled by diquat. However, since efficacy is claimed for above ground species, emergent and riparian weeds like pennywort, cattails, purple loosestrife, black mustard, broad leaf plantain, pigweed, dandelion, Russian thistle or lambsquarters that are found growing on the banks of irrigation and drainage canals are likely to be controlled. Canals typically are constructed with 3:1 bank slopes and are designed to convey peak demand flows without eroding. Irrigation canals can be lined with a variety of materials including earth, blended earth (clay mix to reduce seepage loss), asphalt, concrete or geotextile. Although vegetation may invade the channel over time, vegetation-lined channels are typically not constructed because plant growth can reduce the canal's conveyance capacity. The main objective in canal design is to minimize losses from the canal and to maximize conveyance capacity. Therefore, the irrigation districts actively remove nuisance plant growth. Plant removal operations are usually performed at the end of the irrigation season. The general procedure involves filling and sealing the canal after which the area is treated with a herbicide. The main purpose of diquat applications would be to restore irrigation water flow by eliminating dense stands of purple loosestrife or other emergent or riparian weeds. Because irrigation canals are typically designed to operate at capacity under unvegetated conditions, removal of nuisance plants is unlikely to result in destabilization of irrigation canals. However, depending on site-specific conditions, erosive processes and the amount of sediment trapped by loosestrife or other dense stands of weeds, removal of these plants may contribute to limited sediment erosion and transport.

- **Effects of removal of weeds on habitat**

  Removal of weeds from the newly formed terrestrial habitat may cause additional silt and nutrients to enter adjacent water bodies. Such an increase in nutrient load may lead to algal blooms and eutrification of the water body. Also, the removal of these terrestrial plants will decrease the amount of new terrestrial habitat that terrestrial animals may utilize. Removal of the newly established plants may increase the likelihood of flooding and return the water body to the previous aquatic condition (flooding). Flooding can increase the amount of habitat available for fish and amphibians to utilize for feeding and spawning (Goldman and Horn, 1983). Negative impacts from isolated flooded areas could be stranding or hydrological "jumping" of current flows to a new, but not necessarily, superior channel.

4.2.1.3 Effects on Pristine Sites

In general, the prior use of diquat does not appear to affect the rate of removal of the pesticide from the aquatic environment. However, long-term use of diquat has a small potential to overwhelm the cation exchange capacity of the soil and allow diquat to be re-released to the water column. This is not likely since most sediment and soil have the capacity to adsorb diquat at levels that exceed 250 ppm c.e. (Hiltibran et al, 1972) and there has never been a reported field incident of diquat being released from sediment at concentrations that are high enough to cause adverse impact of the aquatic biota (Dyson and Takacs, 2000 and Shaw et al, 1995) even though diquat has been used to control aquatic weeds for more than 30 years. If the treatment site has not experienced diquat treatments before, the sediment may adsorb diquat at concentrations up to 37 ppm c.e. in 56 weeks after treatment with 0.62 ppm c.e. (Frank and Comes, 1967). However, more typical rates of adsorption to sediment are 0.04 to 0.1, 0.6, 1.7, and 10 ppm c.e. (Fujie, 1988; Newbold,1975 in Hammer 1994; Beasley, 1966 in Hammer, 1994, and Gilderhaus, 1967) with single applications ranging up to 1.0 ppm c.e.

Isolates of bacteria are capable of degrading diquat when grown in pure culture. Species of bacteria capable of degrading diquat include *Pseudomonas fluorescens*, *Rhizobium* spp. and *Aerobacter aerogenes*. This may be an important route of degradation since *P. fluorescens* is capable of withstanding up to 2,500 ppm diquat. Although three separate metabolites have been isolated from tests conducted with 14C-labeled diquat, these metabolites have not been identified. There are indications that diquat adsorbed to kaolinite clay is more susceptible to degradation than diquat adsorbed to montmorillonite clay (Weber and Coble, 1968 in Simsiman et al, 1976). It is not known whether the diquat associated with organic matter at any stage of plant decomposition is more susceptible to microbial degradation than that adsorbed on clay surfaces. However, Burns and Audus (1970, in Simsiman et al, 1976) found that paraquat associated with organic components of soil is more readily degraded by *Lipomyces sarkeyi* than paraquat associated with the mineral components of soil. Although diquat is readily degraded by photolysis in aqueous solutions (Simsiman et al, 1976), it seems unlikely that photolysis would be a significant degradation factor in water that is more than 15-cm deep. However, since adsorption tends to slow the rate of degradation it is likely that diquat will accumulate in soils, particularly in those containing montmorillonite clay.

4.2.1.4 Effects on Contaminated Sites

Although concentrations of diquat accumulate with each exposure, it seems unlikely that bound diquat will accumulate to concentrations that are high enough to overwhelm the adsorptive capacity of most soils. Overwhelming a soil's capacity to adsorb diquat at levels that exceed 250 ppm c.e. (Hiltibran et al, 1972) appears to be very unlikely. Furthermore, there has never been a reported field incident of diquat being released from sediment at concentrations that are high enough to cause adverse impact on the aquatic biota (Dyson and Takacs, 2000 and Shaw et al, 1995) even though diquat has been used to control aquatic weeds for more than 30 years. Although multiple applications and higher treatment rates result in sediment concentrations that are significantly higher at 71 ppm in 24 weeks after the second treatment at 1.0 ppm or 20 to 50 ppm c.e. in 24 weeks after treatment with 3 ppm c.e. (Gilderhaus, 1967), Fujie (1988) found that multiple treatments at the maximum treatment rate did not increase the concentrations typically found in the sediment. No explanation of this was given. However, it was stated "once adsorbed, the residue is bound with little or no vertical movement through the sediment layer or desorption back into the water column", which seems to contradict the fact that the levels of diquat did not increase significantly with subsequent applications. However, Simsiman et al (1976) found that diquat underwent slow degradation after being bound to sediment. The ability of bacteria to degrade diquat has been discussed in the previous section.

4.2.2 Environmental Persistence

Summary: Although diquat does not persist in the water column, it is partially degraded by various species of bacteria on the surface of plants. Concentrations of diquat accumulated within the plant do not degrade. The diquat is released intact from the dead and dying plant and incorporated in the sediment. However, Simsiman and Chesters (1975) found that only 19% of the applied diquat becomes associated with the sediment after treatment and 32% of the applied diquat is metabolized to water soluble products within 22 days (Simsiman and Chesters, 1976). Diquat can persist in sediment for a very long period of time. Peterson et al (1994) estimated that diquat could persist in sediment with a half-life of 1,000 days. This is not an unreasonable assumption since diquat has been known to accumulate in sediment for 36 to 56 days (Simsiman and Chesters, 1976 and Frank and Comes, 1967), and appears to remain in the sediment as unchanged diquat for at up to 180 days. Both longer and shorter resident times in the sediment have been observed. Beasley (1966 in Hammer, 1994) found unchanged diquat in sediment four years after application at 0.3 kg c.e./ha. However, Fujie (1988) did not find substantial evidence for a significant build up of diquat even after four treatments at the maximum use rate. Once diquat has been bound to the sediment, it appears to be inactivated or biologically unavailable and therefore not able to cause significant damage to the biota. For example, diquat in the presence of sediment is much less toxic (LC 50 = 6.8 ppm c.e.) to Hyalella azteca *than in the absence of sediment (LC50 = 0.048 ppm c.e.) (Wilson and Bond, 1969). The aquatic pill bug (*Asellus *spp.) could tolerate diquat at 10 ppm in the presence of sediment but is unlikely to be able tolerate diquat in the absence of sediment (Williams et al, 1984). Diquat bound to sediment or soil has no toxicity to wheat seedlings at concentrations up to 100, 1,000 and 50,000 ppm c.e. on sandy loam soil, kaolinite clay and bentonite clay, respectively (Coats et al, 1966). Daniel (1972) found that Eurasian water milfoil planted in sediment that had been exposed to water previously treated with 2.2 ppm Cutrine® and 3.0 ppm diquat grew with no apparent herbicidal effects.*

Bioconcentration in plants occurs to a very high level. For example, various species of algae and Eurasian water milfoil accumulated diquat at concentrations of 555 to 2,430 ppm c.e. after being exposed to water containing 0.3 to 3 ppm c.e. diquat (Birmingham and Colman, 1983). Fish and invertebrates generally did not bioaccumulate diquat. E.g., the bioconcentration factors of diquat found in bluegill sunfish, Tilapia *spp. channel catfish, and crayfish were generally below 1.0 (Hammer et al, 1987, Schultz et al, 1995, Fugie, 1987, and Fujie, 1987). However, the BCF in freshwater clams,* Daphnia magna*, Pacific oysters, and mayfly nymphs was somewhat higher than 1.0 (Fujie, 1987, Hammer, 1987, Thompson, 1987 and Hammer, 1987). In this case, initial concentrations of even ~1.0 ppm may be high enough to require that the current shellfish tolerance of 0.1 ppm be increased. Another alternative would be to add a restriction to the label specifying how long after treatment that shellfish may be harvested (Fujie, 1987). After diquat has been removed from the system the depuration half-life is usually less than or equal to three days for catfish, bluegill sunfish* Tilapia *sp. and* Daphnia magna*. However, in the pacific oyster, after 14 days only about 30% depuration was seen. The majority of residues in fish edible tissue, skin and kidney remained as diquat while most of the recovered residues in liver and bile were metabolites of diquat.*

Eurasian watermilfoil apparently bioconcentrates diquat to levels of at least 400-fold. (Birmingham and Colman, 1983). The release of diquat from decaying plant material probably has little effect on the concentration of diquat in water since the sediment rapidly adsorbs the released diquat. However, some authors have indicated that plant-released diquat may be high enough in concentration to affect living plants.

The environmental persistence of diquat products in the field can be quite variable; the dissipation half-life in water varies from less than one day to approximately 4 days. However, according to Shaw et al (1995) the most typical half-life under EPA's worst case scenario is 0.75 days. Dissipation in water is typically not due to degradation by microorganisms. However, approximately 32% of applied diquat is degraded to water soluble metabolites by bacteria that are associated with the surface of the resident plant biota (Simsiman and Chesters, 1976). Dissipation of diquat is primarily due to the adsorption by sediment and secondarily by plant tissue. Approximately 19% of applied diquat has been observed to adsorb to sediment directly from the water (Simsiman and Chesters, 1976). Approximately 12 days later, the plants die and release diquat, which is further, adsorbed to the sediment. This final adsorption of diquat released from dying plants can take nearly two months to completely adsorb to the sediment (Gilderhaus, 1967, Simsiman and Chesters, 1976, Hiltibran et al, 1972 and Frank and Comes, 1967).

Diquat is not expected to be found on soils unless an irrigation or flood incident occurs. However, the expected half-life on soils may range to as long as 1,000-days (Peterson et al 1994). Effects of diquat on terrestrial crops after irrigation or flooding are discussed in Section 4.2.5.

4.2.2.1 In Water

A detailed review of the persistence of diquat in water can be found in Section 3.1.3.3. The dissipation half-life for diquat ranges from less than one day (Fujie, 1987) in Bivens Arm, FL to approximately four days (Sedar, 1997) in Lake Steilacoom depending on the conditions found in the water at the time of treatment and during the breakdown of diquat. The conditions that may affect the dissipation of diquat include the amount and type of sediment present and the amount and type of plants present. Some species of bacteria including *Pseudomonas fluorescens*, *Rhizobium* spp. and *Aerobacter aerogenes* are known to degrade diquat. This may be an important route of degradation since *P. fluore*scens is capable of withstanding up to 2,500 ppm diquat. Although three separate metabolites have been isolated from tests conducted with 14C-labeled diquat, these metabolites have not been identified. The level of turbidity in the water is also known to affect the rate of dissipation when diquat is present; this factor alone can decrease the residence time of diquat in the water column from about 27 days under low turbidity to about seven days under high turbidity. Application at the maximum use rate can result in concentrations that vary as follows: 0.11 to 0.13 ppm c.e. (Gravelly Lake and Lake Steilacoom) to 0.37 ppm c.e. in Bivens Arm and Shongaloo Fish Hatchery, FL; 0.1 to about 0.5 ppm c.e. in Finley Lake, NY and Chautauqua Lake, NY; and 1 ppm c.e. in three ponds in Wake, NC (Sedar, 1997; Sewell, 1969, and Langeland and Warner, 1986). The water column dissipation half-life is usually about 0.75 days under the worst case conditions.

Diquat was applied to impounded ponds in Bivens Arm, FL of Shongaloo Fish Hatchery, FL. On June 10, 1987 (one day post-application), the concentration of diquat in water ranged from 0.072 to 0.095 mg/L. After ten days of monitoring, the concentration in these ponds fell below 0.004 ppm c.e. The half-life under this scenario ranged from 1.6 to 2.3 days. An additional three treatments to these ponds with 30 days between treatment resulted in similar residence times for diquat and half-lives that ranged from 0.72 to 1.4 days. Although EPA (in EPA RED, 1995) estimated the half-life for these ponds to be one to two days, Shaw et al (1995) estimated the half-life of these ponds to be closer to 0.75 days.

In Lake Steilacoom, the levels of diquat degraded to 0.047 to 0.091 ppm in one day and remained at approximately this concentration for at least three days. By 12 days the concentration of diquat in Lake Steilacoom had dissipated to 0.003 to 0.007 ppm c.e. This yields a half-life of approximately 2.5 days.

At Finely Lake, NY, the concentration of diquat fell to 0.079 ppm one day after application. Four days after application the concentration of diquat fell 0.023 ppm and to undetectable levels (<0.001 ppm c.e.) in eight days. These results yield a dissipation half-life of approximately one to two days.

At Chautauqua Lake, NY, the concentrations of diquat dropped to below the level of detection (0.001 ppm) in less than one day, which yields a half-life of less than one day. Similarly, at the three ponds in Wake, NC, diquat levels dropped to trace concentrations (0.01 ppm) in 1.5 days, also yielding a half-life of less than one day.

In light of the data presented above, concentrations of diquat will normally drop below the limit of detection in approximately ten days and the half-life will typically be less than one day. However, Hiltibran et al (1972) found that the rate of dissipation can vary dramatically when the application rate ranges from 0.5 to 1.0 ppm. Concentrations of diquat can drop from the application rate to virtually undetectable in less than four days or concentrations applied at 1.0 ppm can persist at levels of >0.21 ppm for up to 13 days. The length of time to complete dissipation seems to be governed primarily by the degree of turbidity with short residence times occurring when algal blooms are present. However, Shaw et al's (1995) estimate of 0.75 days is a good general value for the half-life in a water body when only half of the water body is treated at any one time (as recommended on the label).

In all of these cases, it is not clear what the treatment scenario was. However, it is clear that the entire water body was treated at Lake Steilacoom, WA and Lake Finley, NY; and that only the margins of Gravelly Lake and Lake Chautauqua were treated.

4.2.2.2 In Sediment

A detailed review of the persistence of diquat in sediment can be found in Section 3.1.3.2. Diquat from the application of Reward® LA may have significant concentrations and long half-lives in sediment. The stability of diquat in sediment is very high due to irreversible binding to the sediment that inactivates diquat and makes it biologically unavailable for degradation. Both laboratory and field studies indicate the stability of diquat in association with sedimen. Diquat has been found to be immobile with laboratory adsorption coefficients that vary from 1,882 to 10,740 in sandy loam, sandy clay loam and loam soils (EPA, RED, 1995). Under aquatic aerobic conditions, over 95% of the diquat residues were associated with the sediment; and under aquatic anaerobic conditions more than 89% of the diquat residues were associated with the sediment and in 9 months over 95 % of the identified residues consisted of diquat. Diquat has been observed to be rapidly removed from the water column by becoming irreversibly bound to soil (Dyson and Takacs, 2000). In general, adsorption is very strong until close to the adsorption capacity (Hiltibran et al, 1972). Concentrations of diquat are typically fairly low (<1.2 ppm at 30 days after the fourth treatment, and as high as 10 or 37 ppm in 36 to 48 days). It may take a fairly long time for concentrations of diquat to reach the maximum level that will normally be found in sediment since large amounts of diquat are adsorbed by plants and algae (Birmingham and Colman (1983) and are released only after plants are growing old or dead and have sunk to the bottom (Daniel, 1967). These concentrations can be very stable with little or no degradation occurring on the sediment for up to 180 days after application (Rogers et al, 1992; Fujie, 1988; Gilderhaus, 1967; Frank and Comes, 1967, and Simsiman and Chesters, 1976) (Table 5).

It has been calculated under a worst case scenario that it would take 10 to 50 applications of diquat to overcome a typical sediments sorptive capacity and allow for a release of diquat that would effect sediment associated plants and animals in the water column (Birmingham and Colman, 1983). However, during aging and death, but prior to decay, bacteria associated with the dead and dying foliage degrades 32% of the applied diquat to water soluble metabolites. Typical bacterial species that may contribute to the degradation of diquat have been discussed in previous sections. However, this is a very conservative assumption and assumes that the sediment will adsorb all of the applied diquat and that it does not degrade during its residence in the sediment. Nineteen percent of the applied diquat adsorbs directly to the sediment and less than 50% of the applied diquat subsequently released from dead and dying plants will bind tightly to the soil. When this natural remediation is considered, it will probably take hundreds of applications to overcome the sorptive capacity of the sediment (Shaw et al, 1995 and Dyson and Takacs, 2000).

4.2.2.3 In Soil

The presence of diquat in soil is not anticipated from aquatic treatment unless flooding occurs or the water is used for irrigation. The label prohibits use of diquat treated water for irrigation for a period of time after treatment (Reward® LA label, 2000). The length of this water use restriction varies depending on whether the irrigation water will be used on non-food or food crops and the rate of application of diquat to the water body. However, diquat may not be used under any application rate to irrigate food crops for at least 5 days. However, according to Gangstad (1986), up to one acre-inch of water may be applied by soil or overhead irrigation without significant residues being detected in the crop. While little harm may occur on a crop irrigated one time with water containing concentrations of 0.45 ppm or more on some crops, multiple irrigations could result in substantial harm to crop yield (Shearer and Halter, 1980).

4.2.2.4 Potential for Bioaccumulation or Bioconcentration in Fish, Aquatic Invertebrates, Phytoplankton, Zooplankton, Birds, Mammals and Insects

The potential for bioaccumulation (BAF) and bioconcentration (BCF) is low to moderate for diquat. In many cases diquat will be adsorbed to a fairly high degree by plants. For example, Birmingham and Colman (1983) have found the Eurasian water milfoil (*Myriophyllum spicatum*) adsorbed diquat at concentrations up to 1310 ppm c.e. from water treated with diquat at concentrations of 0.03 to 3 ppm c.e. Algae were also observed to adsorb diquat at concentrations of 0.55 ppm c.e. (*Chlorella vulgaris)*, 2,430 ppm c.e. in *Navicula pelliculosa*, 1,550 ppm c.e. in *Anabaena flos-aquae* and 1,270 ppm c.e. in mixtures of *Spirogyra* sp. and *Cladophora* sp. Other species of algae and plants have been noted to readily adsorb diquat including alligatorweed (*Alteranthera philoxe*roides), American waterweed (*Elodea canadenisis*), sago pondweed (*Potamogeton pectinatus*) and American pondweed (*P. nodosus*), filamentous algae (*Pithophora oedogonia*), and watermeal (*Wolffia columbiana*) (Hiltibran et al, 1972, Langeland and Warner, 1986; Simsiman et al, 1976 cites Funderburk and Lawrence, 1963; and Davies and Seamen, 1968). It is interesting to note that diquat does not bioaccumulate on terrestrial plants and very little or no residues are seen in terrestrial plants even when they are foliarly treated if sufficient time is allowed for photolysis of diquat to pyridine compounds, TOPPS, picolinic acid, and

volatile fragments of pyridine compounds and picolinic acid (Black et al, 1966 in Simsiman et al, 1976). Diquat does not appear to be metabolized in susceptible plants and the fate of diquat in tolerant species has not been reported. Diquat is not bioaccumulated in most animals.
The bioconcentration factor can be predicted throughout most of its range from the following equation: BCF = K_{ow} x 0.05. In fish such as channel catfish (*Ictalurus, punctatus*), bluegill sunfish (*Lepomis macrochirus*) and *Tilapia*, the bioconcentration factor for the whole body is generally $\leq$~1.0. Edible tissues may have concentrations of diquat that are approximately 0.7 ppm c.e. when the water contains ~1.0 ppm c.e. of diquat. However, in aquatic systems where dissipation occurs, the concentration of diquat in edible tissue will typically not be higher than 0.04 ppm c.e. (Schultz et al, 1995, Hammer et al, 1987, and Fujie, 1987). In field studies, diquat was not observed to bioaccumulate in bluegills or catfish after multiple noncrop applications. However, diquat was not detected in the edible tissue of the bluegill sunfish, but was detected at concentrations as high as 0.3 ppm c.e. in the edible tissue of *Tilapia*. These *Tilapia* residues depurated to concentrations below 0.1 ppm c.e. in three days. Crayfish bioaccumulate diquat in a manner similar to fish but bivalves like freshwater clams and oysters may significantly bioaccumulate diquat (BCF = ~10). It is conjectured that these relatively high levels of diquat in bivalves are associated with the siphon and gastrointestinal tract and will be rapidly depurated. However, only 30% depuration is seen in the oyster after 28 days. This bioconcentration in bivalves may be high enough that the tolerance of 0.1 ppm in mantle tissue is exceeded by as much as 3- or 4-fold at the time of harvest (Fujie, 1987 and Thompson, 1987). The bioconcentration factor in daphnia, mayflies, and other fish food organisms can be relatively high (BCF = 8.3 and 32, respectively). However, this is not a high enough concentration to be considered to be a general bioaccumulator (Table 4); and should therefore not be a serious concern with predatory fish. Since the octanol/water partition coefficient K_{ow} is quite low (0.000025) for diquat it can be predicted that bioconcentration (0.05 x K_{ow} = <1.0) would be non-accumulative (Table 4). The bioconcentration factor can be predicted throughout most of its range from the following equation: BCF = K_{ow} x 0.05. Therefore the BCF is predicted to be ~0.0000013 for diquat. Such extremely low K_{ow} values make an accurate prediction of the BCF difficult. However, these values do indicate that diquat will be non-accumulative under most situations.

- **Bioaccumulation and bioconcentration**

  Bioconcentration factors of less than 10 to 60 are not generally considered to be bioaccumulative. When diquat is applied in the laboratory at concentrations from 0.03 to 3.0 ppm c.e., the concentration of diquat found in Eurasian watermilfoil and a variety of algae ranges from 0.55 to 2.43 ppm c.e. dry weight. This suggests that diquat is adsorbed to the surfaces of plants by means of some form of ion exchange mechanism (Birmingham and Colman, 1983). Since plants are important in removal of diquat from the water column, it is apparent that both susceptible (sago pondweed) and non-susceptible species (American pondweed) rapidly adsorb diquat (Hiltibran et al, 1972). In microcosms (250-mL flasks) containing *Myriophyllum spicatum* exposed to 0.03 to 3.0 ppm c.e. diquat for 2 to 7 days, the plants accumulated diquat at concentrations of 1,310 ppm c.e. (dry weight). Total plant collapse (death) can normally be expected 3 to 10 days after exposure (Lawrence et al, 1962). The available evidence indicates that susceptible plants do not degrade diquat (Funderburk and Lawrence, 1964 in Simsiman et al, 1976) and the metabolic fate of diquat in susceptible species has not been reported. However, bacteria associated with the surface of dead and dying plants appears to degrade about 32% of diquat to water soluble metabolites (Simsiman and Chesters, 1976). Although these metabolites have not been described, there is evidence that several species of bacteria including *Pseudomonas fluorescens*, *Rhizobium* spp. and

Aerobacter aerogenes are capable of degrading diquat. *P. fluorescens* is resistant to diquat at concentrations that would normally be found in the environment. After the decay of the plant, the release of any remaining diquat would have only limited effect on the concentration of diquat in the water column since it will be adsorbed rapidly by the sediment (Hiltibran et al, 1967; Dyson and Takacs, 2000, and Shaw et al, 1995).

Diquat is also adsorbed extensively by phytoplankton. Birmingham and Colman (1983) found concentrations of diquat in algae as high as 2,430 ppm (dry weight). *Chlorella vulgaris*, *Navicula pelliculosa, Anabaena flos-aquae* and mixed populations of *Spirogyra* sp. and *Cladophora* sp. adsorbed diquat to levels of 550, 2,430, 1,550 and 1,270 ppm c.e. dry weight. Since the application rate varied from 0.03 to 3 ppm c.e. in water, the bioconcentration factor would be at least 167 to 810. Other authors have reported that mixed populations of weeds adsorbed diquat at concentrations of 40 to 100 ppm (wet weight) (Rogers et al, 1992; Coats et al, 1966 and Way et al, 1971 in Birmingham and Colman, 1983). In spite of this tendency to bind to plants and filamentous algae, there have been no substantiated reports of diquat binding to unicellular algae which may explain the relatively high resistance of unicellular green algae reported by Melendez et al (1993).

There is no extensive non-plant bioaccumulation in the laboratory. The BCF was generally low for all species of invertebrate with the highest value of ~32 being found in the mayfly (*Cloen dipterum*). Bioconcentrations of as high as ~8.3 to ~10 were found in *Daphnia magna* and oysters and bioconcentrations ranging around 10 were found in freshwater clam. Only 30% of 14C-label is depurated from oysters after 28 days and with this level of bioaccumulation, a residue tolerance of 0.1 ppm c.e. may be exceeded in shellfish at the time of harvest (Fujie, 1987 and Thompson, 1987). Fish and crayfish generally had lower levels of bioaccumulation with whole body BCFs typically being less than 1.0. The bioconcentration was significantly lower in edible tissue (BCF = at 0.67) and significantly higher in the viscera (BCF = 2.5).

Field data and data designed to mimic field data on bioaccumulation is not extensive. However, after treatment of a pond in Wisconsin at 1.0 ppm, concentrations in bluegill sunfish were never higher than ~0.5 ppm 10 days after treatment and completely dissipated within 12 weeks (concentration <0.01 ppm). Treatment of the water body at the exaggerated use rate of 3.0 ppm c.e yielded BCF levels as high as 1~1.5 ppm after 10 days, which also completely dissipated within 12 weeks (Gilderhaus, 1967) (Table 5). These concentrations are somewhat higher than typical maximum use rates currently used to control aquatic weeds. However, more recent data has shown that at typical use rates, the concentration of diquat is below the detection limit in bluegill sunfish and was never detected at concentrations greater than 0.3 ppm c.e. in *Tilapia*. This relatively high concentration depurated to <0.1 ppm at 3-days after application (Fujie, 1987). In experiments designed to mimic field studies, two treatments of 1.2 ppm that were placed 2 weeks apart did not produce concentrations in edible bluegill sunfish tissue and edible channel catfish tissue of higher than 0.08 ppm c.e. In a similar experiment, crayfish were found to contain concentrations of 0.22 to 0.56 ppm and freshwater clams were found to contain concentrations of 0.33 to 14 ppm c.e. (Fujie, 1987).

Although the concentration of diquat can be higher than 0.1 ppm in fish tissue after 28 days of continuous exposure at ~1.0 ppm, depuration after placement in water not containing diquat is very rapid. In edible species, 50% of the adsorbed diquat is eliminated within 1.5 to 3 days and in non-edible species, depuration can be even faster with >80% of depuration occurring in 1 day. Since depuration is so rapid, magnification of diquat as it moves up the trophic levels is not likely.

In terrestrial domestic vertebrates, long term (90-day) feeding of animals with silage and hay that contained up to 50 ppm diquat caused no observable ill effects on several animals and no residues in tissues and milk were detected (Black et al, 1966 and Calderbank, 1968 in Simsiman et al, 1976). Drinking water supplied to calves for 30 days at concentrations of 20 ppm c.e. diquat also had no adverse impact (Howe and Wright, 1965 in Simsiman et al, 1976).

- **Persistence within the organism**

Most organisms do not bioconcentrate diquat and those that do rapidly eliminate the compound. Fish that adsorbed diquat from the water eliminated the majority (more than 50%) of diquat from their tissues within a few days (Schultz et al, 1995; Fujie, 1987; Hammer, 1987; Hammer et al 1987). A possible exception to this rate of elimination are bivalves which have been shown to depurate ~30% of 14C-labeled diquat and its degradates in 8 days (Thompson, 1987 and Fujie, 1987).

Over half of the diquat detectable in edible muscle tissue, skin, and kidney of channel catfish remained as unchanged diquat after 3 days of exposure to 14C-labled diquat. However, most of the residues found in liver and bile were metabolites of diquat. Similar experiments performed with goldfish (Beasley, 1966 in Simsiman et al, 1976) and diquat (Hiltibran et al, 1972) found that exposure to diquat for several days c.e. showed that 14C-labeled diquat and its degradates are distributed throughout all organs and tissues but accumulated to the greatest degree in the digestive tract. When fish were returned to clean water, the radioactivity was eliminated from all tissues and organs except the digestive tract. Furthermore, in field tests where pools were treated with 1.0 ppm c.e. 14C-diquat, and catfish were harvested 2 to 5 months later, no radioactivity was found in any tissues except the digestive tract.

In domestic animals, the bulk of the administered dosage (5 to 20 mg/Kg body weight) is eliminated from lactating dairy cow in the feces. Only very small amounts of the dosage are eliminated in the urine (2.6%) and milk (0.02%) after 7-days, primarily as metabolic products (Stevens and Walley, 1966 in Simsiman et al, 1976). In rats, 95% of the administered dosage is eliminated in the feces with about 70% of the eliminated dosage metabolized to presumably less toxic products by bacteria that can be found in the gastrointestinal tract. The remainder of the dosage (~5%) is eliminated in the urine (Daniel and Gage, 1966 in Shearer and Halter, 1980).

One day after the administration of diquat, only small amounts of residue are found in the muscles and other organs of calves. Rats fed 250 mg diquat/Kg body weight exhibited no accumulation of diquat in their tissues.

- **Potential impacts on the food chain**

Diquat has a tendency to accumulate in sediment and plants. The concentrations in sediment will persist for more than six months as unchanged diquat in sediment (Frank and Comes, 1967 and Simsiman and Chester, 1976). The concentrations in plants may also persist until the plants die and release diquat into the water column where it is rapidly bound to the soil. This phenomenon may be the reason that increases in diquat concentration in the sediment can occur for several months after treatment (Frank and Comes, 1967 and Gilderhaus, 1967). While sediment does not readily metabolize diquat, it is inactivated by adsorption to the sediment. Bacteria metabolize up to 32% of the diquat that is associated with dead and dying plants. Animals (terrestrial vertebrates) do not adsorb, and eliminate diquat after metabolizing most of it in their gastrointestinal tract. It is unclear to what degree diquat is metabolized by fish and aquatic invertebrates. However, it is apparent that fish and aquatic invertebrates do not accumulate diquat to levels that should be a problem. Fish and crayfish typically bioaccumulate diquat and its metabolites to levels that are less than the concentration found in water. Although invertebrates bioaccumulate diquat at concentrations of ~10 to ~30, this level of bioaccumulation will generally be rapidly depurated. Bioconcentration factors of less than 10 to 60 are not generally considered to be bioaccumulative. Therefore, animals which eat fish and aquatic invertebrates are not likely to biomagnify diquat as it travels up trophic levels. Higher animals (cattle, sheep and rats) do not bioaccumulate diquat. In rats, 95% of administered diquat is eliminated from the animal's body via the feces after the majority (70%) has been converted to metabolic products. In terrestrial domestic vertebrates (presumably cattle), long term (90-day) feeding of animals with silage and hay that contained up to 50 ppm diquat caused no observable ill effects and no residues in tissues and milk were detected (Black et al, 1966 and Calderbank, 1968 in Simsiman et al, 1976). Drinking water supplied to calves for 30 days at concentrations of 20 ppm c.e. diquat had no adverse impact on calves (Howe and Wright, 1965 in Simsiman et al, 1976).

Therefore, diquat should not bioaccumulate; it should be rapidly eliminated from most organisms that ingest it; and it should not be bioaccumulated (biomagnified) as it is passed up the food chain.

4.2.3 Potential Impacts of Water Quality on Survival of Aquatic Organisms

4.2.3.1 Effects of Physiological Sustaining Water Chemistry

Summary: Exposure of living plant tissue to diquat products or other herbicides usually results in secondary effects that may impact the biota. When plants start to die, there is often a drop in the dissolved oxygen content associated with the decay of the dead and dying plant material. Reduction in dissolved oxygen concentration may result in aquatic animal mortality or a shift in dominant forms to those more tolerant of anaerobic conditions. There may also be changes in the levels of plant nutrients due to release of phosphate from the decaying plant tissue and anoxic hypolimnion. Ammonia may also be produced from the decay of dead and dying plant tissue which may reach levels toxic to the resident biota. Ammonia may be further oxidized to nitrite (which is also toxic to fish), and the almost nontoxic, nitrate. The presence of these nutrients may cause an algal bloom to occur. Hardness and pH may have an effect on the toxicity of diquat. Since typical concentrations of diquat in the environment at the time of application are <0.224 ppm, most species will not be affected at either high or low water hardness. For example, the LC50 for diquat in several species of fish ranges between 35 to 125 ppm c.e. in hard water and 7.6 to 43 ppm in soft water. In most species of fish the toxicity in hard water is 2- to 10-fold lower than the toxicity in soft water.

- **Potential impacts of dissolved oxygen**

The key factor to survival and maintenance of the aquatic environment is normally adequate dissolved oxygen. The oxygen content of the water should ideally be as close to saturation as possible. For warm water environments (15 to 25°C) oxygen saturation is 10 mg/L at 15°C and 8.2 mg/L at 25°C. For cold water environments (5 to 15°C), oxygen saturation is 12.2 mg/L at 5°C and 10 mg/L at 15°C. Cold and warm water are somewhat arbitrary designations. Table 6 shows the sea level saturation concentration for oxygen at temperatures from 5 to 25°C.

Warm water fish like sunfish, bass, catfish, carp, and shiners can generally survive and reproduce at oxygen concentrations of about 5 mg/L (Litler, 1983, personal communications). However, while cold water fish are able to survive for short periods at dissolved oxygen concentrations as low as 1 to 3 mg/L, concentrations needed for long term survival are much higher. It is unlikely that these cold water species could go through a life-cycle at dissolved oxygen concentrations below 9.0 mg/L (Welch, 1992 in Shearer et al, 1996).

Treatment with diquat products has been shown to decrease dissolved oxygen content. Oxygen depletions are to be expected following application of diquat due to the bacterial breakdown of dead plants. This has been verified by a study conducted in a pond containing eutrophic hard water in Franklin, Wisconsin (Daniel, 1972) and Lake Marion, North Carolina (Inabinet, 1976). After application of Cutrine® plus diquat at concentrations of 2.2 ppm plus 3.0 ppm to microcosms placed in a pond, the dissolved oxygen concentration (DO) dropped from pretreatment DOs of 8 to 11 ppm to <1.0 ppm for 4 to 8 days. Surface water DOs returned to normal within 12 days of application but DOs at 30 and 90 cm depth took nearly 40 days to return to normal. These applications were effective in controlling Eurasian watermilfoil. However, adverse aquatic impact was seen after this application. One day after treatment, the entire resident fish population in this treated microcosm was dead or dying. It is not entirely understood if mortality was primarily due to the low DO, the toxic effects of the treatment, or a combination of both. However, it was noted that combined treatments with Cutrine®, diquat and Aquathol® at 2.2 ppm plus 1.5 ppm plus 1.5 ppm produced similar decreases in the DO, but no fish-kill was noted.

Work done by Inabinet (1976) was conducted after application of 2,4-D for control of water primrose and diquat for the control of American waterweed. It is not entirely clear what the application rate was for this experiment, but 30 tons of 2,4 –D granular formulation appears to have been applied on August 4-6, 1976 to 500 acres of Lake Marion, North Carolina. 1,500-gallons of diquat formulation appear to have been applied on August 27 after the overlying water primrose infestation had been controlled. During the course of the experiment, the dissolved oxygen levels dropped from 3.0 to 6.0 ppm on July 24 (before treatment) to 0.9 to 2.0 ppm at the surface and 0.5 to 1.2 ppm at the bottom on August 20 (after 2,4-D application). Seven days after treatment with diquat (September 2), the dissolved oxygen concentration continued to decrease with surface water concentrations of 1 to 2.7 ppm and bottom water concentrations at 0.3 to 0.6 ppm. These dissolved oxygen concentrations continued to remain low through September 11. However, by October 3, the dissolved oxygen concentrations had recovered significantly (2.8 to 3.8 ppm at the surface and 3.1 to 3.3 ppm at the bottom) and by November 5, the dissolved oxygen concentration had returned to pre-treatment levels.

The above field studies were conducted at rates that would typically be used to control Eurasian watermilfoil, American waterweed and pondweed. However, recent studies (Serdar, 1997) at Lake Steilacoom and Gravelly Lake, maximum application rates indicate that treatment with diquat does not necessarily result in a decrease in dissolved oxygen concentration. Lake Steilacoom was treated as a whole lake treatment at 0.13 ppm c.e.

diquat, and the dissolved oxygen content remained above 11.0 ppm during the 12 days of the study. Furthermore, Gravelly Lake was treated with diquat on its periphery at 0.11 ppm c.e. diquat, and the dissolved oxygen concentrations did not drop below 10.5 ppm during the 12 days of the study.

The drop in dissolved oxygen concentrations at Franklin, Wisconsin and Lake Marion, North Carolina has the potential to adversely impact the more oxygen sensitive organisms. However, the dissolved oxygen content at Lake Steilacoom and Gravelly Lake approaches saturation and effects of dissolved oxygen on even the most sensitive organisms should be minimal. Since the metabolic pathway in natural aquatic systems is not known, it is not known if the effects of low oxygen concentration affect the degradative pathway of diquat. It is reasonable to assume that it does affect the degradative pathway since both aerobic and anaerobic metabolism of diquat occurs on the surfaces of dead and dying aquatic plants.

- **Potential impacts of ammonia, nitrite and nitrate production**

It is rare when nitrogen is the limiting factor for production within a freshwater body. Several species of blue-green algae have the ability to fix nitrogen. Therefore, an addition of nitrogen to water bodies is not a major issue. However, the toxicity of ammonia and nitrites to aquatic organisms can be important. Experimental Cutrine® plus diquat treatments in microcosms (Daniel, 1972) produce marked increases in the levels of ammonia and nitrate/nitrite. On the day of treatment the concentration of ammonia in these microcosms was 0.03 to 0.05 ppm. Eight days after exposure (when the dead plants would be rapidly decaying), the concentration of ammonia in these treated microcosms increased to 0.380 to 0.490 ppm. By 16 days after treatment, the ammonia concentrations had dropped below the levels found in the microcosms prior to treatment. Similar observations were observed for nitrate/nitrite with concentrations of these oxidized nitrogens being non-detectable prior to treatment with diquat and rising to 0.030 to 0.060 ppm at 12 days after application. By 16 days after treatment, the nitrate/nitrite concentrations had returned to levels found prior to application of diquat. No algal blooms occurred for 240 days after treatment. However, at 240 days after application, there was a low level of algal regrowth that cannot be attributed to increases in ammonia or nitrate/nitrite concentrations. Secondary increases in ammonia concentrations occurred at 170 and 290 days after application while secondary increased in nitrate/nitrite concentration did not occur during the course of the study.

Although ammonia and nitrate/nitrite concentrations were not measured, at Lake Marion, North Carolina, phytoplankton density increased from 200 to 300 organisms per mL to 1,200 to more than 2,000 organisms per mL seven days after the application of diquat. This is the time frame that would be expected for the production of ammonia and nitrate/nitrite due to decay of dead and dying plants. This increase in numbers was likely due to a classical algal bloom since the phytoplankton densities returned to pretreatment levels in seven days to 1 month after peak phytoplankton densities were observed.

In Washington waters, even a small release of ammonia can be a serious issue. The whole lake levels of ammonia-nitrogen in Lake Steilacoom during the 1995 season exceeded the criterion of 0.100 mg/L during the months of May and October. These levels of ammonia are toxic to fish and near-shore runoff containing fertilizers may have contributed to the October ammonia peak (Shearer et al, 1996). These levels of ammonia are higher than the maximum recommended levels for the culture of aquatic organisms and are higher than the EPA criterion (0.091 mg/L) for 4-day exposure of salmonids. Since the concentrations of ammonia in the Franklin, Wisconsin microcosm study reach levels of nearly 0.5 ppm, there is potential for ammonia concentrations exceeding the criterion level or the maximum 4-day exposure level for salmonid exposure. However, in a study conducted during the 1996 season at Lake Steilacoom and Gravelly Lake, concentrations of ammonia and nitrate/nitrite only increased slightly after treatment with diquat at the maximum use rate. For ammonia, this increase was typically from concentrations of <0.010 ppm prior to treatment to concentrations of approximately 0.020 ppm 3 days after treatment at the maximum use rate. Concentrations of ammonia at these two Washington lakes returned to pretreatment levels at 12 days after treatment. The concentrations of nitrate/nitrite did not seem to be affected by treatment with diquat. However, the concentrations of nitrate/nitrite remained high throughout the 12 days of the study (0.329 ppm to 0.659 ppm). No changes water clarity that could be attributed to an algal bloom were seen at Lake Steilacoom or Gravelly Lake (Serdar, 1997).

The toxicity of ammonia increases with both temperature and pH. As temperature and pH increase, the amount of unionized ammonia increases (Table 7). The unionized forms of ammonia (NH3) are toxic to aquatic animals. The ionized form of ammonia ($NH4^+$) is almost harmless (Goldman and Horne, 1983).

Adsorption of nitrogen containing nutrients by aquatic macrophytes and algae can influence the seasonal dynamics of nitrite and nitrate concentrations. The levels of nitrite are often higher at the surface of a non-flowing water body than at the bottom because under anoxic conditions some bacteria utilize nitrate as a terminal hydrogen receptor when oxygen is not available.

Nitrite, although fairly toxic, is rarely a problem in well aerated waters because it is rapidly converted to nitrate and under anoxic conditions it is rapidly converted to ammonia. Nitrate is usually not toxic in the quantities found in lakes and rivers (up to 1 ppm). The drinking water standard is set at about 10 ppm). Polluted streams can contain up to 2 ppm of nitrite and small areas near the thermocline may contain relatively large quantities of nitrite.

If nitrogen is the limiting nutrient, nitrate can participate in the next algal bloom. Nitrate and nitrite are formed from the oxidation of ammonia and may persist long after algae and plants have utilized the ammonia in their biological processes. The next algal bloom can be due to the presence of nitrate. However, it may take several days from the time ammonia becomes in short supply for the next bloom to occur because nitrate uptake is slow relative to ammonia uptake and induction of nitrate reductase in algae is also fairly slow. Nitrate must be reduced to ammonia in algae prior to the initiation of an algal bloom
(Goldman and Horne, 1983).

- **Potential impacts of nutrient cycling and the release of phosphates and other plant nutrients**

  Phosphate is usually the limiting nutrient in aquatic systems. The sediment typically retains phosphorus under aerobic conditions and releases it under anaerobic conditions. This released phosphate may result in growth of phytoplankton in the hypolimnion provided the depth is not so great that photosynthesis is precluded. When plants are treated with diquat or other herbicides, they die and degradation of plant tissue by microbes can cause phosphate and other nutrients to be released. Most phosphorous in its organic form, cannot be utilized and must first be converted to phosphate (PO_4) by excretion and decay. Normally, phosphates rarely exceed 0.020 mg/L in the summer or 0.030 mg/L in the winter. Phosphorous depletion in the epilimnion is likely in freshwaters under normal circumstances during the growing season. Therefore, the treatment of a water body with diquat, which causes release of phosphates from the decaying tissue after the plants have died, has the potential to cause an algal bloom.

Similar to the effects of Cutrine® plus diquat treatment on the levels of ammonia and nitrate/nitrite, the concentrations of all forms of phosphorous including total phosphorous, total dissolved phosphorous, particulate phosphorous, ortho-phosphate and total dissolved organic phosphorus increased with respect to the control. For example, while orthophosphate concentrations remained low (<0.029 ppm) in the controls for the 370 days of the study, the concentration in the treated microcosm rose to ~0.230 ppm one day after treatment with diquat and remained at these levels till 80 days after treatment. Similar phosphate levels were observed for total phosphate and total dissolved phosphate. As with nitrogen, algal blooms did not seem to be correlated with concentrations of phosphate seen in the water column (Daniel, 1972). A similar release of phosphate and a correlated rise in the chlorophyll content was seen due to nutrient release after treatment of ponds with 1.0 ppm diquat for control of American waterweed (Peverly and Johnson, 1979).

Although phosphorous concentrations were not measured, Lake Marion, North Carolina phytoplankton density increased from 200 to 300 organisms per mL to 1,200 to >2,000 organisms per mL seven days after the application of diquat. This is the time frame that would be expected for the production and release of phosphate due to the decay of dead and dying plants. This was likely due to a classical algal bloom since the phytoplankton densities returned to pretreatment levels in seven days to one month after the peak phytoplankton densities were observed (Inabinet, 1976).

At Lake Steilacoom and Gravelly Lake, the concentrations of orthophosphate and total phosphorous did not appear to be correlated with the application of diquat for the control of American waterweed (*Elodea canadensis*) or pondweeds (*Potamogeton* spp.). No changes in the Secchi depth that could be attributed to an algal bloom were seen at Lake Steilacoom and Gravelly Lake (Serdar, 1997).

The only other nutrient, frequently in short supply, is iron. Ferric iron may either react with or be adsorbed with phosphate into the sediments under typical aerobic conditions and become biologically unavailable. Under anaerobic conditions, ferrous iron is formed from ferric iron/phosphate complexes and is released into the hypolimnion where plants may utilize it for growth provided that the light is sufficient for photosynthesis to occur. Eh (oxidation/reduction potential), pH and DO (dissolved oxygen content) govern this reaction. The heterogeneous nature of water/sediment phase reactions prevents easy extrapolation of laboratory results to real lake sediment systems. Iron availability may limit the growth of algae in lakes and streams especially when the production of ammonia (due to nitrogen fixation) is the limiting factor in algal growth (Goldman & Horne, 1983 and Reid, 1961).

Nutrient cycling will include a bloom of algae, which ends when one of the nutrients and/or other factors becomes in short supply. As the algae dies it releases phosphates, iron and ammonia through the degradative process. When enough of the nutrient in shortest supply becomes sufficient to sustain growth, algae will start growing again in the lag phase and will result in an algal bloom if conditions of temperature, pH, N: P ratio and iron concentration are adequate to sustain a log phase growth.

- **Potential impacts of pH changes**

  The pH of most natural waters falls between 4 and 9. A pH of 7 is neutral, neither acid nor basic. One important way in which pH is controlled is by removing carbon-dioxide from the water. A pH of greater than 8 in a lake or pond is probably due largely to a high rate of photosynthesis. Anthropogenic causes of high pH may be due to enrichment of the water with fertilizers containing organophosphates. If the pH of a lake or pond is low (<6) it is likely due to leaching of organic acids from peat, and anthropogenic sources such as acid rain or leachate from mines. Bottom waters are typically lower in pH than surface waters because bacterial respiration and decomposition of organic matter produces carbon-dioxide and organic acids which lower pH (Shearer, 1996).

  After aquatic macrophytes die, due to either a natural process or treatment with an herbicide, the pH may drop. If an algal bloom occurs after the release of nutrients, the pH may rise due to the removal of carbon-dioxide from the water column by photosynthesis. A pH greater than 9 can be directly lethal to fish. Toxicity to high pH levels arises from the inhibition of ammonia secretion by gills and respiratory alkalosis (Heath, 1995 in Shearer, 1996). Sub-lethal alkaline or acidic conditions can indirectly harm fish and other aquatic animals by increasing their susceptibility to other stresses such a pollutants (like diquat), ammonia, high temperatures and low dissolved oxygen.

  Hardness can have an effect on the toxicity of diquat. Although not directly connected to pH, hardness can have an effect on the toxicity of herbicides. Hard waters due to the presence of calcium and magnesium have a tendency to be alkaline (basic) while soft water due to the presence of low calcium and magnesuim levels has a tendency to be acidic. This appears to be true for diquat also. However, the concentration of diquat that kills fish in hard water is typically much higher (5- to 10-fold higher) than that found to kill fish in soft water. For example, diquat is more toxic by 10-fold to fathead minnow in soft water than in hard water (Soft water LC50 = 7.6 ppm c.e.; hard water LC50 = 70 ppm c.e.); diquat appears to be more toxic to grass carp by about 4-fold in soft water than in hard water (soft water LC50 = 29 ppm c.e.; hard water LC50 = 125 ppm c.e.); diquat appears to be more toxic to bluegill sunfish by about 2-fold in soft water than in hard water (soft water LC50 = 39 ppm c.e.; hard water LC50 = 76 ppm c.e.). However, this may not be so for all species. For example, the toxicity of diquat to common carp appears to be about the same in soft water and hard water (LC50 = ~40 ppm) (Table 8).

  In Washington State, hard waters with higher pH are generally found in Eastern Washington lakes (relative to Western Washington lakes). Diquat appears to be more toxic to most species of fish in soft water than in hard water. Since most fish are unaffected by diquat concentrations that are likely to be encountered in the environment, this observation is largely of no practical concern. However, for those species and stages like striped bass sac-fry and walleye fry with a soft water and hard water (LC50 of 0.54 ppm and 0.75 ppm, respectively), potential remediation of toxic effects could be seen if diquat was used in hard water only when these species are present in their most sensitive stages.

4.2.3.2 Effects of Diquat in Water

Summary: In the state of Washington, pesticide residues that exceed the federal drinking water standard (MCL) have not been found in public drinking water for most counties east of the

Cascade Mountains. Although diquat is persistent, it binds irreversibly to soil. Soil bound diquat does not appear to be biologically available to aquatic organisms in surface water. Therefore, the U.S. EPA does not require a surface-water advisory and it is unlikely that diquat would have significant impact on surface water used for domestic or agricultural purposes (EPA RED, 1995). Because the soil distribution coefficient is greater than 15.0 on all sediments and soils tested, it is not believed that diquat is likely to leach into the ground water. Therefore, recharge areas are not at risk for diquat leaching into the ground water. Downstream surface- water treatment plants may experience concentrations of diquat higher than the ederal drinking water standard (0.02 mg/L). In order to avoid using water that is above the Federal drinking water standard, a set back distance of 1,600 feet from a water intake located in a river must be observed; a set back of 400 to 1,440 feet from a water intake must be observed in the treated area of reservoir that is 0.32 and 50.2 acres, respectively; and a set back distance of 400 feet from a water intake must be observed in a treated canal. Diquat is not likely to be found in the water of sewage outfalls since waste water treatment plants only process water from household waste and water runoff from street level. Due to the short dissipation half-life, high levels of water exchange and dilution of diquat in water bodies, additional procedures for removing diquat from outfalls or potable water systems is probably not necessary. However, the addition of montmorillonite clay, or bentonite clay to water contaminated with diquat may remove it from the water to levels where it is safe to drink (Shearer and Halter, 1980 and Coats et al, 1966).

According to Scott Fink (2000, personal communications) of the Spokane Department of Health, Drinking Water Division, herbicides have never been detected in a surface water system at concentrations that exceed the federal drinking water standard in any Washington State county east of the Cascades. Also, in eastern Washington, there has never been a herbicide detection that exceeds the EPA’s Drinking Water Criterion in public well water. The current MCL for drinking water is less than 0.02 mg/L for diquat products. However, there have been a few cases where herbicides were found in well water at concentrations that exceed Washington State’s detection limits.

- **Potential impacts on recharge areas**

  In light of the above findings, it is unlikely that diquat will have an adverse impact on sensitive well recharge areas. Because of its extremely high adsorption coefficient and its tendency to bind irreversibly to sediment and soil, diquat is unlikely to have an adverse impact on either surface or ground water. The soil distribution coefficient (Kd) is typically 15 for sand sediment, 36 to 42 in sandy soils and 1,882 to 10,740 in sandy loam, sandy clay loam and loam soils respectively. Kds of this magnitude would normally classify diquat as immobile. Since diquat is stable after binding to soil it is unlikely to be converted to more mobile degradates. Therefore, the EPA does not require a surface-water advisory and diquat is not likely to leach to the ground water through aquatic, riparian or palustrine recharge areas (EPA RED, 1995). Generally speaking, Coats et al (1966) has found that in all of the natural soils tested that diquat was not leached below 0.5 inches when 10 acre-inches of water were applied to a three-inch soil column. This is further evidence that diquat will not leach to the ground water under any condition likely to be encountered in the environment.

  Using these observations, the modeling work done by Travis (1994) indicates that diquat is immobile in saturated sediments and subsoils. Therefore, use of diquat as an aquatic herbicide will not and cannot result in the concentration of the diquat in surficial aquifers in Florida. Florida appears to be a worst case scenario, and it is unlikely that groundwater would be contaminated by the use of diquat as an aquatic herbicide in other states of the U.S.A.

- **Impact of pesticide application on downstream water treatment plants**

  Recent modeling work on the effects of diquat on downstream water treatment plants has been conducted (Ritter et al, 1995 and Singh and Williams, 1997). Under a worst case scenario based on modeling the most conservative view on the diquat application, bed sediment type, bulk density of bed sediment, sediment-water partition coefficient, fraction of organic carbon content in sediment, plant mass in treatment area, plant water partition coefficient, longitudinal dispersion coefficient, horizontal dispersion coefficient, vertical dispersion coefficient, stream velocity on the riverbank, stream velocity in the river center, water half-life, plant and sediment half lives, set back distances and waiting periods required to ensure safety of water to be used for domestic purposes was determined. For the maximum use rate of 2.0 gallons /acre, the set back distance from a potable water intake is 400 feet for reservoirs and 1,600 feet for rivers. If this set back distance were not attainable, it would be necessary to wait eight hours before using water from a treated reservoir or 48 hours before using water from a treated river.

  Singh and Williams (1997) came to a slightly different conclusion. They found that a set back distance was not necessary in reservoirs where the treatment area was less than $\leq$ 0.32 acre and if water was to be taken from the treatment area and used for household purposes that the waiting period must be at least one hour. However, a set back distance of 1,440 feet was necessary if the treated area of a reservoir was 50.2 acres; if this set back distance is not obtainable, the waiting period before using water for household purposes is 20 hours. In a canal, the set back distance should be 400 feet from a potable water intake. No mention was made of a waiting period if this set back distance from the water intake could not be obtained.

- **Presence of pesticide in the outfall**

  Because concentrations of diquat are low at water intake pipes only three or four days post treatment, the amount of diquat in the outfall of drinking water or waste water treatment plants is likely to be negligible. Since wastewater treatment plants only process water from household waste and water runoff from street level, diquat from treatment of lakes, ponds, streams, and irrigation canals will not be present in the outfall (Jim Milton, Ecology CRO Manager of Sewage Treatment Plant Permits, verbal communication, 2000).

- **Need for additional procedures to remove pesticide from the outfall**

  In cases where the concentration of diquat or paraquat is higher than the federal drinking water standard, it is feasible to clean up the water by adding montmorillonite clay or bentonite clay to the contaminated water. Due to the high number of cation exchange sites on these agents, diquat and paraquat are likely to be removed to levels where the drinking water standard is not likely to be exceeded (Shearer and Halter, 1980 and Coates et al, 1966). However, such procedures should rarely be necessary due to a short dissipation half-life, and relatively low chronic toxicity to aquatic wildlife.

  Although as yet not proposed, the use of biofilms or chemostats containing bacteria that are known to degrade diquat also has potential for mitigating cases of diquat water pollution. Species of bacteria known to degrade diquat include *Pseudomonas fluorescens*, *Rhizobium* spp. and *Aerobacter aerogenes*.

4.2.4 Mixtures with Other Pesticides and Incidental Presence of Other Pesticides

Summary: Aquatic herbicide tank mixes have not been evaluated in an Evironmental Impact Statement and are therefore not permitted in Washington State. However, when liquid diquat products are used to control floating aquatic weeds, low levels of non-ionic surfactants can improve the efficacy of liquid diquat products. If surfactants are used, care should be taken to use non-ionic surfactants that are registered for aquatic use since they have low toxicity to fish.

There are some claims that combinations of diquat and certain copper products can be useful in increasing the spectrum of algae and macrophyte species controlled, allowing for better adsorption of diquat by removing certain epiphyte algae (Komeen® label, Nautique® label, Captain® label and Cutrine®-Plus label). The potential for a more than additive effect in combination with diquat is also claimed by these labels against Hydrilla verticillata*. Diquat may also synergize the effects of copper sulfate by allowing some species of aquatic macrophytes to more readily adsorb cupric ion (Sutton et al, 1970).*

There have been reports that carbaryl and diquat, and nabam and diquat are synergistic in their effects against some species of aquatic vertebrates. High concentrations of carbaryl and diquat have been reported to be synergistic in toxicity to mosquito fish (Krieger and Lee, 1973 in Shearer and Halter, 1980). However, concentrations of diquat and nabam at environmentally relevant concentrations have a synergistic effect on the development of African clawed toad (Anderson and Prahlad, 1976). While diquat is frequently mixed with endothall, paraquat and copper products, these mixtures do not typically produce higher than additive effects. However, combinations of Cutrine® and diquat at 0.162 ppm copper plus 5.5 ppm diquat have been noted to produce higher than additive toxicity to brown trout (Simonian and Skea, 1977); and mixtures of Cutrine® at 2.2 and diquat at 3.0 ppm have been note to produce a complete fish-kill in semi-

field tests when these concentrations were not likely to affect survival when used alone (Daniel, 1972).

There have been no substantiated reports of reduced effectiveness when diquat is used with other pesticides

There have also been reports of diquat and triclopyr exhibiting additive effects on the control of waterhyacinth and waterlettuce (Langeland and Smith, 1993).

There has been some discussion of possible cumulative effects of diquat in its effects on higher animals. Since diquat damages higher animals by the formation of free radicals, there is a potential for irreversible and cumulative cellular damage to occur with exposure to diquat. This has potentially serious ramifications for mutation effects, carcinogenic effects and the production of cataracts. Although Shearer and Halter (1980) note potential cumulative effects, no experimental evidence has been presented to prove these assertions.

Formulations of diquat may act in combination with other pesticides under three scenarios: 1) Applied as a mixture; 2) Broadcast in separate applications (e.g., areas where pesticides are applied for mosquito and aquatic vegetation control), or 3) Accidentally combined as a result of over-spray in marginal areas or of run-off from neighboring areas treated with different products. Herbicide mixtures may result in antagonistic, synergistic, additive or cumulative effects (same herbicide applied more than once). It should be noted that tank mixes of aquatic herbicides are not permitted in Washington State for control of aquatic weeds.

Because very little work has been done on the effects of pesticide combinations, it is unclear whether pesticides applied for other purposes could substantially enhance the toxicity and persistence of diquat.

In the state of Washington, diquat products are rarely mixed with other products. A number of nonionic surfactants are registered for use with water-soluble ionic herbicides like diquat when they are applied to floating or emergent plants. Generally, when a liquid pesticide is applied to floating or emergent vegetation a surfactant and/or drift control agent should be used.

Only one formulation of diquat (Reward® LA) is typically used for the control of aquatic weeds in public waterways. Therefore, at the current time the effects of different formulations on aquatic organisms are not relevant. Although Reward® LA does not appear to impact most species of fish and aquatic invertebrate adversely, this is not a guarantee that other formulations of diquat that may become registered for use in public waterways will not adversely impact these species. Different "inert materials" and contaminants in formulations from different manufacturers may interact with the pesticide to give antagonistic, additive, cumulative or synergistic effects against target species (aquatic weeds and algae) and non-target fish and aquatic invertebrates. It is not necessary to use adjuvants with subsurface injections of diquat. However, in other states, a thickener is often used with liquid products to allow the treatment to sink more deeply into the water column where it can be most effective; and non-ionic surfactants are recommended when the species of weed to be controlled is emergent or floating.

- **Adjuvant effects**

When liquid diquat products are used to control floating weeds like pennywort, Salvinia, waterlettuce, or waterhyacinth, or emergent weeds like cattail by direct contact with a spray, the use of a non-ionic surfactant and a thickening agent is recommended. The non-ionic surfactant should be used to allow for better wetting of floating or emergent weeds and the thickening agent should be used to prevent drift. There are a number of non-ionic surfactants registered for aquatic use in Washington State, although only R-11, X77 and LI700 are allowed under the EIS for aquatic use at this time. Most surfactants should be mixed at 0.25% to 0.5% by weight of application solution when diquat is being applied to floating (surface) aquatic macrophytes. The toxicity of these adjuvants to bluegill, rainbow trout and daphnia has been well DOumented. None of these aquatic adjuvants should be toxic to fish or aquatic invertebrates when applied at labeled rates. However, Watkins et al (1985) noted that some aquatic adjuvants have a potential to be toxic to aquatic organisms when applied in shallow water. For example: 1) If Spar-Mate® is applied at the labeled use rate to water with a depth of less than 1.5 meters, it can be toxic to bluegill sunfish. 2) If Cide-Kick®, X-77®, Formula 403®, or IVOD® are applied at the labeled use rate to water with a depth of less than 0.1 meters, they may be toxic to fish. Since the depths given are for concentrations of the adjuvant that will kill 50% of the treated animals, an additional safety factor of ~10-fold would need to be added to assure safety of the adjuvant to the biota. Lee and Furtado (1977) have also recommended these safety factors controlled by depth specifically for diquat when controlling *Salvinia molesta*. Details of the toxicity and depth considerations for a number of aquatically applied adjuvants can be found in Table 9. Although adjuvants are typically considered to be "nearly inert", they are not entirely inert. However, adjuvants labeled for aquatic use should not be subacutely, acutely or chronically toxic to fish or other aquatic animals. Adjuvants can enhance, diminish, or have no effect on the activity of herbicides. Because ionic surfactants may react with ionic herbicides, they should not be used with diquat, which has an organic cation as the toxic agent. Although acute aquatic testing has been done on a number of adjuvants, insufficient data exists on the toxic effects of adjuvants when mixed with herbicides and applied to the aquatic ecosystem.

No specific surfactants are specified in the Reward® LA label. Therefore, experts should be consulted prior to the use of a specific non-ionic surfactant with diquat for the control of floating of emergent weeds.

- **Antagonistic effects**

Antagonism is defined as a less than additive effect when using pesticides in combination with each other. There are no specific studies that indicate diquat is antagonistic with other pesticides. However, ionic-surfactants must be avoided since they may react with or be antagonistic to the action of diquat dibromide.

- **Additive effects**

Langeland and Smith (1993) have reported that mixtures of diquat and triclopyr are strictly additive and that no interaction between these two herbicides has been observed for the control of waterhyacinth or waterlettuce. However, mixtures of these two herbicides have been recommended because triclopyr at 3.3 Kg a.e. /ha controls waterhyacinth and diquat at concentrations as low as 0.14 Kg c.e./ha controls waterlettuce.

Diquat is often used in combination with endothall, paraquat or copper compounds in order to control various species of weeds. These combinations, while generally additive in effect, do not appear to be supra-additive; that is, they are not synergistic (Shearer and Halter, 1980 cite Berry, 1975; Water Investigation Branch, 1977 and Yeo and Dechretz, 1976).

- **Cumulative effects**

  Although Shearer and Halter (1980) pointed out that the mode of action of diquat on higher animals has the potential to produce cumulative effects, there is no concrete evidence that this occurs. Diquat's mode of action on higher animals is due to the formation of free radicals. Free radicals have been shown to cause cumulative effects with respect to mutations, cancer and cataracts when other free radical forming compounds or free radical forming radiation are tested. Although the mode of action of diquat makes it a suspect in causing cumulative effects, no credible demonstration of these cumulative effects have been seen (Shearer and Halter, 1980). The only confirmed chronic effect of diquat in higher animals is the production of cataracts in rats and dogs and additional work would be necessary to show cumulative or lack of cumulative effects.

- **Synergistic effects**

  There are few reports of potential synergism of diquat by other pesticides. The potential for synergism with copper sulfate pentahydrate has been shown when it is combined at 1.0 ppm copper plus 0.1 to 2.0 ppm c.e. diquat. At these concentrations, *Hydrilla verticillata*, *Egeria densa* and *Naja guadalupensis* accumulate copper ion at concentrations that are significantly higher than in the absence of diquat (Sutton et al, 1970). This improved adsorption of cupric ion could possibly explain the better than additive control of *Hydrilla verticillata* claimed by the Komeen®, Cutrine®-Plus, Nautique® or Captain® labels when these products are combined with diquat at the labeled use rates.

  True synergistic effects with diquat in combination with nabam fungicide or carbaryl insecticide have been claimed. Diquat and nabam at concentrations ranging from 0.41 ppm c.e. diquat and 2.0 ppm nabam to 0.68 ppm diquat and 2.0 ppm nabam) have been reported to have synergistic effects on development and survival, growth and behavioral effects in both larvae (tadpoles) and embryos of the African clawed toad (*Xenopus laevis*)(Anderson and Prahlad, 1976). Although this species is not relevant to Washington waters, the treatment concentrations are environmentally relevant and therefore, of potential concern to more sensitive species that may be found in Washington waters.

  The mosquito fish (*Gambusia affinis*) is synergistically affected by concentrations of diquat (17.5 ppm and 275 ppm) in combination with carbaryl (10 ppm). It is likely that microsomal fraction oxidases (MFOs) are inhibited by diquat, which is a one electron transfer agent, with potential to synergize a variety of anthropogenic toxins. However, since the combination of carbaryl and diquat is only synergistic at concentrations well above the maximum use rate, it is unlikely that this combination of pesticides will adversely impact the aquatic environment.

  Diquat and Cutrine® in combination has been seen to affect fish adversely in both the laboratory and semi-field situations. Diquat plus Cutrine® at concentrations of 5.5 ppm c.e. plus 0.162 ppm copper has been seen to cause greater than additive mortality in the brown trout (Simonin and Skea, 1977). In the semi-field situation, diquat plus Cutrine® has been observed to cause complete fish-kills at concentrations of 3.0 ppm plus 2.2 ppm respectively (Daniel, 1972). These concentrations of diquat or Cutrine® alone should only affect the most sensitive species of fish. Therefore, it is possible that the low dissolved oxygen concentration in this test could have contributed to an observed fish-kill.

4.2.5 Potential Impacts on Agriculture

Summary: At typical use rate concentrations, single irrigation events or flooding of crops with water treated with diquat is not likely to cause damage to most edible crops. Diquat has been used as an aquatic herbicide for over 20 years without a single case of phytotoxicity being reported due to the effects of irrigation water. Most crops irrigated at up to 0.5 ppm are not likely to be damaged. Irrigation studies with water containing 0.01 ppm diquat have shown no evidence of phytotoxicity or residues higher than the U.S. FDA tolerance. At much exaggerated exposure rates(>0.45 ppm), no evidence of damage or residues higher than the allowable tolerance was seen in potatoes, sorghum, soybeans, carrots, lettuce, or onions (White 1994 and Gangstad, 1986). Despite the safety margin for use of diquat on crops appearing to be very high (>50-fold), it is not advisable to treat crops with irrigation water that has been treated with 4.0 gallons of Reward ® LA/acre more than one-time per season unless the water has been restricted from irrigation use for at least 5 days.

If water use restrictions are followed as described in Section 1 and the Federal Use labels, there should be no impact on agriculture. The Reward® LA label (2000) does not permit the use of diquat to control weeds in water to be used for irrigation, agricultural sprays, watering dairy animals or domestic water supplies for a period of time specified on the label. The water use restriction period for Reward® LA, used at the maximum use rate (2 gallons/acre), is 3, 1, 3 and 5 days for drinking, livestock consumption, irrigation of ornamentals, and irrigation of food crops, respectively. However, if an U.S. EPA/FDA approved method of analysis is used, diquat treated water may be used for any purpose if the concentration is below 0.02 ppm c.e.

There are very strong indications that diquat will not persist at concentrations greater than 0.02 ppm c.e. for more than 5 days even under EPA's worst case scenario. Typical concentrations after five days are <0.004 to 0.027 ppm c.e. in Florida ponds. Typical concentrations seen after treatment (Lake Finley, New York, Lake Chautauqua, New York, ponds in Colorado, ponds in North Carolina, Lake Marion in South Carolina, Miller Pond in Illinois, Guntersville Reservoir in Alabama, Gravelly Lake and Lake Steilacoom in Washington) range from non-detectable (<0.004 ppm) to ~0.06 ppm (Sewell, 1969; Frank and Comes, 1967; Rogers, 19992 and Serdar, 1997). Concentrations are occasionally seen that are in the range of 0.5 ppm, but this typically occurs when the treatment concentrations are ≥1.0 ppm (Gilderhaus, 1967 and Hiltibran et al, 1972). The persistence can be strongly affected by the application rate. However, modeling work indicates that typical spot treatments with diquat on the perimeter and middle of lakes in the Northwest will not exceed a four-day average of 0.021 ppm c.e. when the application rate is 4.0 lbs c.e./acre (4.48 Kg c.e./ha) (Ritter et al, 2000). Spot treatments are very common while whole lake treatments with the maximum use rate, as was done in Steilacoom Lake in 1996, are rather uncommon but can yield concentrations of diquat as high as 0.059 ppm c.e. after three days of dissipation. However, whole pond treatments with the maximum use rate can yield modeled concentrations with a four-day average of 0.014 ppm (Ritter, 2000) while the maximum concentration based on a simple first order half-life can be as high as 0.059 ppm. A five-day water use restriction for irrigation is considered to be adequate for almost all scenarios. However, in order to provide 100% assurance that the concentration of treated irrigation water does not exceed 0.02 ppm, residue analysis would be necessary.

Although a worst case scenario is given above, more typical persistence based on EPA's worst case scenario would allow for a four-day average environmental concentration in pond water of 0.014 ppm (Ritter, 200). Other modeling work by Ritter et al (1995) and Singh and Williams (1997) would indicate that water use restrictions in reservoirs need be only one hour when the treatment area is 0.32 acres or 20 hours when the treatment area is 50.2 acres. The recommended water use restriction period for rivers based on Ritter et al's (1995) model is 48 hours. These water use restriction periods were determined based on standard models that assume the worst case situation likely to be encountered in the United States.

A water use restriction is not necessary if the distance from the treatment area to the water withdrawal pipe is 400 feet in a reservoir where the treatment area is only 0.32 acre, 1,440 feet where the treatment area is 50.2 acre, 400 feet in a canal or 1,600 feet in a river.

- **Potential impacts of water on irrigation**

  If water used for irrigation contains less diquat than mandated by the MCL (0.020 ppm c.e.), diquat should not have an adverse impact on crops irrigated with treated water. According to Scott Fink (Public Health Department: Drinking Water Division, 2000 personal communication), the levels of herbicides in public drinking water have always been below the current MCL.

  In general, broad leaf plants are susceptible to diquat, while grasses and grains are resistant. Diquat is therefore toxic to many non-target broadleaf plants, including both crops and native vegetation. Adverse effects depend on rate of application, number of applications over a confined period and relative susceptibility of individual species. Plant susceptibility to diquat has been qualitatively described for a number of aquatic weeds and terrestrial crops (Campbell, 2000; White, 1995, and Gangstad, 1986).

  The use of diquat as a weed killer is described on the Reward® LA label, with appropriate water use restrictions specified for specific applications. If the water use restrictions are followed, minimal damage should occur to non-target native and crop species. However, because diquat is a non-specific broadleaf herbicide, it may adversely affect some crops and other non-target species exposed to irrigation water containing the active ingredient.

  Many species of plants may survive repeated exposure to diquat, if key periods where they are more or less susceptible are considered. These periods include germination (more susceptible), seedling (more susceptible), dormancy (less susceptible) and aging (less susceptible) periods. Other factors include plasticity, seed dispersal, hardiness and tolerance (Ebasco, 1993). For additional information please see Section 4.3.3.2.3.

  Indications are that when water containing up to 0.01 ppm c.e. diquat was used to water various crops, the maximum residues of diquat in each crop were either non-detectable or lower than the FDA tolerances for these crops (White, 1994). However, Gangstad (1986) found that even when crops were irrigated with concentrations $\geq$0.45 ppm c.e., no residues exceeding the FDA tolerances were found and presumably no significant crop damage was observed. These crops and their residue levels were potatoes (<0.05 ppm), grain sorghum (<0.1 ppm), soybeans (<0.1 ppm), carrots (<0.05 ppm), lettuce (<0.05 ppm), and onions (<0.05 ppm). It is noteworthy that the rates of diquat in irrigation water of 0.45 ppm to 2.25 ppm described by Gangstad (1986) are much higher than would typically be encountered in the field. However, White (1994 citing Calderbank) concluded that phytotoxicity damage is unlikely in crops irrigated with water containing $\leq$0.5 ppm. Although this provides a 50-fold safety factor, it is not recommended that crops be irrigated with water containing these high levels of diquat more than once per growing season (Shearer and Halter, 1980 and Ecology, 1992) (Table 10).

- **Potential impacts of water used to water livestock**

If water used for watering dairy animals contains less diquat than mandated by the MCL (0.020 ppm c.e.), diquat (applied as Reward® LA) should not have an adverse impact on the animals or milk production (Simsiman et al, 1976). In general, diquat is rapidly dissipated in natural waters by adsorption to sediment and plant tissue and experiments done to mimic EPA's worst case scenario contained concentrations of <0.004 to 0.027 ppm c.e. after 5 days of dissipation (Fujie, 1988). Concentration of diquat under typical use scenarios rarely falls outside the range of <0.004 ppm to 0.060 ppm (Fujie, 1988 and Serdar, 1997). Diquat has an LC50 to rats of > 600 mg test material /Kg body weight and is therefore not significantly toxic to rats (EPA RED, 1995). The toxicity to birds for diquat acid ranges between 61 and 90 ppm c.e. for the most sensitive species. Diquat is therefore moderately toxic to birds (EPA RED, 1995). After oral dosing, ^{14}C-labeled diquat is rapidly eliminated from the body of rats with 90% to 95 % of the residues eliminated in the feces (Daniel and Cage, 1966 in Shearer and Halter, 1980, and EPA RED, 1995). Almost the entire remainder of this dose is eliminated in the urine. The bulk of the residues (70%) detected in rat feces had been eliminate as metabolic products.

In studies with sheep, 60% of the diquat ingested with silage could not be accounted for, indicating that it was eliminated from the animal as volatile [probably respiratory products (Simsiman et al, 1976)]. Concern has been expressed over the distribution of diquat in animal tissues. In chickens fed ^{14}C-labeled diquat at 32 ppm c.e. (36X the maximum dietary burden) for 4 days, the total radioactive residue found in eggs and tissues was <0.058 ppm c.e. Typical tissue concentrations were <0.001 ppm c.e. for egg yolks, 0.004 ppm c.e. for egg whites, 0.004 ppm c.e. for fat, 0.003 ppm for muscle, 0.042 to 0.058 ppm for kidney and 0.030 to 0.045 ppm for liver. The bulk of these tissue concentrations were diquat *per se* and not metabolites. Concentrations of diquat and metabolites found in the feces was not discussed but is presumed to have been relatively high based on the low concentrations of ^{14}C-labeled diquat and its metabolites in eggs and tissues (EPA RED, 1995).

Lactating dairy cows were dosed at 5, 8 and 20 ppm ^{14}C-labeled diquat from a drenching bottle. No ^{14}C-labeled residues were found in leg muscle of cows dosed at 5 ppm ^{14}C-labeled diquat and the concentration of residues in milk was <0.002 ppm c.e. in cows dosed with 20 ppm ^{14}C-labeled diquat. Total residue concentrations in a bullock dosed with 8 ppm were 1.071 ppm, 0.033 ppm, <0.04 ppm in kidney, liver and other tissues, respectively. The concentration of residue that could be attributed to diquat and its metabolites were 0.03 and <0.01 ppm in kidney and liver, respectively. In a similar experiment, the bulk of the residues were found in feces and 2.6% of the residues were found in the urine while less than 0.02% of the residues were found in the milk (Stevens and Walley, 1966 in Simsiman et al, 1976). Based on the rat data and general comments found in the reviewed articles, it is believed that the residues consisted primarily of metabolites due to the action of bacteria that would typically be found in the gastrointestinal tract. Concentrations of ^{14}C-diquat and metabolites found in the feces was not discussed, but is presumed to have been relatively high based on the low concentrations of diquat and its metabolites in milk and tissue (EPA RED, 1995).

Since the residues of concern are diquat *per se*, it is not likely that cattle or chickens will accumulate enough diquat residue to effect the human consumer (EPA RED, 1995) or cause biomagnification as it moves up through the trophic levels to man.

Although the above information applies to food ingested by livestock, no toxic signs were seen in calves or sheep supplied with drinking water that contained 20 ppm diquat for 30 days (Howe and Wright, 1965 in Simsiman et al, 1976).

- **Potential impacts of water used for agricultural sprays**

  Crops were irrigated with water that had been treated with diquat at rates much higher than would normally be expected to occur. Typical concentrations of diquat in soil or sprinkler irrigation water were >0.45 ppm c.e. No significant residues were noted in crops irrigated with water containing 0.45 to 2.25 ppm diquat. Crops were irrigated only once in either early or late stage of maturity. The irrigation rate was 1.0 acre-inch and the water was delivered by sprinklers or through furrows. Although it was not discussed, since a crop was available for harvest, it is presumed that the concentrations of diquat in the irrigation water did not cause significant damage to the crops (Table 10). Use of overhead irrigation can probably be equated with the effects of using water for agricultural sprays. However, sprinkler irrigation does not take into account the potential antagonistic, additive or synergistic effects that might occur due to the presence of other pesticides, or adjuvants (surfactants, accelerator, thickeners, et cetera); and these effects have been known to occur with other herbicidal products. See Section 4.2.4.

- **Potential impacts on fishing and the consumption of fish**

  Diquat is not expected to cause adverse impact on recreational or commercial fishing. In addition, when the Guntersville Reservoir, Alabama was treated with diquat at the maximum use rate of 2.0 gallons formulation per acre (0.3 to 0.4 ppm c.e.) no significant fish-kill was mentioned (Rogers et al, 1992, Bain and Boltz, 1992,). Even when the treatment rate was as high as 1.0 ppm in ponds in Wisconsin, no significant fish- kill was seen (Gilderhaus, 1967). In similar experiments conducted in Nigeria, no significant fish-kill was seen after treatment with 2.0 Kg a.i./ha (Olaleye et al, 1992).

  It has been previously shown that diquat does not extensively bioaccumulate or bioconcentrate in fish (Section 4.2.2.4). Diquat bioconcentration studies have been conducted in the laboratory on several animal species. Diquat has been tested in the laboratory for bioaccumulation in bluegill sunfish, channel catfish, crayfish, freshwater clams and oysters. These species were typically exposed to ~1.0 ppm c.e. except for oysters that were exposed to ~0.10 ppm c.e. During exposure of 28 days, fish typically did not accumulate diquat in their edible tissues. The residues of diquat typically did not exceed 0.7 ppm in muscle tissue (Hammer et al, 1987) and were typically less than this value in studies designed to mimic the dissipation of diquat seen in field studies (Fujie, 1987). In these simulated field studies, concentrations seen in edible bluegill and channel catfish tissues were typically <0.01 ppm. c.e. after one 14-day exposure to diquat. Similar studies conducted with clams and crayfish yielded residue concentrations of 0.40 and 0.07 ppm c.e., respectively after one 14-day exposure to diquat. Residue tolerance levels for fish are 0.10 ppm c.e. and therefore it is unlikely that people will be exposed to concentrations of diquat that exceed the residue tolerance. However, the concentrations in shellfish demonstrate that the diquat tolerance of 0.1 ppm may be inadequate. Field studies to determine residue levels were poorly designed since the limit of detection was 1.0 ppm for fish, shellfish, plants and sediment and the measured residues were <1.0, <1.0, 100 and 2.16 ppm, respectively 48, 48, 48 and 144 hours after treatment at 0.4 ppm diquat (Rogers et al, 1992).

Similar field tests with diquat were conducted with bluegill sunfish. When these fish were exposed to initial concentrations of 1.0 ppm c.e., the concentrations in water were ~0.14 ppm 3 days after treatment, ~0.45 ppm 10 days after treatment and ~0.12 ppm 21 days after treatment. The high concentrations of diquat at ten days after treatment are difficult to understand since the concentrations in water were low 10 days after treatment (~0.14 ppm) in field studies conducted in Wisconsin ponds (Gilderhaus, 1967) (Table 5). However, release of diquat from dead and dying plants into the benthic region could have impacted the concentration of diquat found in fish.

In well designed field studies, the concentrations of diquat in edible catfish tissue was always below <0.1 ppm c.e. Similarly, in field tests done with *Tilapia*, the concentration of diquat in edible tissue was 0.3 ppm c.e. one day after treatment and 0.1 ppm c.e. three days after treatment (Fujie, 1987).

In laboratory metabolism studies where fish were exposed to 1.0 ppm c.e. ^{14}C-labeled diquat, total residues in edible trout or carp tissue did not exceed 0.01 ppm in edible tissue and 65% of this residue was diquat *per se*. This would lead to diquat concentrations of <0.0065 ppm c.e. (EPA RED, 1995). It is noteworthy that these diquat tissue concentrations do not exceed the tolerance even at exposure concentrations, which slightly exceed the expected maximum use rate concentration of 0.75 ppm c.e.

Diquat is rapidly eliminated from the edible tissue of fish. The elimination half-life is usually less than three days. Therefore, exposure to diquat is not very likely if any time is allowed for depuration prior to reopening the fishery. However, current label guidance indicates that fishing and eating of fish caught from a treated water body should be allowed with no time restrictions and since the current diquat tolerance in fish is 0.10 ppm c.e., this will usually be an acceptable practice. However, data from shellfish experiments indicate that a tolerance of 0.1 ppm c.e. may not be adequate for the consumption of shellfish fished from water treated at 1.2 ppm c.e.

- **Potential impact of air quality on crop plants and livestock**

  The Reward® LA label states that "The pesticide should only be applied when wind is blowing away from adjacent sensitive areas (e.g. residential areas, bodies of water, known habitat for threatened or endangered species and non-target crops)". This implies that vapors from this product or contact with the spray drift may injure susceptible plants in the immediate vicinity. The Reward® LA label further warns, "Applications should not occur during temperature inversions because drift potential is high. Temperature inversions restrict vertical air mixing, which causes small suspended droplets to remain in a concentrated cloud. This cloud can move in unpredictable directions due to light variable winds common during inversions. Temperature inversions are characterized by increasing temperatures with altitude and are common on nights with limited cloud cover and light-to-no wind. They begin to form as the sun sets and often continue to morning. Their presence can be indicated by ground fog; however, if fog is not present, inversions can also be identified by movement of smoke from a ground source or an aircraft smoke generator. Smoke that layers and moves laterally in a concentrated cloud (under low wind conditions) indicates an inversion, while smoke that moves upward and rapidly dissipates indicates good vertical air mixing." This implies that diquat should not be applied when weather conditions favor drift from target areas, as this product may injure non-target plants. Most terrestrial plants are susceptible to the effects of diquat and even a 5% drift can result in 0.2000 lbs cation/acre being deposited outside the target when the maximum use rate is applied. Even with low drift, corn, sweet corn, wheat, cotton, and soybeans may be adversely affected. For example, treatment rates as low as 0.0047 to 0.016 lbs/acre may cause 25% damage to these sensitive crop species (EPA RED, 1995) (Table 11). The main methods of using Reward® LA largely preclude the effects of drift. This liquid product is either injected by subsurface methods (which precludes drift) or applied as large droplets at low pressure which mitigates the effects of drift. It is also recommended that a thickening agent be used to control drift when applying liquid herbicides to the water surface. However, even small amounts of drift can be an issue if many swaths are applied, and particularly if the product is applied from and aircraft (Forsythe et al 1997).

Due to the low vapor pressure of commercial products of diquat (2.4 x 10^{-7} mm/Hg at 25° C for the active ingredient diquat), diquat products should have very little tendency to effect air quality or cause crop damage. The mode of application is usually subsurface injection for liquid formulations making drift outside the treatment area unlikely. For those cases where a boom sprayer applies a liquid formulation, as much as 1% of the application may drift out of the treatment area. It has been estimated for general herbicides that this amount of drift could have an impact if 120 swaths were applied and 1% of the applied pesticide drifted out of the treatment area on each pass. In this case, dosage levels higher than that intended for the target could accumulate downwind of the treatment area. This could cause an effect on non-target plants that may damage habitat and decrease the amount of forage available for waterfowl and fish in non-target areas (Forsythe et al, 1997). In cases where aerial application might be necessary, as much as 17% of the treatment would not strike the target area. In aerial application, drift out of the treatment area could impinge on non-target organisms at a great distance from the site of application. Depending on how much diquat was deposited per unit area outside the site, there could be a significant impact on non-target wild plants or crops. In addition to effects on plants, non-target sensitive terrestrial wildlife may be adversely impacted.

Odor is unlikely to be noticed except for short periods of time following application of diquat. Posting and communication requirements specified in the aquatic weed control permit should make the public aware of any potential odor problems and how long the odor problem will exist. Since there are rarely more than one or two applications of diquat per water body per year in the state of Washington, any adverse impact on quality of life due to problems with odor from diquat applications should be weighed carefully with the impact on quality of life due to the effects of poor navigability, and effects on the recreational use of the water body. Direct effects from breathing the vapor are unlikely for the general public. However, there have been cases of applicator exposure to the vapors when the applicator was known to have passed through a cloud of vapor during treatment. Typical effects that were seen from inhalation of the vapor by an applicator include inflammation and bleeding of the nasal mucosa, myalgia, periorbital headache, and a cough productive of thick, red sputum. A few days later, the patient may become confused, have neck stiffness, a cough, and a high fever. Consolidation of exposed lung tissue is also likely. Approximately one week after exposure, the fever may continue to be high, and wet rales and rashes on exposed skin may be apparent. A slight jaundice may be apparent. Treatment with prednisone is recommended with rapid improvement apparent and chest X-rays normal after 5 days (Shearer and Halter, 1980 cite Wood et al, 1976).

- **Potential impact of flooding on agriculture**

  Flooding of agricultural land with diquat-treated water should be a rare occurrence. When flooding occurs, dilution effects should mitigate the effects of the concentration of diquat. Flood irrigation is typically practiced with very few crops. Although the effects of soil irrigation are similar to those of the sprinkler irrigation described above, flood irrigation has the potential to cause damage to a crop even if soil and sprinkler irrigation at 1 acre-inch does not. Further research to address this point may be necessary.

- **Potential impacts on aquaculture**

  Under most conditions, it is not anticipated that the use of diquat should have acute effects on aquaculture when the concentration of diquat is below 1.0 ppm c.e. The target concentration for diquat in aquatic use is usually 0.2 to 0.75 ppm (Shaw et, all 1995) but can be as low as ~0.1 ppm c.e. (Serdar, 1997). The most typical use rate in the United States is 0.224 ppm to 0.37 ppm c.e. (Ritter et al, 2000). Target concentrations in Gravelly Lake and Lake Steilacoom, Washington were 0.11 to 0.13 ppm after application to a water body with a depth of 10 to 11 feet. At Lake Steilacoom, the concentration of diquat was measured as 0.033 to 0.091 ppm during the first three days after application but the concentration fell below 0.007 ppm c.e 12 days after application. Since only the Southern and Northeast margins of Gravelly Lake were treated, the measured concentration of diquat at one to three days after treatment was 0.0006 to 0.0028 ppm c.e. The concentrations in Gravelly Lake are unlikely to cause acute aquatic impact to fish or aquatic invertebrates. However, the concentrations in Lake Steilacoom may be high enough to affect the most sensitive invertebrate that has been tested; *Hyalella azteca* has a 96-hour LC50 of 0.048 ppm c.e. Water is not generally used for agricultural purposes until the concentration in the treated water body falls below the MCL (0.02 ppm c.e. for diquat). If water used for aquaculture is taken in through surface intake pipes, the concentration will be in the range of 0.02 ppm if the setback distances specified by Ritter et al (1995) and Singh and Williams (1997) are followed. Please see section 4.2.3.2 for information on setback distances for surface water intake pipes. Fish and invertebrates will be more sensitive to diquat when exposed chronically than when exposed acutely. However, chronic exposure is generally not considered to be a serious issue because the rate of dissipation of diquat from the water column is very rapid (Campbell et al, 2000).

  Diquat has been used to treat disease in fish in hatcheries, and for the species tested; it did not affect the breeding rate of fish or cause moralities of juveniles (Gilderhus 1967). Rates of 1ppm diquat applied up to 3 times and 3 ppm applied once or twice, with 8-week intervals between applications, had no adverse effect on hatching and growth rates of bluegills in seven different pools. Channel catfish fry were not affected at 10 ppm diquat and bluegill fry were not affected at 4 ppm diquat. Largemouth black bass fry were more sensitive and were affected at levels greater than 1.0 ppm at 22° C (Jones 1965).

  Because the concentrations of diquat that acutely effect the most sensitive commercial species run from very low to very high (0.54 ppm c.e. for striped bass sac-fry to 141 ppm c.e for 2-year old common carp), diquat is not likely to adversely impact cultured aquatic organisms since the exposure concentration of properly used Reward® LA would typically not be higher 0.02 ppm c.e. In order to protect these commercial species from the effects of acute toxicity, the maximum concentration would have to be five to ten-fold lower than the lowest LC50 of 0.54 ppm c.e. Since and the expected environmental concentration (EEC) at the point of water withdrawal would be $\leq$ 0.02, fish in fish hatcheries should be protected from the adverse effects of diquat. However, for the most important commercial species (salmonids), the safety margin would even be higher than the thirty-fold demonstrated for striped bass sac-fry since the LC50s for these desirable species range from 6.1 ppm c.e. for rainbow trout fingerlings to ~20 ppm for brown trout fingerlings and Coho salmon yearlings (Table 2 and Table 10).

Although chronic exposure is likely to produce an MATC somewhat lower than the acute LC50, inadequate research has been done to determine the MATC for most commercial species. The chronic MATC has been determined for fathead minnow and channel catfish to be 0.2 and >1.0 ppm c.e., respectively, but chronic MATCs for rainbow trout have not been determined. However, the chronic LC50 for rainbow trout is 2.1 ppm which is well above the maximum acute EEC (0.02 ppm) value likely to be found at a water intake pipe (Table 2 and Table 19).

4.3 ENVIRONMENTAL TOXICITY REVIEW – DIQUAT TOXICITY TO THE BIOTA AND RISK ASSESSMENT

Diquat products are not chronically toxic to most aquatic life except for their direct contact effect on plant foliage and algae. However, formulated diquat is acutely toxic to some species of algae (7-day EC50 = 0.050 to 0.175 ppm c.e. for blue-green algae; 0.047 to 0.48 ppm c.e. for green algae; 0.022 to 0.127 ppm c.e. for freshwater diatoms; >2.94 to 3.24 ppm for marine diatoms, and 6.5 to 43 ppm c.e. for marine green algae) and aquatic macrophytes (14- to 16-day EC50 = 0.0072, ~0.02, ~0.12, 0.023, 0.046, 0.08, 0.1, 0.11 and ~0.01 ppm c.e. for *Potamogeton natans*, *Lemna minor, Azolla caroliniana, Elodea canadensis, Myriophyllum spicatum, Hydrilla verticillata, Eichhornia crassipes and Pistia stratioites and Spirodella polyrhyza,* respectively) (Tables 13 and 17). All environmentally relevant species of fish appear to be tolerant to diquat with the most sensitive stage/species being sac-fry of striped bass and the least sensitive stage/species being 1-2 year old common carp with a 96-hour LC50 = 141 ppm c.e. Important fish to the Northwest include 8- to 10-day walleye fry, fingerling rainbow trout, fingerling brown trout and fingerling to yearling Coho salmon with 96-hour LC50s = 0.75, 6.1 to 18, 18 to 20 and 21 to 30 ppm c.e., respectively (Table 18). Most species of aquatic invertebrates are not susceptible to the acute effects of diquat; 48-hour and 96-hour LC50s are typically greater than 5.0 ppm [mayfly (96-hour LC50 = 16 ppm c.e, lined scud (96-hour LC50 = 19 to ~50 ppm c.e.), Odonata (96-hour LC50 = >100 ppm c.e., bloodworm (96-hour LC50 = >100 ppm c.e.), juvenile eastern oyster (96 hour EC50 = 141 ppm), and various ostracods, copepods and marine shrimp (48-hour LC50 = 2.0 to ~25 ppm c.e.)]. There have been reports that the marine amphipod (*Hyalella azteca* and the water flea (*Daphnia magna* and *Simocephalus vetulus*) are highly susceptible to diquat with 96-hour and 48-hour LC50s = 0.048 and <1.0 ppm c.e., respectively (Table 14). This may be of concern since concentrations of diquat shortly after application to surface waters (0.224 to 0.37 ppm) are often higher than the EC50 for these sensitive species (Shaw et al 1995, and Campbell et al 2000). However, the usual half-life for diquat is approximately 0.75 days. This value is based on the EPA's worst case scenario of 0.224 ppm in a farm pond (EPA RED, 1995, Campbell, 2000 and Shaw et al, 1995). The initial concentration may also be toxic to algae and plants since the 7- and 14-day EC50s are less than 0.224 ppm [except for *Euglena* (EC50 = >2.94 ppm c.e.), *Chlorella vulgaris* EC50 = ~0.3 to 0.4 ppm, c.e. and marine green algae and diatoms (EC50 = >2.94 to 43 ppm c.e.)]. However, after 4 to 7 days of dissipation the expected environmental concentration (EEC = 0.059 to 0.034 ppm) of diquat is lower than the typical concentration that effects most species of algae. Possible exceptions could be various species of diatoms like *Chryptomonas ozolini* (7-day EC50 = 0.019 to 0.075 ppm c.e.)

and *Ochramonas danica* (EC50 = 0.022 to 0.124 ppm), and the green algae, *Selenastrum capricornutum* (EC50 = 0.026 to 0.48 ppm c.e.). However, since the range of concentrations that affect these species encompasses risk quotients that are both less than and more than one it is not anticipated that algae will be adversely impacted by diquat for protracted periods of time.

All tested species of macrophytes would be affected adversely for 7-days and duckweed, giant duckweed, and American pondweed would probably be affected adversely for at least 14 days since the 14-day EC50 is less than the 14-day EEC. Species may start to recover in 14 days including common duckweed, waterhyacinth, Hydrilla and Eurasian watermilfoil since the 14-day EC50 is greater than the 14-day EEC. Fish and most aquatic invertebrate habitat will be protected since in all cases except the for *Hyalella azteca* (96-hour EC50 = 0.048 ppm c.e.), the 4-day LC50 or EC50 is usually greater than 10 times the 4-day EEC (0.059 ppm). The presence of sediment may be a mitigating factor, which can raise the EC50 for *Hyalella azteca* to 6.8 ppm c.e.

Diquat's effectiveness may be improved on floating emergent weeds by adding nonionic surfactants and accelerators so that diquat is more readily adsorbed. Diquat products are primarily applied from boats using a spray boom or subsurface injection. Diquat liquid products may occasionally be applied from a shore vehicle using a spray boom. It is unusual for diquat to be applied by aircraft except for application to remote sites. Aerial application is usually avoided due to public perception that drift problems may have an adverse impact on the human habitat (Getsinger, 1999, personal communications). A drift of only 5% (0.2 lbs/acre) could have significant impact on susceptible crop plants (Table 13) which are affected at the EC25 level at concentrations as low as 0.0047 to 0.016 lbs/acre. The activity of liquid formulations of diquat may be improved by adding a thickening agent to assure that subsurface applications drop lower in the water column where they can do the most good or by decreasing drift when the formulation of diquat are applied by a spray boom.

The acute effects of diquat are not of major concern. However, possible problems could occur with food chain issues, disruption of habitat (Fronda and Kendrick, 1986; Tatum and Blackburn; 1962, Hillsenhoff, 1966; Berry et al, 1975; Gilderhaus, 1967; Newbold, 1975 and Fujie, 1987), potential disruption of nesting (breeding behavior) in cladoerans (Gilderhaus, 1967 and Newbold, 1975) and maturation behavior in salmon (Lorz et al, 1979). Some sub-lethal effects cause avoidance to treated areas by invertebrates (Folmar, 1978) and fish (Berry, 1984), as well as a change in swimming speed or rheotaxic response in fish (de Peyester 0& Long, 1993 and Dodson and Mayfield). Sub-acute effects which disrupt biochemistry or respiration in fish have been seen by Holtz and Winter (1989), El-Deen (1992 & 1993) and Bimber et al (1975) and may cause pathogenic conditions. Changes in numbers, diversity and quality of aquatic macrophytes and animals associated with them (Hillsenhoff, 1966; Tatum and Blackburn; 1962 Newbold, 1975; and Olaleye et al, 1992) have also been observed.

Due to the mode of action (inhibition of photosynthesis) diquat can take from a few days to about 2 weeks to control aquatic weeds, particularly if low rate technology is an issue. It has been suggested that much lower concentrations than are typically used for control of aquatic weeds may be effective for the control of more sensitive weeds like sago pondweed, coontail, American waterweed and southern naiad (Hulbert, 1987 and Tatum and Blackburn, 1965). These species are typically completely controlled at 0.5 and 1.0 ppm c.e. but effective (95% control) is obtainable at 0.25 ppm c.e. Concentrations of diquat sufficient to control milfoil are typically high enough to adversely impact most aquatic plants. Furthermore, concentrations effective against milfoil will normally not be effective for longer than two months in Northwest waters.

Therefore, in the Northwest, more than one treatment per season may be necessary to control milfoil (Water Investigations Branch, 1977 in Shearer and Halter, 1980). While sago pondweed is effectively controlled by diquat (Hiltibran et al, 1972), certain species of pondweed, including American pondweed (Hiltibran et al, 1972) Richardson's pondweed (Reward® LA Label), reed canarygrass, white clover, *Chara* spp., *Nuphar* spp., *Nymphaea* spp., duckweed or curlyleaf pondweed are either not controlled by diquat or are controlled only for very limited periods of time (<6 weeks after application) (Eady and Renney, 1965; Ecology, 1992; Hulbert 1987; Tatum and Blackburn, 1965).

Biochemical degradation of diquat is not extensive. Soil bacteria metabolize diquat rapidly to at least three water soluble metabolites. The toxicity of diquat to microorganisms is fairly high. Diquat concentrations as high as 100 ppm caused an initial decrease in the number of bacteria. However, after 12 to 24 hours, this concentration of diquat stimulated bacterial growth to over that seen in controls. This is probably due to selection against susceptible microorganisms and selection for microorganisms that have become resistant to diquat and are now able to utilize it as an energy and carbon source. It is unknown if this is primarily a laboratory phenomenon or whether diquat at levels used for aquatic plant control would stimulate the natural bacterial populations (Yazar et al, 1975 and Camper and Shively, 1974 in Shearer and Halter, 1980). Some of the soil sediment microbes that are known to metabolize include *Pseudomonas fluorescens*, *Rhizobium sp.* and *Aerobacter aerogenes*. *Pseudomonas fluorescens* can tolerate diquat up to 2,400 ppm c.e. Once diquat has been adsorbed to soil, it is unavailable for degradation by bacteria. The rate of degradation on soil varies with the type of soil; diquat adsorbed to kaolinite clay was more susceptible to degradation than diquat adsorbed to montmorillonite clay. Diquat adsorbed to montmorillonite in aqueous nutrient suspensions was not degraded microbially and remained intact for a period of one year (Weber, 1972 in Simsiman et al, 1976).

In an experiment using four species of aero-aquatic hyphomycete fungi, it was found that *Pseudoaegerita matsushimae*, conditioned leaves in water exposed to concentrations of up to 20 ppm c.e. diquat so that the pond snail *Lymnaea elodes* was able to utilize the leaves and fungus for food. However, other species of fungus including *Helicoon elegans, Beverwykella pulmonaria,* and *Hormiacitis ontariensis* were not able to condition leaves in water exposed to diquat at more than 5.0 ppm c.e. Such leaves were not palatable to the pond snail, thus disrupting the food chain and nutrient uptake for those individuals that depended on fungi other than *Pseudoaegerita matsumishimae* to condition their food and detoxify diquat for them (Frondra and Kendrick, 1986). Although conditioning of leaves by *P. matsumishimae* reduced the toxicity of diquat to the pond snail, reduced survivorship was observed at concentrations as low as 1.0 ppm c.e.

Plants do not appear to metabolize diquat directly. It is apparent that alligatorweed (an aquatic plant) and beans (a terrestrial plant) do not metabolize 14C-diquat. In these species the only significant radiolabel seen after extracting the plants is diquat. Diquat is typically degraded by bacteria associated with aquatic plants and up to 32% of this diquat can be metabolized to water soluble metabolites by the bacteria that are associated with dead and dying plants (Simsiman and Chesters, 1976). Residues of diquat are not usually found at high levels because diquat is photolytically degraded to TOPPS and pyridene compounds which are further degraded to picolinic acid and volatile fragments (Black et al, 1966 in Simsiman et al, 1976). This degradation occurs only if sufficient time between spraying and harvest is allowed. If the crops are harvested within a few days of foliarly diquat application, concentrations of diquat on the surface of the plant can be very high [0.02 to 4.0 ppm (Calderbank et al, 1968 in Chester et al, 1976)]. In small grains, residues are often very high since cereals are desiccated when diquat comes into direct contact with them. It is likely that the herbicide adheres to grain (0.5 ppm to 4.0 ppm for barley, up to 1.3 ppm for wheat, 0.7 to 5.0 ppm for rice, and up to 0.2 ppm for sorghum). Diquat residues in corn and root crops are usually much lower than for small grains (0.02 to 0.05 ppm for onions, up to 0.04 ppm for potatoes and rarely detected in corn).

Animals do not appear to directly metabolize diquat. However, 70% of the diquat that passes through the gastrointestinal tract can be metabolized by bacteria associated with the gut. Total 14C-labeled residues found in muscle (<0.003 ppm c.e.), liver (<0.05 ppm c.e.), kidneys (<0.05 ppm c.e.), eggs (0.004 ppm c.e.) and milk (0.077 ppm c.e.) is usually less than the residue tolerance of 0.02 to 0.05 ppm even when the treatment levels are much higher than the expected body burden. Note that the concentration of diquat *per se* is much lower than the total residue concentration with diquat concentration being 0.002 ppm c.e. in milk, 0.003 ppm in cattle kidney, <0.01 ppm in cattle liver and no residues in cattle muscle tissue. A more detailed discussion of the residue issue in chickens and cattle can be found in section 4.2.2.4.

4.3.1 Effects and Selectivity on Aquatic Plants

Summary: Diquat is toxic to most algae and macrophytes when applied at the maximum use rate of 0.75 ppm. Almost all species of algae or plant have EC50s that are less than this initial concentration of diquat used at the maximum use rate. Only Chlorella vugaris *(EC50 = 0.395 to > 2.94 ppm c.e.),* Euglena gracilis *(≥2.94 ppm c.e.), the marine diatoms (3.24 ppm c.e.), and marine green algae (6.48 to 43 ppm c.e.) are not affected by this concentration of diquat when exposed for short time periods). According to Peterson et al, (1994), this concentration inhibits growth in all algae and macrophyte species tested by at least 50% and more typically 70% to 100%. This level of inhibition gives a risk quotient (RQ) higher than the level of concern of 1.0 with a presumption of very high risk to non-target algae and aquatic macrophytes. The most resistant algae/plant species not discussed in this paragraph is* Lyngbya wollei *blue-green algae with a 3- and 7-day EC50 of ~0.15 ppm, which yields a risk quotient of 5 (RQ = 0.75 ppm/ 0.15 ppm). The most susceptible algae species not yet discussed above is the pyrrophyte (*Peridinium cinctum*) with a 7-day EC50 of 0.001 ppm, which yields a risk quotient of 750 (RQ = 0.75 ppm c.e./0.001 ppm c.e.). Therefore, it is likely that both target and non-target aquatic algae and plant species will be adversely impacted by short-term exposure to diquat.*

The most typical exposure concentration under EPA's worst case scenario is 0.224 ppm c.e. applied to a 200-cm deep pond at 4.48 Kg c.e./ha. Short-term exposure to this concentration of diquat is likely to adversely impact algae and aquatic macrophytes since the risk quotient is greater than the level of concern (RQ 2.0 = 0.224 ppm c.e./0.15 ppm c.e. or RQ = 224 = 0.224 ppm c.e./0.001 ppm c.e.). Even if the 4-day geometric EEC is 0.059 ppm c.e., assuming a half-life of 0.75 days, all macrophyte species will be adversely impacted since the 4-day risk quotient is greater than the level of concern of 1.0 for a very insensitive species (RQ = 1.28 = 0.059 ppm c.e./ 0.046 ppm c.e. for Eurasian water milfoil). Most species of algae will not be adversely impacted since the EC50 for 3 to 7 days is generally higher than the EEC. Since less than 20 % of the algae species are adversely impacted (at very high risk in all tests), control and adverse impact on most species of algae in the field is unlikely.

Laboratory results are similar to the effects observed in field studies where unicellular and filamentous algal blooms were observed to occur 7 to 16 days after treatment with diquat in excess of 1.0 ppm (Cooke, 1977 and Inabinet, 1976). This is likely to be a secondary effect due to *the degradation of macrophytes and the release of phosphate and/or ammonia/nitrate/nitrite that occurred in the same time frame.*

Treatment with 1.0, 0.5 or 0.25 ppm c.e. diquat caused nearly complete clearance of a variety of waterweeds and some species of filamentous algae in field and semi-field tests. For example, 0.5 ppm to 1.0 ppm c.e. has been seen to eliminate or strongly inhibit the growth of watermeal and the aquatic species listed in the label. Diquat has also controlled Eleocharis acicularis, Lemna minor *and* Callitriche *sp., watermeal (*Potamogeton richardsonii*), water stargrass,* Mougeotia *spp.* green algae *and* Zygnema *spp., and net algae (*Hydrodictyon *sp.) at rates of 0.5 to 1.0 ppm. Rates of 0.25 ppm c.e. diquat will control most target species of aquatic weeds and algae. However, rates of up to 0.5 ppm c.e. diquat may be necessary to control dense growths of these species. Field studies indicate that treatment with diquat plus Komeen® at 0.3 ppm c.e. plus 0.3 ppm copper for control of aquatic vegetation does not effect the numbers, size, condition or movement of largemouth bass. However, this may be a reflection of the small size of the treated areas (5 to 10 acres).*

Most species of algae are not strongly affected by the use of diquat. However, species like Spirogyra *spp.,* Pithophora *spp.,* Piridinium cictum, *and* Chryptomonas ovata *which are adversely affected by exposure to diquat will be, at least temporarily, eliminated from the population with a subsequent effect on diversity. Although most algae are affected at higher concentrations of diquat (1.0 to 30.0 ppm), the filamentous and unicellular green forms appear to increase significantly (13- to 19-fold), which can affect the diversity of the species present (Melendez et al, 1993). However, these high concentrations may not have environmental relevance since they are higher than typical treatment rates of 0.11 to 0.37 ppm used to control aquatic weeds. Various species of protists are adversely impacted as to diversity at concentrations of as low as 0.3 c.e. diquat. While these effects can last as long as 21 weeks in static laboratory microcosms, the presence of sediment can reduce this decrease in protist diversity to just 1 week. Certain species of macrophytes appear to be unaffected by diquat including* Chara *spp.,* Nuphar *spp.,* Nymphaea *spp., and Richardson's pondweed. Furthermore,* Myriophyllum *spp., duckweed (*Lemna *spp.), curlyleaf pondweed and Robinson's pondweed (*P. robertsonii*) are not controlled for periods of time (<2.0 months) for the use of diquat to be considered practical for their control (Johnson, 1962 and Hulbert, 1987). These resistant and difficult to control species may become dominant in a treated water body after the use of diquat if other control measures are not taken.*

Diquat is a fairly non-selective broadleaf herbicide, which also affects certain monocotyledonous plants. However, the labeled use for diquat in aquatic ecosystems is limited. Diquat is used in Washington at low concentrations (~0.1 ppm c.e.) for the control of pondweeds (*Potamogeton* spp.) and American waterweed (*Elodea canadensis*); control of these species by diquat has been achieved at both Lake Steilacoom and Gravelly Lake (Serdar, 1997). However, it also has utility in the control of other species, i.e. *Myriophyllum* spp*., Ceratophyllum demersum* (coontail) and *Utricularia* spp. (bladderwort), Naiad (*Najas* spp.), *Hydrilla verticillata,* and the filamentous algae (*Spirogyra* spp. and *Pithophora* spp.) at 1 to 2 gallons per acre when applied as a subsurface or bottom placement application. For the control of floating and emergent aquatic weeds like waterlettuce (*Pistia stariotes*), waterhyacinth (*Eichhornia crassipes*), pennywort (*Hydrocotyle* spp.), frog's bit (*Limnobium spongia*), or *Salvinia* spp. control may be obtained with an application of 0.5 to 0.75 gallons formulation per acre when applied to floating foliage; higher application rates (1 to 2 gallons formulation per acre may be necessary to control duckweed (*Lemna* spp.) or cattails (*Typha* spp.) (Reward® LA label, 2000). The aquatic macrophytes currently of greatest concern in the Northern Tier of States (including Washington) are *Myriophyllum spicatum* (Eurasian watermilfoil), *Potamogeton crispus* (curlyleaf pondweed), *Egeria densa* spp. (Brazilian elodea), Monoesius hydrilla*,* *Spartina alterniflora* (smooth cordgrass), *Lythrum salicaria* (purple loosestrife), *Phragmites australis* (common reed), and *Nymphaea* spp. (water lilies). Of these, only Eurasian watermilfoil and curlyleaf pondweed are effectively controlled with a Reward® LA according to its label. However, Robinette (1998-1999) and Westerdahl and Getsinger (1988) also claim good to excellent control with diquat or diquat plus complexed copper products against *Brazilian elodea*.

Treatment under an experimental use permit at Lake Steilacoom and Gravelly Lake had a multi-fold purpose: 1) To control aquatic weeds (American waterweed and pondweed); 2) Determine the persistence of diquat in the water column; 3) Assess the acceptability of the product label restriction in Washington; 4) Assess the degree to which diquat drifts from the treatment area; 5) Evaluate the accuracy of a model currently used to predict diquat concentrations in water. It was unclear as to the level of control that was achieved at these lakes (Serdar, 1997). In a few field studies, diquat was noted to clear treated lakes of aquatic weeds when applied at concentrations of 0.5 ppm c.e. Treatment at 0.5 ppm cleared Spooner Pond in Wisconsin of vegetation, including *Potamogeton richardsonii*, *P. pectinatus*, *Eleocharis acicularis*, *Lemna minor*, *Callitriche* spp. and the filamentous algae (*Mougeotia* and *Zygnema*) (Johnson, 1962) and no vegetation had re-grown even two months after treatment. In a similar treatment in a pond in Broward County, Florida, southern naiad (*Najas guadalupensis*) was cleared from the pond by the seventh day after treatment and no re-growth was seen by the end of summer (Tatum and Blackburn (1965). In semi-field plots conducted in plastic pools in Alabama, treatment with 0.5 ppm c.e. diquat cleared pools of waterhyacinth (*Eichhornia crassipes*), pondweed (*Potamogeton* spp.), water stargrass, American waterweed (*Elodea canadensis)*, Naiad (*Najas* spp.), an *Azolla* within 1 to 2 months after treatment and over 90% control was obtained within two weeks of application (Lawrence et al 1965). However, a number of species were not controlled adequately or were resistant to diquat including alligatorweed *(Alteranthera philoxeroides*), (Lawrence et al, 1962), *Chara* spp. (Johnson, 1962 and Tatum and Blackburn, 1965), *Nymphaea* spp., *Nuphar* spp. (Shearer and Halter, 1980) and the filamentous algae (*Vaucheria dicotoma)* (Robson et al, 1976 in Shearer and Halter, 1980). Some species of macrophytes may be controlled only for brief periods of time. For example: duckweed may be controlled for only 4 weeks at concentrations of diquat up to 0.5 ppm c.e.; and curlyleaf pondweed may be controlled for only 6 weeks at concentration up to 1.0 ppm (Hulbert, 1987). American pondweed has also been noted to be tolerant of diquat treatment (Hiltibran et al, 1972). The watermilfoils, including parrotsfeather and Eurasian watermilfoil (*Myriophyllum spicatum*), may be controlled for two weeks to a month but often are no longer controlled after two months (Lawrence et al, 1962 and Shearer and Halter, 1980). The case of Eurasian watermilfoil is especially alarming since this failure to control for more than two months has been observed in Washington waters including Union Bay where concentrations of diquat high enough to achieve temporary control of watermilfoil, killed almost all other aquatic plants.

Most species of aquatic plants are entirely cleared from ponds at 0.5 ppm c.e. However, this may not be the most economical rate since 0.25 ppm c.e. diquat will control these same species at the 95 percent level except in the case of very heavy infestations (Johnson, 1962).

4.3.1.1 Acute Effects on Aquatic Plants

The indicator species for aquatic toxicity in aquatic plants and algae are *Lemna gibba* (duckweed, aquatic macrophyte), *Anabaena flos-aquae* (blue-green algae), *Selenastrum capricornutum* (green algae), *Navicula pelliculosa* (fresh water diatom) *and Skeletonema costatum* (marine diatom) (Tables 2 and 13). However, in the case of diquat, a wide variety of surrogate macrophyte species were also tested including *Spirodela polyrhyza* (giant duckweed), *Eichhornia crassipes* (waterhyacinth), *Pistia stratioites* (waterlettuce), *Hydrilla verticillata*, *Potamogeton natans* (floating leaf pondweed), *Elodea canadensis* (American waterweed), Eurasian watermilfoil (*Myriophyllum spicatum*), *Wolfiella floridana*, *Azolla caroliniana* and watermeal (*Wolffia columbiana*). Other species of algae that may be use as surrogate plant species include various blue-green algae (*Microcysitis aeruginosa*, and *Lyngbya wollei*), green algae (*Chlorella vulgaris*, *Chloroccocum* spp., and *Dunaliella tertiolect*a), diatoms (*Cyclotella meneghiana*, *Nitzchia* spp., *Isochrysis galbana*, *Phaeodactylum tricornutum*, *Ochromonas danica*, and *Cryptomonas ozolini*), *Euglena gracilis* and *Peridinium cinctum*. Aquatic algae and plant species are often not tested at concentrations that exceed the expected environmental concentration (EEC). Therefore, the effects of the maximum EEC of 0.75 ppm c.e. according to the Canadian guidelines (Peterson, 1994 and 1997) is often not ascertained. For those species tested with diquat concentrations that equaled the maximum use rate and EEC of (~0.75 ppm), blue-green algae, diatoms, green algae, pyrrophyte euglenophyte and macrophytes had at least short-term effects. However, *Chlorella vulgaris*, *Euglena gracilis, Isochrysis galbana*, *Phaeodactylum tricornutum*, and *Skeletonema costatum* could withstand concentrations of diquat that ranged between 2.95 and 43 ppm c.e. Even more typical application concentrations of 0.224 to 0.37 ppm are high enough to cause acute damage to most species of algae and macrophytes.

Because plants are the intended targets of aquatic herbicides containing diquat, a risk assessment would not usually be conducted to determine the safety of diquat products on plants. A realistic level of concern may be used for aquatic plants since even a reduction in growth of 50 percent will leave a significant amount of forage and habitat (refuge). However, except for the species discussed above, freshwater macrophytes and algae can be adversely affected by diquat (Reward® LA) since the concentrations that they would be exposed to could be expected to reduce growth by more than 50 percent. Since algae species can be an important element of the food chain (Goldman and Horne, 1983), the high risk quotient leads to a level of concern for those species and animals that depend on them for food. Aquatic macrophytes are also very susceptible to the effects of diquat which means that the risk quotient would be higher than unity; and it is (RQ = 1000 = 0.75 ppm c.e./0.00075 ppm c.e. or RQ = 298 = 0.224 ppm c.e./0.00075 ppm c.e.) for the most sensitive species (*Spriodela polyrhyza*) in EPA RED (1995). Aquatic (emergent) macrophytes are of importance in providing both food and habitat to fish, amphibians, aquatic invertebrates, wild birds and mammals (Frank, 1972); therefore, this high RQ exceeds the level of concern for aquatic macrophytes. Peterson et al (1994) expressed concern that the use of a risk quotient of 1.0 as the level of concern does not provide an adequate safety margin for algae and plants. However, since the risk quotient is greater than one for almost all species of plant and algae tested, this seems to be a mute point (Peterson, 1994) (Table 14). The other species with a potentially low to moderate hazard rating (RQ = < 0.1 to 0.26) were *Chlorella vulgaris*, *Euglena gracilis*, *Dunaliella tertiolecta, Isocrysis galbana*, *Phaeodactylum triconutum* and *Skeletonema costatum*.

4.3.1.2 Chronic Effects on Aquatic Plants

Laboratory work to determine the chronic effects of herbicides on algae and aquatic plants is currently not conducted for the purposes of registration.

4.3.1.3 Potential Impacts of Single versus Multiple Applications

Studies performed in both the field and laboratory indicates that algal response to a variety of diquat formulations varies from no effect to stimulation of growth at low concentrations (1-10 ppm c.e.). These concentrations initially cause a decrease in filamentous green algal density but by the 21st day after treatment in a laboratory microcosm, the filamentous green algal density increased to 13- to 19-fold above control levels. Since the unicellular green algae appear to be resistant to diquat at levels up to 10 ppm, it is likely that these species will also exhibit increases in density when exposed to diquat (Cullimore, 1975 in Melendez et al, 1993). Other algal groups including green colonial algae, diatoms, and blue-green algae were reduced in numbers at all time periods tested .

Since nitrogen fixation by blue-green algae grown in anoxic culture is inhibited by diquat at 20 ppm, diquat has the potential to affect the overall nitrogen economy of soils/and sediments (DeSilva et al, 1975). This may be due to the algae killing effects that such a concentration of diquat may have and not directly due to inhibition of nitrogen reductase or other enzymes involved in nitrogen fixation. However, to determine if inhibition of nitrogen fixation by blue-green algae is likely to be a factor influencing the systems nitrogen economy, it would be necessary to conduct similar studies at concentrations of diquat that might be found in the field (e.g., <1.0 ppm c.e.). Other older studies (1969 to 1975) have also shown diquat to stimulate or inhibit growth of algae (Tables 15 and 16).

Laboratory results indicate that green algae survival, growth and productivity are not adversely affected at moderately high concentrations (1 to10 ppm) of diquat. Furthermore, field observations indicate that low concentrations of diquat appear to stimulate algal growth (Cooke, 1977). It is generally agreed that diquat exposure can lead to algal blooms. However, since the concentrations that are directly stimulatory to algal growth are typically higher than the maximum use rate, it is likely that stimulation of algal growth in the field is due to the release of plant nutrients rather than a direct stimulatory effect from the chemical (Melendez et al, 1993; Peverly and Johnson, 1979; Inabinet, 1976; Daniel, 1967). In the field inhibiting effects on phytoplankton growth are probably limited to the first few days after diquat treatment (Inabinet, 1972; Tatum and Blackwell, 1962, and Cooke, 1977). Therefore, it seems unlikely that diquat will have a significant direct impact on phytoplankton growth sufficient to adversely impact the food chain.

Sorption and metabolism of diquat have also been investigated. Diquat appears to be adsorbed extensively by many species of algae and macrophytes. Diquat is adsorbed extensively by alligatorweed, sago pondweed, American pondweed, *Chlorella vulgaris, Navicula pelliculosa*, *Spirogyra* spp., *Cladophora* spp., Eurasian watermilfoil, American waterweed and *Chara vulgaris* (Funderburke and Lawrence, 1963; Simsiman et al, 1976; Hiltibran, 1972; Birmingham and Colman, 1983; Coats et al, 1966; Way et al, 1971; Birmningham and Colman, 1983; and Rogers et al, 1992). Therefore, diquat appears to bioaccumulate in plant tissue. The bioconcentration factor can be as high as ~800 on a dry weight basis and in excess of 250 ppm on a wet weight basis (Birmingham and Colman, 1983 and Rogers et al, 1992). Greater detail on the bioaccumulation of diquat in plants can be found in section 4.2.2.4. Typically, 32% of the applied diquat is degraded on the surface of dead and dying plants (Simsiman and Chesters, 1975). However, little or no diquat is directly metabolized by aquatic plants (Funderburk and Lawrence, 1967 in Simsiman et al, 1976).

Aggressive aquatic herbicide treatment may create more open water for fish habitat. However, aggressive treatment may eliminate areas containing milfoil and other macrophytes that are used by juvenile fish as a refuge from predators and as general habitat (Killgore et al, 1987 in Ecology, 1989). Invertebrates are more abundant on macrophytes other than milfoil, so a community shift to other plant species may result in greater abundance of invertebrates, which would provide more food for the grazing planktovoric fish. Therefore, in most cases where an adverse effect has occurred on fish food organisms, it has been as a result of anaerobiosis rather than loss of habitat (Frank, 1971).

Ecology (1992) suggests retaining 20% to 25% of native vegetation as fish-rearing habitat in treated areas. This also creates more open water with fewer macrophytes and increases habitat for post- and non-breeding adult fish, while at the same time allowing for increased invertebrate habitat, which increases the food source for fish. This intermediate approach provides improved habitat and food source for juvenile, sub-adult and adult fish.

Although these approaches have a large element of common sense behind them, a decrease in fish populations due to lack of sufficient macrophyte habitat is still primarily a hypothesis (Bain and Boltz, 1992, and Swingle, 1956 and Swingle, 1950). Swingle indicated that for the best forage fish to game fish ratio (3 to 6), there is an ideal areal plant cover due to macrophytes and an ideal level of foodstuff filamentous algae. However, it was not clear what these ideal levels are and they probably vary considerably from pond to pond. However, Ecology's (1992) suggested value of 20 % - 25% seems to be a reasonable compromise when good experimental data is not available to support such a position. There does seem to be good evidence for the need of adequate levels of filamentous algae to support vertebrate and invertebrate populations in the field. For example, Cooke (1977) found that treatment of a pond in Woodwalton Fen National Nature Reserve with 1.0 ppm diquat initially resulted in a slight decrease in the weight of Rana *temporia* and *Bufo bufo* tadpoles. This seemed to be due to the removal of algae from the treated ponds by the action of the herbicide. By 18 to 32 days the weight of these tadpoles had doubled relative to the control which was considered to be a significant increase in weight. This increase was due primarily to a bloom in filamentous green algae (particularly *Mougeotia* spp.) The report also showed that there were more (7- to 26-fold more) unicellular green algae eaten by these tadpoles than by the controls. Although the number of diatoms eaten at 18 days was greater in the diquat-treated ponds than in the control ponds, fewer diatoms were eaten 32 days after exposure to diquat than in the untreated ponds. Invertebrate populations also increased in number after treatment with diquat. There was a massive increase in claDOeran (*Simocephalus vetulus*) populations due to subsequent algal blooms of *Mougeotia* spp. at this site. Tatum and Blackburn (1965) showed massive increases in glassworm (*Chaoboridae*) shortly after treatment with diquat at 0.5 ppm c.e., which could be due to increased foodstuffs (decaying southern naiad) or possibly the improvement of benthic habitat by eliminating excessive course weeds from the bottom of the treated pond (Gilderhaus, 1967). With the increase of fish food organisms like rotifers, glassworms and claDOerans, the number of fish which use them as food may also increase. The effects of plant removal and its impacts on habitat for fish and invertebrate animals will be discussed in subsequent chapters.

- **Potential impact on plant and algae densities**

  Field studies with both algae and macrophytes indicate that the numbers of these plants can be strongly affected by the use of diquat at concentrations that are typically used in the field. Johnson (1962), Hulbert (1987), Tatum and Blackburn (1962), Cooke (1977), Melendez et al (1993), Olaleye et al (1993), Berry et al (1975), and Hilsenhoff (1966) cite a number cases, which indicate that use of diquat at approximately normal field rates can eliminate macrophytes and filamentous algae from a treated water body and cause planktonic blooms. The elimination of macrophytes and filamentous algae is due to the direct toxic action of diquat. Algal blooms can be due to a stimulatory affect in the case of filamentous green algae, removal of competition in the case of single cell green algae or the release of nutrient nitrogen or phosphorous in the case of filamentous green algae or unicellular green algae.

Effects of diquat on increases in phytoplankton can begin as quickly as 3 days after treatment and may persist for a short period of time or until the water body has been entirely cleared of the decaying weed mass.

Treatment of a laboratory microcosm at concentrations of 10 to 30 ppm diquat can cause blooms of unicellular green algae with an increase in numbers from 400,000 cells/mL to up to 2,000,000 cells per/mL within 21 days. The increase in unicellular green algae numbers is dosage related. However, concentrations of diquat below 3.0 ppm seem to have no impact on the growth rate of unicellular green algae. Within the microcosm, unicellular green algae appear to be relatively resistant to the effects of diquat with *Oocystis* spp. being more resistant than the other species (Melendez et al, 1993). Unicellular green algae typically appear to be more tolerant of diquat (EC50 = 0.65 to 43 ppm c.e.) for most species (Table 13). However, most species of blue-green algae and diatoms are more susceptible to the effects of diquat (EC50 = 0.02 to 0.175 ppm c.e.) which was verified by the field work of Shealy (1971 in Shearer and Halter, 1980) where 0.5 ppm diquat killed diatoms, blue-green algae and other filamentous forms. In field studies, similar observations were noted. For example, the phytoplankton density increased form 200 to 300 organisms /ml to 1,200 to >2,000 organisms seven days after treatment of Lake Marion in North Carolina with 3 gallons formulation/ha (Inabinet, 1976). Cooke (1977) found that both filamentous green algae and unicellular green algae bloomed within 32 days of treatment with 1.0 ppm diquat, which provided additional foodstuffs for frogs (*Rana temporia*) toads (*Bufo bufo*) and the claDOeran (*Simocehalus vetulus*). While exposure to diquat in laboratory microcosms appears to have a direct stimulatory effect on the growth of unicellular green algae, the increases in the number of phytoplankton seen in the field appear to be due to releases of nutrients from dead and dying macrophytes. Peverly and Johnson (1979) were able to correlate the release of phosphate from *Elodea canadensis* after treatment with 1 ppm diquat with a 0.200 ppm increase in chlorophyll. Although nitrogen nutrients also increased in this time frame, it could not be attributed to nutrient release from diquat-treated macrophytes. Although decreases in phytoplankton (particularly unicellular greens and filamentous green) were seen for a few days after application, the algal densities appeared to rebound within 18 to 32 days (Cooke, 1977 and Tatum and Blackwell, 1962). Similarly, 0.5 ppm diquat plus 0.5 ppm paraquat was not effective in reducing the standing crop of phytoplankton in a British Columbia, BC boat marina, but population growth was stimulated within 3 weeks post-treatment (Water Investigation Branch, 1977 in Shearer and Halter, 1980). Both laboratory and field effects of diquat on phytoplankton are summarized in Tables 15 and 16, respectively.

The numbers of macrophytes in treated ponds decreased dramatically in those treated for aquatic weed control. Treatment with diquat at 2.0 Kg a.i./ha killed weed mats of waterhyacinth at Kofawei Creek in Nigeria within two weeks of application (Olaley et al, 1993). Treatment with 0.5 ppm diquat at a pond in Broward County in Florida, controlled southern naiad by the seventh day after treatment and there was no re-growth by the end of the summer season (Tatum and Blackburn, 1962). Treatment of a pond in Price County in Wisconsin with 1.0 ppm destroyed most of a benthic habitat that consisted of American waterweed. Treatment with 0.5 ppm c.e. diquat completely eliminated susceptible vegetation from Spooner Pond in Wisconsin. Susceptible species included *Potamogeton richardsonii*, *P. pectinatus*, *Eleacharis acicularis*, *Lemna minor*, *Callitriche* spp. and the filamentous algae (*Mougeotia* an *Zygnema*) (Johnson, 1962) Other species controlled in the field include *Elodea canadensis*, *Ceratophyllum demersum*, *Potamogeton crispus* and *Lemna* spp., watermilfoil species (*Myriophyllum* spp.), various pondweed species, water stargrass

(*Heteranthera* spp.) and *Azolla* spp. which are typically controlled at the 90% to 100% level within 1 month of application of diquat at 0.5 ppm c.e. (Johnson, 1962; Hulbert, 1987 and Lawrence et al, 1962). Unfortunately, many of these other species are not effectively controlled for a full two months. For example: duckweed and curlyleaf pondweed are only controlled for 4 to 6 weeks after application of diquat at 0.5 ppm. Watermilfoil is also difficult to control and applications at typical use rates fail to control these species for a full two months (Shearer and Halter, 1980).

- **Potential impacts on diversity**

  The dominant species of phytoplankton can change after treatment with diquat. Melendez et al (1993) and Pratt et al (1990) found that the species diversity in laboratory microcosms appeared to shift. For example, treatment with concentrations as low as 0.3 ppm cause a 20% decrease in the number of taxa protista. The decrease in diversity for taxa protista was related to dosage. However, it is not clear that concentrations higher than 1.0 ppm, which decreased protist diversity by 50%, had any environmental relevance since such concentrations are higher than the maximum use rate. The green unicellular algae came to dominate the microcosm after treatment with diquat at 10 to 30 ppm. It is not clear that such high concentrations have environmental relevance. Nevertheless, since 0.3 ppm diquat can inhibit the growth of blue-green algae and diatoms, it is likely that these species will become fewer while species of green algae will become greater in number which may both decrease the algal diversity of an ecosystem exposed to diquat and allow for a shift in the dominant organisms from general unicellular green algae to *Oocysis* spp.

  The dominant macrophytes in a pond can vary depending on the history of the pond. However, in a pond in Broward County, Florida, the dominant macrophyte prior to treatment with 0.5 ppm diquat was southern naiad (*Najas guadalupensis*). Similarly, in Spooner Pond, Wisconsin, pondweeds, naiads and *Lemna minor* appeared to dominate the water body prior to the application of 0.25 to 0.5 ppm diquat. In another pond in Wisconsin, the water body was dominated by Eurasian watermilfoil prior to the application of 2.2 ppm Cutrine® plus 3.0 ppm diquat. However, after treatment of these ponds with diquat the dominant pest species was rapidly eliminated from the water body. The dominant species were typically eliminated within one to two weeks. Without competition from the former dominant species, *Chara* spp. often became the dominant species within one or two months since it is resistant to the effects of diquat (Tatum and Blackburn, 1965; Daniel, 1972, and Johnson, 1962). In the case described by Daniel (1972), watermilfoil did not re-grow on its own by 240 to 320 days, and various species of pondweed had become abundant. Duckweed and curlyleaf pondweed rapidly re-infest reservoirs when more competitive species like *Elodea canadensis*, *Potamogeton pectinatus* and *Ceratophyllum demersum* have been eliminated (Hulbert, 1987). Other species that could become dominant due to their tolerance to diquat include water lilies (*Nuphar* spp. and *Nymphaea* spp.) (Shearer and Halter, 1980). Production of dense monospecific stands will also decrease the diversity of animals, which require other species of plants for adequate foodstuff and habitat. It may, therefore, be necessary to remove these new undesirable dominant plants by using another method of control.

- **Naturally occurring re-growth and reproduction of non-noxious or non-invasive plants**

  Most noxious plants are substantially reduced upon treatment with diquat. Nevertheless, it is clear that while native and desirable pondweed species do recover, some of the more difficult to control species like *Chara* spp., Duckweed (*Lemna* spp.), curlyleaf pondweed, *Nymphaea* spp., *Nuphar* spp., and even watermilfoil (*Myriophyllum* spp.) in the Northwest may come to dominate a treated water body (Johnson, 1962; Hulbert, 1987; Tatum and Blackburn, 1965; Daniel, 1972, and Shearer and Halter, 1980).

Diquat is not very selective when used at concentrations that will control water milfoil in the Northwest. However, diquat appears to be extremely effective in controlling *Elodea canadensis*, and many species of pondweed (*Potamogeton*), although some species like *P. richardsonii*, *P. robbinsii* and *P. nodosus* are difficult to control. Although Reward® LA is effective at controlling some species of algae; it appears to have a stimulatory affect on some species of green unicellular and green filamentous algae which can potentially lead to an algal bloom (Cooke, 1977). Such stimulatory effects on algal growth may be advantageous in developing increased foodstuff and habitat for certain amphibian species and claDOerans.

- **Post treatment plantings of non-noxious or non-invasive species**

  In a general review article, Frank (1972) recommended the planting of non-noxious, and non-invasive native plants after the elimination of exotic noxious and invasive plants. However, some scientists have found efforts to reestablish native plant species are often unsuccessful. He indicated that such plantings would be competitive with the faster growing exotics once they have been eliminated. These native species can serve as both food and habitat for waterfowl, fish food organisms and fish. For a further discussion of diquat on numbers and diversity of aquatic animals (please see Section 4.3.2.3).

  Although diquat has usually been inactivated by adsorption to sediment (Birmingham and Colman, 1983, Daniel, 1972), only limited success has been noted with post treatment plantings. Although you would not plant this species to re-vegetate a water body, Daniel (1972) found that some success with transplants could be obtained by transplanting Eurasian watermilfoil 268 days after treatment had elapsed. Birmingham and Colman (1983) found that Duckweed could be planted in sediment containing up to 170 ppm diquat without completely eliminating growth. However, at those concentrations residual phytotoxicity becomes apparent in this closed system.

- **Effects on aquatic plants: potential impacts of single versus multiple applications**

  Initial elimination of exotic plants should increase habitat for fish (Bain and Boltz, 1992, Olaley et al, 1993). Growth and reproduction of bluegill sunfish does not appear to be impacted by treatment with diquat at concentrations that eliminated American waterweed and inhibited the growth of filamentous algae. However, multiple treatments (3-times) at concentrations of 1.0 ppm appeared to decrease the growth of individual adult fish without effecting the overall standing crop weight of the treated ponds (Gilderhaus, 1967). The treatment of Guntersville Reservoir with Komeen®, plus diquat at 0.3 ppm copper and 0.3 ppm c.e., though probably not necessary to control *Hydrilla verticillata* in the year of application, had no effect of the abundance, size structure, condition or movement of largemouth bass in the treatment area (Bain and Boltz, 1992). Killing mats of waterhyacinth with 2.0 Kg a.e./ha allowed for an increase in both fish numbers (from 8 to 67) and diversity (from 1 to 6 species) and an increase in maximum weight in *Stenopoma kingslayae* (from 35.0 to 95.0 g). The elimination of American waterweed by treatment with 1.0 ppm diquat resulted in a decline in numbers for four species of mollusks and an amphipod (*Hyallela azteca*) (Hilsenhoff, 1966). The effects of the elimination of American waterweed may be mitigated by the use of an artificial substrate like conservation webbing (Berry et al, 1975).

4.3.1.4 Effects on Endangered Plant Species

The current literature does not discuss the effects of diquat on endangered plant species. However, a few general comments can be made. Reward® LA (liquid diquat) is normally applied at or below the water surface; thus accidental "drift" exposure to upland vegetation during application would be minimal with the exception of emergent aquatic plant communities bordering the treated area. If any proposed "sensitive" plants or candidate species under review for possible inclusion in the state list of endangered or threatened species occurs along the banks of waterways to be treated with diquat products, the applicator should leave a protective buffer zone between the treated area and the species of concern (Ecology, 1989). Sensitive upland plant species could potentially be damaged if treated water was improperly used for irrigation or extensive flooding from irrigation canals treated with diquat for the control of terrestrial or emergent weeds growing on the ditchbanks before significant herbicide dissipation had occurred. Use of treated water for irrigation is normally prohibited for three to five days after treatment with Reward® LA or until an acceptable analytical method shows the concentration of diquat has decreased below the MCLG (0.02 ppm). To protect endangered aquatic plants, some knowledge must be gained on the toxicity of diquat to these plants, or diquat must not be applied in areas that will adversely impact the habitat or population of these plants. In the case of threatened aquatic plants, the Endangered Species Act does not allow for the control of noxious weeds to take precedence over the protection of endangered species. However, if conditions indicate that removal of noxious weeds will improve habitat for threatened/endangered plant species, removal of the noxious species by chemical or other means should be considered. The permit for treatment of water bodies to control noxious or invasive plants may be denied or amended if Ecology believes that populations of plants may be adversely impacted by treatments to control these weeds (McNabb, 1999 and Dorling, 1999 personal communications).

Endangered plant species in the State of Washington that are either fully aquatic, palustrine or riparian are as follows: Ute Ladies' Tresses, Golden paintbrush, and Nelson checker mallow (terrestrial species); water howellia and marsh sandwort (aquatic species). For current listings see: http://www.wa.gov/dnr/ocn/pg62.html.

4.3.1.5 Risk Analysis for Aquatic Species of Plants

It is not standard procedure to conduct a risk assessment with a herbicide for aquatic plants and algae. Although DaSilva et al (1975) has shown that diquat at concentrations of 20 ppm a.i. (10.8 ppm c.e) completely inhibits nitrogen fixation in various species of blue-green algae; it is unlikely that concentrations of this magnitude will be encountered in the field. *Anabaena flos-aquae* is at moderate to high risk (RQ =0.42 to 1.18) when exposed to diquat based on laboratory experiments. However, treatment of water bodies with 0.5 to 5.0 ppm diquat in the field kills filamentous blue-green algae and in some cases induces algal blooms of *Anabaena* spp. and *Staurastrum* spp. (Table 16) (Shearer and Halter, 1980 cite Shealy, 1971 and the Water Investigation Branch, 1977). Rapid recovery in the field and lack of testing at environmentally relevant concentrations indicates that destruction of the nitrogen fixation capacity is not likely to be a problem when diquat is used according to the Reward® LA Label (2000).

The most sensitive species of algae are the diatom *Ochramonas danica* and the pyrrophyte *Peridinium cinctum,* which appear to be at least moderate risk when the population of concern is adjacent to the treatment area or up to 375 meters outside the treatment area. (Table 17) (Ritter et al, 2000). This data is confirmed by previous work done by Ritter et al (1995) that indicates a set back distance of at least 400 meters should be use when spot treating weed infested areas within a reservoir. Although there is evidence that treatment with diquat plus paraquat at 0.5 plus 0.5 ppm did not reduce the standing crop of phytoplankton in a British Columbia boat marina, it is not clear which species were monitored. Therefore, when pyrrophytes or diatoms are major components of an algal population, cautious judgment should be used to protect these algal forms from being damaged. Other species of blue-green algae and freshwater diatoms appear to be at similar risk after diquat treatment and caution should be used to protect these algal forms if they are significant as foodstuffs in areas adjacent to the treatment area. In most cases, green algae appear to be at only moderate to low risk when they are in a spot treatment zone or outside the treatment zone (RQ = <0.5) (Table 17). However, Cooke (1977) found that both unicellular and filamentous green algae were reduced in number for only a few days after treatment of a pond with diquat. These experiments were particularly interesting since the method of monitoring algal levels was determining the numbers of various algal forms that were eaten by algaephytic frogs and toad tadpoles. Such utilization of a food source would seem to be key to the ecological health of an ecosystem.

Very little work has been done with aquatic macrophytes, but diquat would be expected to cause high risk to most species when they are located within the treatment area. All species of plants tested were at high to very high risk (RQ = 0.53 to >1.0) when a pond was treated according to EPA's worst case scenario even if a 4-day geometric mean on the EEC was factored into the risk evaluation. Common duckweed, giant duckweed, American waterweed and floating leaf pondweed (*Potamogeton natans*) appeared to be at high to very high risk (RQ = 0.91 to 15) even when diquat was used as a spot treatment at 0.30 ppm c.e. Other tested species were only at low to moderate risk (RQ = <0.1 to 0.46) when diquat was applied as a spot treatment to control aquatic weeds. Most macrophytes located outside the treatment area appeared to be at low to moderate risk from the application of spot treatments of diquat for the control of aquatic weeds. However, common duckweed, giant duckweed and floating leaf pondweed appear to be at risk even when they are located outside the treatment area. Although moderate to very high risk for Eurasian watermilfoil and duckweed is indicated by these laboratory studies, Hulbert (1987) and Shearer and Halter (1980) indicate that these species are effectively controlled for less than 2 months in the field at treatment rates of 0.25 to 0.5 ppm c.e. Other species including sago pondweed (*Potamogeton pectinatus*) coontail (*Ceratophyllum demersum*), American waterweed, *Elodea canidensis*, naiad (*Najas flexilis*), and *Eleacharis acicularis* were controlled in the field at 0.25 to 0.5 ppm c.e. for over two months in experimental ponds managed by Chevron Chemical Company, Wisconsin.

It is unfortunate that concentrations that will control Eurasian watermilfoil in Washington State waters have been shown to kill almost all of the native aquatic species (Shearer and Halter, 1980). Therefore, diquat is very effective in controlling most pondweed species, American waterweed, Hydrilla and watermeal amongst other species, but it may not be the best material for controlling watermilfoil in light of cost and efficacy.

Certain species appear to be tolerant of diquat. For example, duckweed, *Chara* spp., curlyleaf pondweed, Richardson's pondweed, Robbin's pondweed, *Nuphar* spp. and *Nymphaea* spp. are tolerant or controlled for only short periods of time after application. These species may dominate a water body after more competitive species are removed by the use of diquat (Shearer and Halter, 1980; Hulbert, 1987 and Reward® LA label).

The conclusion that can be drawn from risk assessments on algae and macrophytes based on Peterson et al's (1994) conservative approach is that this segment of the biota is at high risk for ecological damage from diquat. However, this conclusion must be tempered by considering various use rates and how far the at risk population is from the treatment area (Ritter et al, 2000 and Shaw et al, 1995). The conclusions based on these understandings are as follows: 1) Treatment at the maximum use rate (2 gallons of Reward® formulation in 2 feet of water = 0.75 ppm c.e.) places the algal and aquatic macrophyte biota at very high risk since the risk quotient is greater than EPA's level of concern for high risk (1.0) for the most sensitive species; RQ = 750= 0.75 ppm.c.e./0.001 ppm c.e. for the pyrrophyte algae *Peridinium cinctum* or RQ = >38 = 0.75 ppm c.e./<0.02 ppm c.e. for the macrophyte, *Lemna minor*; 2) A similar conclusion of high risk is drawn when diquat is applied under EPA's worst case scenario (2 gallons of Reward® LA in 200 cm of water = 0.224 ppm); RQ = 224 = 0.224 ppm c.e./0.001 ppm c.e. or RQ = >11 = 0.224 ppm/<0.020 ppm c.e; 3) If the 4-day geometric mean of the EEC (0.059 ppm c.e.) after a worst case scenario application is used, this segment of the biota is still at very high risk; RQ = 59 = 0.059 ppm c.e./0.001 ppm c.e. or RQ = >3.0 = 0.059 ppm c.e./<0.020 ppm c.e.; 4) Under the scenario of a worst case spot treatment in a Northwest reservoir (4-day geometric mean of a 0.30 ppm treatment = 0.021 ppm), this segment may still be judged to be at very high risk; RQ = 21 = 0.021 ppm c.e./0.001 ppm c.e. or RQ = >1.1 = 0.021 ppm c.e./<0.02 ppm c.e.; 5) When the area of concern is adjacent to a reservoir section that has been spot treated with 0.30 ppm c.e. (4-day EEC = 0.008 ppm c.e.), very high risk to algae is likely but moderate to very high risk is likely to macrophytes; RQ = 8.0 = 0.008 ppm c.e./0.001 ppm, or RQ = 0.4 to 5.7 = 0.008 ppm c.e./0.02 ppm c.e. or 0.008 ppm c.e./0.0014 ppm c.e.; 6) When the area of concern is 375 meters from the reservoir section that has been spot treated as described above (4-day EEC = 0.00028 ppm), moderate risk to algae is likely and moderate to low risk is likely for macrophytes; RQ = 0.28 = 0.00028 ppm c.e./ 0.001 ppm c.e., or RQ = 0.014 to 0.2 = 0.00028 ppm c.e./0.02 ppm c.e. or 0.00028 ppm/ to a c.e./0.0014 ppm c.e.

The only situation where algae and plants will not be at significant risk from diquat application is when the treatment area is sufficiently removed from the ecologically sensitive area. In this case, algae and plants can be judged to be acceptable risk from the effects of diquat. Under all other treatment scenarios, algae and plants should be judged to be under high risk from treatment with diquat. However, this does not imply that all species of algae or aquatic macrophytes will be at risk within the treatment area. For example, 80% of the algal species would not be at very high risk (RQ = >1.0) when they are located within the treatment zone; the 4-day EECs in these cases are less than the EC50s for these algal species.

4.3.2 Effects of Diquat on Aquatic Animals

*Summary: Diquat is generally safe to fish, free-swimming aquatic invertebrates, and benthic invertebrates when the EC50/LC50 is compared to typical 4-day geometric means of the expected environmental concentration (EEC). When the toxicity of diquat is compared to other pesticides, it is classified according to the U.S. EPA Ecotoxicological Categories highly toxic (LC50 = 0.1 to 1.0 ppm) for striped bass (*Morone saxatilis*) sac-fry in soft water (LC50 = 0.54 ppm c.e.) to practically nontoxic (LC50 = >100 ppm) to grass carp (*Ctenophryngodon idella*) and yearling common carp (*Cyprinus carpio*) in hard water (LC50 = 125 to 141 ppm c.e.). Striped bass, smallmouth bass (*Micropterus dolomieu*) and largemouth bass (*Micropterus salmoides*) are known for being particularly sensitive to the laboratory effects of diquat (LC50 = ~5.0 for fingerlings of both species). More typical toxicities (LC50s) for fish native to Washington range from 10 to 125 ppm for bluegill sunfish (*Lepomis macrochirus*), 20 to 30 for salmon*

*(*Oncorhynchus *spp.), 10 to 15 ppm c.e. for rainbow trout (*Oncorhynchus mykiss*) and 19 ppm c.e. for brown trout (*Salmo truta*).*

*Several species of invertebrates are extremely susceptible to the laboratory effects of diquat. For example: diquat would be classified as very highly toxic (LC50 = <0.1 ppm) to the amphipod (*Hyalella azteca*) (LC50 = 0.048 ppm c.e.), highly toxic to the pocket shrimp (*Mysidopsis bahia*),* Daphnia pulex *and apple snail (*Pomacea pludosa*) (LC50 = 0.42, 0.16 and 0.34 ppm, respectively). However, for most species of invertebrates diquat is classified as much less toxic; e.g. diquat is classified as moderately toxic (LC50 = >1.0 to 10 ppm) to practically nontoxic (LC50 = 100 ppm) for mayflies (*Callibaetis *spp.), oysters (*Crassostrea virginica*),* Daphnia magna, Diapotomus *spp*., Eucyclops *spp., various odonates, caddisfly (*Limnephilus *spp.), various species of marine shrimp and bloodworms (*chironomidae*) (LC50 = 16, 55 to 141, ~1.0, ~5, 25. >100, 33, >1.0 to 8.5 and >100 ppm c.e.). Most other species of free swimming and benthic invertebrates show similar affects in laboratory testing.*

*Diquat appears to be safe for use in aquatic ecosystems. When comparing typical expected environmental concentrations (EEC) of diquat with laboratory LC50s, the highest concentration that may be encountered immediately after application (0.75 ppm c.e.) may affect more sensitive fish species. For example, bass (sac-fry and free swimming) (LC50 = 0.54 to 5.4 ppm c.e.) and walleye (*Stizostedion vitreu*) fry (LC50 ppm c.e. = 0.75 to 4.9 ppm) would be adversely impacted. However, if EPA's worst case scenario is used (0.224 ppm c.e. at application), diquat can be used as a restricted use compound without exceeding the level of concern risk quotient (0.5) for restricted use compounds (RQ = 0.41 = 0.224 ppm c.e. / 0.54 ppm c.e. for the most sensitive fish species). Since diquat is classified as a restricted use compound, it can be used for control of the labeled aquatic weeds without great potential for adverse impact to the fish. When factoring in an appropriate dissipation half-life (0.75 days) or considering a typical use to be a spot treatment of 0.30 ppm c.e., the 4-day geometric means become 0.059 ppm c.e. and 0.021 ppm c.e., respectively. With these refined EECs, the risk quotient does not significantly exceed the level of concern (0.1) for a general use aquatic herbicide; (RQ = 0.11 = 0.059 ppm c.e./0.54 ppm c.e. or RQ = 0.079 = 0.059 ppm c.e./ 0.75 ppm c.e).*

Invertebrates will not be entirely safe from the effects of diquat. Even if the refined EEC values of 0.059 and 0.021 ppm c.e. are used, the more sensitive benthic species may be affected. The LC50 value for Hyalella azteca *is less than or approximately equal to the EEC under the worst case and spot treatment scenarios. Therefore, the level of concern risk quotient (0.1) is exceeded for this species (RQ = 1.2 = 0.059 ppm /0.048 ppm c.e. or RQ = 0.44 = 0.021 ppm c.e. / 0.048 ppm c.e.). Other species that may be at risk include Daphnia pulex (LC50 = ~0.16 ppm c.e.), apple snail (LC50 = 0.34 ppm c.e.) and pocket shrimp (LC50 = 0.42 ppm c.e.). The level of concern risk quotient is not exceeded if diquat is treated as a restricted use compound when these species are present. Therefore, although the invertebrate biota appears to be at risk for one species (*Hyalella azteca*) out of 26, diquat can probably be used according to the Reward® LA restricted use requirements without significant risk to most of the invertebrate biota.*

The chronic toxicity (MATC) for diquat is also low with predicted or empirical long-term MATCs ranging from 0.045 ppm c.e. for Daphnia magna *to 0.2 ppm c.e. for fathead minnow (*Pimephales promelas*). Although chronic toxicity was evaluated for* Fundulus heteroclitis, Xenopis laevis *and* Rana pipiens *to be 0.017 ppm, 0.64 ppm c.e. and 1.7 ppm c.e., respectively, the studies were conducted for less than 28 days, the species were not environmentally relevant and/or the lowest treatment concentrations resulted in mortality that exceeded that of the controls making the studies difficult or inappropriate for interpretation. Although the long-term (28-day)* *geometric EEC is estimated to be 0.0087 ppm, it is unlikely that aquatic organisms will encounter such concentrations in the field and therefore, the chronic toxicity of diquat should be a mute point. However, if one assumes that the acute/chronic toxicity ratio should be the same for all species of fish (38X) and invertebrate (19X), the chronic effect of diquat on bass will be 0.014 ppm c.e. (0.54ppm c.e./38) and the chronic effects on* Hyalella azteca *will be 0.0025 ppm c.e. (0.048 ppm/19) and on* Daphnia pulex *will be 0.0084 ppm c.e. (0.16 ppm c.e./19).*

For these most sensitive species, the fish biota will be protected from the chronic effects of diquat since the risk quotient is less than the level of chronic concern for 28 days (1.0) (RQ = 0.6 = 0.0084 ppm c.e./0.014 ppm c.e.). However, since the two most sensitive species of invertebrates yield 28-day risk quotients equal to or higher than the level of concern (RQ= 3.48 = 0.0087 ppm c.e./0.0025 ppm c.e. or RQ= 1.0 = 0.0087/0.0084 ppm c.e.), sediment and free-swimming invertebrates may not be protected from the effects of diquat treatment. However, in the presence of sediment, the sediment species may be protected since the LC50 for Hyalella azteca *is significantly higher under these more natural scenarios (LC50 = 3.4 to 6.8 ppm c.e.). Since the risk quotient would be less than the chronic level of concern (RQ= 0.048 = 0.0087 ppm c.e./0.17 ppm c.e.) there is a strong possibility the* Hyalella *and other similarly sensitive species will be protected from the effects of diquat under field conditions.*

Laboratory exposure of Coho salmon may adversely impact the parr to smolt metamorphosis at concentrations as low as 5.0 to 20 ppm diquat. Smolting salmon exposed to this concentration of diquat experienced 15% to 87.5% mortality when subsequently exposed to seawater. Additional experiments with concentrations of up to 3.0 ppm diquat did not produce mortality when Coho salmon were exposed to diquat. However, exposure to concentrations as low as 0.5 ppm interfered with the ability of Coho salmon to migrate downstream. Concentrations of 3.0 ppm diquat produced histopathological effects on eyes, kidney, gills, and liver (Lorz et al, 1979). However, similar experiments with Chinook salmon at concentrations ranging from 0.125 to 0.5 ppm c.e. produced no mortality or osmoregulatory effects in a subsequent seawater challenge (Merill, 1997). Other anadromous fish species like steelhead, sea-run cutthroat trout or American eel have not been tested for their ability to osmoregulate or migrate appropriately. Since there is some evidence that diquat may affect seawater survivability and migration at concentrations that may be encountered in the environment, this is worth further investigation. It also indicates the permits should be written so that breeding and smolting andromonous species are not exposed to diquat.

Behavioral effects have been observed on exposure to diquat. Rainbow trout do not exhibit active avoidance behavior at concentrations of diquat up to 10 ppm c.e. (Folmar, 1977). However, passive avoidance behavior, a tendency to drift with the current, has been observed in rainbow trout exposed to concentrations of diquat as low as 0.5 ppm c.e.(Dodson and Mayfield, 1979). Similar effects were observed in fathead minnow at concentration of diquat as low as 9.2 ppm c.e. (de Peyster and Long, 1993). An active avoidance response to diquat was seen in goldfish at concentrations as low as 1.1 ppm diquat (Berry, 1984). In the field, carp and suckers would avoid diquat at very high field use rates (26.7 ppm). However, rainbow trout did not display any reaction to the diquat during field exposure (Hesser et al, 1972). Passive avoidance may have been the result of respiratory distress (coughing). Respiratory distress has been displayed in yellow perch exposed to concentrations of 1 to 5 ppm diquat (Bimber et al, 1976). This may be environmentally relevant since even at concentrations of 1.0 ppm, respiratory stress is demonstrated within 6 hours of exposure.

Field studies indicate that treatment with diquat appears to have an affect on some species of free-swimming or benthic invertebrates in ponds. Diquat applied to the field at use rates of 2.5 ppm caused a reduction in the numbers of Hyalella azteca *associated with acute toxicity. However, other factors could have contributed to this population reduction including a decrease in dissolved oxygen concentration and from the destruction of submerged plants (Wilson, 1967). Similar work by Hilsenhoff (1966) at concentrations of 1.0 ppm caused a decrease in the numbers of* Hyallela azteca *and 4 genera of snails. However, these decreases were due primarily to the decrease in habitat from the destruction of* Elodea canadensis*. Berry et al (1975) found that shallow water treated with diquat (0.11 ppm) plus endothall (0.17 ppm) often had lower numbers and diversity of invertebrates than deep water. However, this effect was also seen in the controls, and therefore could not be attributed to treatment with herbicides. The addition of conservation webbing may have a mitigating effect on the elimination of* Egeria densa *or* Elodea canadensis *from treated water. Various species of cladoerans were also affected by application of 1.0 ppm diquat. Exposure to these concentrations of diquat in the field caused* Simocephalus vetulus *to disappear from the water column for up to 64 days (Newbold, 1975 in Shearer and Halter, 1980). Similar work by Gilderhaus (1967) caused unspecified claodoceran species to shift their time of peak abundance in a Wisconsin pond by several months. Multiple treatments with diquat at 1 ppm caused the average and peak number of cladoerans to decrease by two- or three-fold in addition to a shift of peak population times. Treatment with diquat at 0.5 ppm to control southern naiad was accompanied by a large increase in* Chaoborus *spp. numbers (2-fold) due to an increase in decaying vegetation, which served as a food source. Field data supports the laboratory data that indicates* Hyallela azteca*, various cladoerans and possibly more than one species of pond snail would be adversely impacted by treatment with diquat to control aquatic weeds.*

Fish species like largemouth bass, sunfish and others are not adversely affected by typical field concentrations of diquat. There was no adverse effect on numbers (experimental largemouth bass catch) and no adverse effect on mean total length, condition, and movement within the treatment area. Furthermore, fingerling bluegill sunfish grew at the same rate in control and treatment ponds while the average increase in the size (length and weight) of adults was much reduced in the treated ponds. However, the total harvest weight did not differ between treated and untreated ponds. Fall spawning of bluegills from treated ponds was successful with 100 adults/acre producing approximately 100,000 fry per acre. In studies conduced in Nigeria, control of waterhyacinth with 2 Kg a.e./acre diquat caused a marked increase in number, maximum size, and diversity of native species. For example, there was an increase in numbers from 8 to 67 fish/unit area; increase in maximum size for Ctenopoma kingslayae *from 65 grams to 95 grams; and an increase in diversity from 1 species to 6 with the dominant species changing from* Ctenopoma kingslayae *to* Channa obscura.. *Field studies appear to support the results of the laboratory studies, which indicated that no adverse impact would occur from treatment with diquat to control aquatic weeds.*

Sensitive, endangered and threatened species of aquatic animals that may need protection through mediation include Coho salmon, chum salmon (Hood Canal summer chum), Chinook salmon, sockeye salmon, bull trout, steelhead trout, cutthroat trout, Coastal cutthroat trout, Olympic mudminnow, mountain sucker, lake chub, leopard dace, Umatilla dace, and river lamprey. Other species which may need protection within Puget Sound, the San Juan Islands, and the Strait of Juan de Fuca east of the Sekiu River are Cherry Point Herring, Discovery Bay Herring, and South Pacific cod.

Diquat applications to fully aquatic (lentic and lotic) systems may be toxic to some aquatic animals (Table 2, 18 and 19). Diquat will generally be safe to the most ecologically relevant species. However, diquat is acutely toxic to a few species including bass (*Morone saxatilis*) sac-fry (LC50 = 0.54 ppm), smallmouth bass (*Micropterus dolmieu)* sac-fry (LC50 = 1.3 to 3.9 ppm c.e.), largemouth bass (Micropterus salmoides) fingerlings (LC50 = 4.2 to >10 ppm) and the walleye (*Stizotedion vitreu*) fry (LC50 = 0.75 to 4.9 ppm c.e.). Although diquat is less toxic to bluegills, fathead minnows, and grass carp in hard /basic water (LC50 = 70-125 ppm c.e.) than in soft/neutral water (LC50 = 7.6-39 ppm c.e.), the difference is not great enough to afford significant protection to these species due to pH and hardness alone (Table 8). However, for walleye and those species of bass that are fairly susceptible to diquat, water bodies with a high degree of hardness may mitigate the effects of diquat particularly right after treatment at rates approaching the maximum use rate (0.75 ppm c.e. according to the label or 0.224 ppm in EPA's worst case scenario).

However, diquat is acutely toxic to several species of invertebrates including the amphipod (*Hyallela azteca*), cladoeran – waterfleas (*Daphnia pulex*), the pocket shrimp (*Mysidopsis bahia*) and the apple snail (*Pomacea pludosa*) with LC50s = to 0.048, 0.16, 0.42 and 0.34 ppm c.e., respectively). The presence of sediment in the system lessens the toxicity of diquat to sediment-dwelling invertebrates. For example, the toxicity of diquat in the presence of sediment is 3.4 to 6.8 ppm to *Hyalella azteca*, which provides up to a ~140 additional safety factor over diquat in the presence of water alone. Nevertheless, for most species of invertebrates, diquat has a low toxicity; e.g., mayflies (LC50 = 16.4 ppm c.e.), oysters (55 to 141 ppm c.e.), *Cypria* spp. (7.5 ppm), various copepods (~5.4 to 25 ppm c.e.), odonates (>100 ppm c.e.), *Limnephilus* spp. (33 pm c.e.)*, Nymphulla responsalis* (87 ppm c.e.) or bloodworm larvae (>100 ppm c.e.). Since both amphipods and cladoerans are important in the food chain, the ecological relevance of adversely affected species may be high. However, Wilson (1967) indicated that the affected species may be replaced by species of a similar size class and therefore, the impact of the few species which may be eliminated from a water body, is likely to be minimal.

Diquat and its formulations have a low tendency to bioaccumulate in fish and crayfish with bioconcentration factors less than 1.0 for the whole body, 0.67 ppm for edible tissue and 2.5 ppm for the viscera. Typically, the bioconcentration factor for fish is 0.01 for the edible tissue of trout and carp is lower than that for bluegill sunfish (EPA RED, 1995). The tendency for bioaccumulation in bivalves, insects and daphnids is somewhat higher with the bioconcentration factor ranging from ~10 in oysters and freshwater clams to ~30 in mayflies (Hammer et al, 1987, Thompson, 1987). In most cases, the depuration half-life is one to three days. So bioaccumulation should not be an issue and there should be no biomagnification as diquat travels up the trophic levels. However, the current residue tolerance of 0.1 ppm may not be sufficient in bivalves and possibly crayfish (Fujie, 1987; Thompson, 1987). Biconcentration and residue levels in fish tissue, aquatic plants, terrestrial plants, and livestock are discussed in much greater detail in sections 4.2.2.4 and 4.2.5.

Diquat is usually applied in a water carrier or as an invert emulsion through boom trailing hoses carrying nozzle tips to apply the dilute spray below the water surface to ensure adequate coverage. While diquat is combined with other herbicide products in some cases, this is not allowed in Washington State. Typically, diquat may be combined with copper complexes for the control of *Hydrilla verticillata*, pondweeds (*Potamogeton* spp.), cattail (Typha spp.), naiad (Najas spp.), waterhyacinth (Eichhornia *crassipes*), Eurasian watermilfoil (*Myriophyllum spicatum*), duckweed (*Lemna spp.),* pennywort (*Hydrocotyle* spp.) or bladderwort (*Urticularia* spp.), common Salvinia (*Salvinia rotundifolia*) and other weed species specified in the label. The diquat plus copper complexes with the broadest spectrum of control include Komeen® Aquatic Herbicide and Nautique® Aquatic Herbicide. However, K-Tea™ Algaecide, Cutrine®-Plus Algicide, and Captain® Liquid Copper Algicide may be combined with diquat for the control of *Hydrilla verticillata*.

There have been very few cases where diquat has been shown to be synergistic. For example, diquat has been shown to be synergistic with Cutrine®-Plus on the brown trout (Simonin and Skea, 1977), and a variety of fish species in the field (Daniel, 1972). Diquat has also been shown to affect the mosquito fish (*Gambusia affinis*) in a synergistic manner when combined with parathion (Krieger and Lee, 1973 in Shearer and Halter, 1980). Also, diquat has been shown to be synergistic with the fungicide nabam in inhibiting the development, survival and growth of tadpoles and embryos *of Xenopus laevis* (Anderson and Prahlad, 1976). More detail on the effects of diquat and other pesticides as synergistic, antagonistic, cumulative or additive effects agents can be found in Section 4.2.4.

Although diquat is a one-electron transfer agent with potential to inhibit microsomal fraction oxidases (MFOs), very few cases of synergism have been documented with diquat. However, since diquat is an MFO inhibitor, the presence of accelerators/surfactants, other “inerts”, or other pesticides in either tank mix situations with diquat or incidental exposure from treatment with other pesticides may increase the potential for damage to the biochemistry or physiology of fishes. These potentiating effects could increase acute or chronic (early life-stage) toxicity or increase the biochemical or pathological effects of diquat in fish exposed to sub-acute dosages.

A number of sub-acute effects have been noted due to the exposure to diquat. In the yellow perch (*Perca flavescens*), even very low concentrations of diquat during a 96-hour exposure caused changes in the hematocrit, plasma corticol steroid concentrations and plasma glucose concentration (Holtz and Winter, 1989). These changes were seen at all exposure concentrations (0.1, 1.0, 10 and 50 ppm a.i. = 0.054, 0.54, 5.4 and 27 ppm c.e., respectively) during at least one point in time (0.16, 1, 2, 3 or 4 days). Similar observations were made for the grass carp exposed to diquat. However, effects on grass carp, including decreases in plasma and muscle protein levels, increases in plasma and muscle transaminase levels, and increases in hematocrit values were seen at exposure concentration of 53 ppm diquat (29 ppm c.e.). However, while the lower concentration of diquat (2.0 ppm) caused increases in the hematocrit levels, this concentration had no adverse impact on protein concentrations or transaminase levels in grass carp (El-Deen and Rogers, 1992 & 1993). Since the changes in grass carp biochemistry are only noted at concentrations that greatly exceed any likely environmental concentration, there does not appear to be real environmental relevance in these changes. However, since even very low concentrations of diquat cause changes in yellow perch biochemistry, it is likely that this species is highly stressed due to exposure to diquat. And such stresses can lead to greater susceptibility to predation, disease or environmental assault. Pathological changes in the tissues of Coho salmon included histopathological effects of eyes, kidneys, gills, and liver that indicated a stress from concentrations of diquat as low as 3.0 ppm c.e. Since 3.0 ppm diquat is close to the maximum use rate of 0.75 ppm c.e., it is possible that lower dosages may cause pathological conditions undetectable by standard necropsy but still of importance in interfering with the parr to smolt metamorphosis, which is a potential problem with Coho salmon even at concentrations of 0.5 ppm c.e. However, most of these effects can be considered minimal in absence of environmental assault from sources other than the presence of diquat at typical expected environmental concentrations (EECs). Typical EEC concentrations are much lower than the 3.0 to 53 ppm tested in most sub-acute toxicity studies. To discover the long term effects of diquat at environmentally relevant concentrations would require the conduct of multigenerational laboratory experiments with species considered to be ecologically sensitive.

Accelerators and thickening agents are rarely used with herbicides sprayed directly on the surface of a water body, but some applicators and scientists believe that surfactants like CideKick® and X-77® improve effectiveness and should be used with diquat products when surface (floating) weed control is necessary (Getsinger, 2000 personal communications). A thickener like Nalquatic® or Polysar® will often be used to allow a subsurface application to sink down into the water column where it will be most effective against rooted aquatic macrophytes. If the herbicide is sprayed on, thickeners also control potential drift. Although all adjuvants registered for use with aquatic herbicides should be safe to fish and other aquatic animals when used according to the label, they are not without risk to aquatic life (Watkins et al, 1985). Their 96-hour toxicity (LC50) ranges from 0.96 mg/L to > 1000 mg/L. In lakes and ponds with reasonable depth, dilution should prevent toxic effects from occurring due to the use of additives. This is particularly so if the control measure is a spot or margin treatment. A more detailed discussion of the effects of adjuvants can be found in Section 4.2.4 and in Table 9. Care should be taken that only non-ionic surfactants are used since ionic surfactants may react with diquat to decrease its effectiveness or damage sensitive animal biota.

4.3.2.1 Acute Effects on Aquatic Animals

- **Acute effects on fish**

  Toxicity information indicates that commercial products of diquat have limited acute toxicities to most species of fish tested (Table 2 and Table 18 and Appendix 1); that is, it has a LC50 greater than 10 ppm for most of the species considered to have high commercial or recreational value. Diquat has a 24- to 96-hour LC50 that ranges from ~10 to ~30 ppm for all the species of salmonid tested. These include rainbow trout, Coho salmon, Chinook salmon, brown trout and Eastern brook trout. Most other species including Northern pike (*Esox lusius)*, Muskellunge (*Esox masquinongy)*, black bullhead (*Ictalurus melas*) and bluegill sunfish are relatively tolerant of diquat's acute toxicity effects (LC50 =5.0 to 10 ppm). Some growth stages and species are very tolerant (LC50 = 10 to 156 ppm c.e.). However, bass species and the walleye (*Stizotedion vitreu*) can be very sensitive to diquat. For example, striped bass sac-fry have an LC50 of 0.54 ppm c.e.; smallmouth bass sac-fry have an LC50 of 1.3 ppm c.e.; largemouth bass fry have an LC50 of 4.9 ppm c.e.; and 8- to 10-day walleye have an LC50 of 0.75 ppm c.e.

  Based on these LC50s, diquat can be placed in the ecotoxicological risk categories as follows: highly toxic (LC50 = >0.1 ppm to 1.0 ppm) for the bass and walleye; moderately toxic (LC50 = >1.0 to 10 ppm) for *Esox* spp., bluegill sunfish, *Ictalurus* spp., yellow perch, and live-bearing toothed carp; slightly toxic (LC50 = >10.0 to 100 ppm) to American eel, zebra fish, cyprinid carp, northern pike, shiners, salmonids, and fathead minnows; to practically non-toxic LC50 = >100 ppm) for mosquito fish (*Gambusia affinis*). These risk category classifications do not mean diquat will or will not have an adverse impact to fish when they are exposed to the expected environmental concentration. This determination of risk compares the general toxicity of diquat with other registered pesticides; based on this comparison, diquat is moderately toxic to slightly toxic to most species of fish.

  The application rate for diquat in the United States to control aquatic macrophytes typically ranges from ~0.1 to 0.75 ppm. Typical use rates in the United States are around 0.224 ppm for whole pond treatments and 0.30 to 0.37 ppm c.e. for spot treatments in reservoirs or canals (Campbell et al, 2000). Therefore, fish biota should be largely unaffected by normal treatments with less than 50% mortality occurring when the most sensitive species are present during treatment.

Although the risk quotient level of concern of 0.1 is exceeded with the most sensitive species, less than 50% of the most sensitive species would be affected since the RQ is less than 1.0. However, if a safety factor is necessary, the use of the diquat would not be acceptable unless mitigating factors could be considered. An acceptable mitigating factor would be the assumption that a 4-day geometric mean of the EEC gives a more accurate picture of real exposure. These assumptions are investigated further in Section 4.3.2.5 where the final conclusion is that diquat does not pose a significant risk to the fish biota since the EEC is less than or equal to the LC50 for the most sensitive species and growth stage.

- **Acute effects on aquatic invertebrates**

  Toxicity information indicates that the commercial products of diquat have a slight acute toxicity to most species of aquatic invertebrates tested (Table 2, Table 19 and Appendix 2). That is, they have a LC50 greater than 10 ppm for most of the species considered to have high ecological value as fish food (Table 19 and Appendix 2). Diquat has a 24- to 96-hour LC50 that ranges from ~10 to ~100 ppm for most non-crustacean and non-mollusk species tested. Exceptions to this appear to be with sediment (benthic) organisms like the amphipod (*Hyalella azteca*) with a 96-hour LC50 = 0.048 ppm c.e., pocket shrimp (*Mysidopsis bahia*) with a 96-hour LC50 = 0.42 ppm c.e. and the apple snail (*Pomacea pludosa*) with a 96-hour LC50 of 0.34 ppm c.e. A number of free swimming cladoerans also appear to be acutely affected, including *Daphnia magna* with 48-hour LC50 = <1.0 ppm c.e. and *Daphnia pulex* with a 48-hour LC50 = 0.16 ppm c.e.

  Based on these LC50s, diquat can be placed in the following ecotoxicological categories: Very highly toxic (LC50 = <0.1 ppm) to *Hyalella azteca*; Highly toxic (LC50 = 0.1 to 1.0 ppm) for the cladoerans, pocket shrimp and mollusk; Moderately toxic (LC50 = >1.0 to 10 ppm) for *Cypria* spp. and *Diapotomus* spp.; Slightly toxic (LC50 = 10.0 to 100 ppm) for mayfly, oysters, *Eucyclops* spp., *Gammarus fasciatus*, caddisfly, and *Nymphula responsalis*; and Practically non-toxic for damselfly, dragonfly, and bloodworms. These risk categories do not imply that diquat will or will not have an adverse impact on these specific groups of invertebrates when they are exposed to the expected environmental concentrations (EEC). However, this determination of risk compares the general toxicity of diquat with other registered pesticides; based on this comparison, diquat has a high to very high acute toxicity to cladoerans, *Hyalella azteca*, apple snail, pond snail and pocket shrimp. However, the toxicity to other species tested was moderately toxic to practically nontoxic.

  Even if a very liberal federal drinking water standard (MCGL = 0.02 ppm c.e.) is used as being typical of pore water or over-lying water concentrations, the most sensitive benthic species may still be affected by environmental concentrations of diquat since the LC50 exceeds the EEC (MCLG) by only two-fold. Diquat may partition to the sediment at typical concentrations of 25 ppm c.e. However, sediment-bound diquat is generally considered to be inactive and biologically unavailable (Dyson and Takacs, 2000). Therefore, the only concentrations likely to impact free-swimming invertebrates and benthic organisms would be found in the water column. In the worst field case, concentrations of diquat in Lake Steilacoom were typically ~0.05 ppm c.e. for 1 to 3 days after application of diquat at 0.13

ppm c.e. Since this concentration exceeds the 96-hour LC50 (0.048 ppm c.e.) for the most sensitive species, adverse impact is likely for the sediment invertebrate biota.

It is likely that diquat will have adverse impact on benthic invertebrates even if further analysis is conducted. However, depending on half-life considerations for diquat and the manner in which diquat is applied, it may prove to be safe to the free-swimming (zooplankton) biota. The concentrations of diquat found in water shortly after treatment can vary considerably depending on the treatment rate, rate of uptake and release from plant material, and mass of water movement through the treatment area. For example: the diquat concentrations seen in the water of Lake Chautauqau, New York was <0.001 ppm c.e. one day after a maximum use rate application to the periphery of the lake (Sewell, 1969); in a whole lake treatment of Lake Steilacoom at the maximum use rate, diquat was detected at concentrations ranging 0.033 to 0.059 ppm after three days of dissipation (Serdar, 1997). More detailed discussion of the levels of diquat found in both water and sediment can be found in Sections 4.2.2.1 and 4.2.2.2. For a determination of risk see Section 4.3.2.5.

The exact toxicity categories may not be of great importance, since the evaluation presented here does not constitute a risk assessment and exposure to diquat is likely to be negligible for most species even though potential exposure concentrations exceed the LC50 in the more sensitive species tested. The most sensitive species appear to be benthic and sediment invertebrates and free-swimming invertebrates. The toxicity of diquat to these species has been discussed extensively in early paragraphs of this section.

When environmental concentrations dramatically exceed the LC50 with the most sensitive species, the use of the compound would not be acceptable unless mitigating factors could be considered. Fortunately, whole lake applications are not considered to be a typical exposure scenario. The most typical exposure scenario is a spot treatment in reservoirs or canals at 0.30 to 0.37 ppm c.e. Under these scenarios, one would generally expect concentrations of diquat to dissipate in northwest waters to time-weighted EEC levels of 0.042 to 0.021 in 48 to 96 hours (Ritter et al, 2000). At these concentrations, even *Hyalella azteca* is likely to be spared from the direct toxic effects of diquat. However, for this protection to occur, diquat would have to be used as a restricted use herbicide, which is described in the Reward® LA label. These concentrations are far more acceptable. However, even at these more reasonable rates, the most sensitive species may not be protected. A formal risk assessment in Section 4.3.2.5 supports the conclusions in this toxicity review.

4.3.2.2 Chronic Effects of Diquat on Aquatic Animals

- **Chronic effects on fish and amphibians**

To this date, the amount of chronic or early life-stage effect data for diquat on aquatic animals (fish) is rather minimal (Table 2, Table 20 and Appendix 3). Most studies deal with early life-stage (egg, egg to sac-fry or egg to free-swimming fry). There are studies that deal with the early life stage (egg to fry) toxicity of diquat in the fathead minnow, channel catfish, eastern brook trout, killifish, leopard frog and African clawed toad.

Early life-stage (egg to fry) toxicity of diquat was investigated with the fathead minnow. After 34 days of exposure the no observed effect concentration (NOEC) maximum allowable toxin concentration (MATC) and lowest observed effect concentrations (LOEC) were 0.12, 0.2 and 0.32 ppm c.e., respectively, for weight reduction. For eastern brook trout, these values were 2.7, 3.8, and 5.4 ppm c.e. respectively, for abnormal sac-fry development. For the channel catfish, the LOEC was >1.0 ppm c.e.; so, the NOEC and MATC could not be determined for channel catfish. For rainbow trout, the LOEC was <1.4 ppm c.e.; so the MATC and NOEC could not be determined but the Chronic LC50 was determined to be 2.9 ppm c.e.

For the killifish, accurate early life-stage toxicity values could not be ascertained because all fish eventually died at concentrations of 0.0054 ppm c.e. However, the NOEC, MATC and LOEC for abnormal pigmentation and slow development were determined to be 0.0054, 0.017 and 0.054 ppm c.e., respectively.

In the leopard frog, the 14-day early life-stage toxicity values for NOEC, MATC and LOEC were 1.08, 1.7 and 2.7 ppm c.e., respectively, for developmental abnormality and survival. In a chronic toxicity study based on 15-day tadpole survival, chronic toxicity values for the NOEC, MATC, LOEC and LC50 were all greater than 5.4 ppm c.e.

For the African clawed toad, accurate early life-stage toxicity values can be ascertained because the LC50 (0.41 ppm c.e.) for this test was lower than the LOEC (0.68 ppm). Therefore, the NOEC, MATC and LOEC for abnormal pigmentation, body size reduction, altered body shape, muscular development and survival were 0.54, 0.64 and 0.68 ppm c.e., respectively.

Not all of the above listed studies are of adequate design to pass current EPA guidelines as early life-stage studies. The only well designed study was an early life-stage study with fathead minnow. The other studies did not yield adequate information for evaluation of chronic effects either because the duration of the study was less than 28 days or the mortality effects were high enough to interfere with the developmental effects that are most important in chronic studies. The studies conducted by Hiltibran (1967) provide good supplemental data but were not conducted for a long enough periods of time to be considered chronic studies. However, Hiltibran’s data will be considered definitive if it is the only data available with a particular product and a particular species. In these studies the 12-day NOECs and LC50s were >11.0 ppm c.e. for bluegill sunfish.

The EEC for diquat will be quite low since applications at environmentally relevant rates will not typically be measurable after 1 to 12 days of dissipation since the half-life under most use scenarios is less than 1 day. Although formal calculations were not found in the literature to predict the long-term geometric mean EEC after 12, 13, 15, 21, 28, 34 or 40 days, we have calculated these values to be 0.02, 0.019, 0.016, 0.012, 0.0087, 0.0071 or 0.0061 ppm c.e., respectively. These EEC values were calculated assuming an application rate of 0.224 ppm c.e. and a half-life of 0.75 days. These EEC values assume EPA’s worst case scenario of a maximum use rate application to a water impoundment with a depth of 200 cm deep impoundment. In this case, the EEC (0.0087 ppm c.e.) for the 28 day standard assumption is less than the lowest NOEC (0.12 ppm c.e.), MATC (0.20 ppm c.e.) or NOEC (0.32 ppm c.e.). Therefore, there should be little or no impact to fish and amphibian species from exposure to diquat at typical chronic field rates.

However, since the database is so small, one cannot say that credible risk does not exist for chronic exposure of fish to these products. Further research to expand this database on the chronic toxicity of diquat to fish needs to be conducted to give the chronic toxicity MATCs greater credibility. Typical tests that would be conducted are early life-stage tests with fathead minnow, rainbow trout and sheepshead minnow. These species are easy to rear in the laboratory and the procedures for conducting early life-stage tests are accepted by state and federal regulatory agencies. A formal risk assessment in Section 4.3.2.5 supports the conclusion of this toxicity review.

- **Chronic effects on Invertebrates**

  The amount of data that has been generated on life-cycle effects of diquat on aquatic invertebrates is minimal (Table 2, Table 21 and Appendix 4). Twenty-one day life-cycle tests have been conducted on diquat with *Daphnia magna*.

  The life-cycle NOEC, MATC and LOEC for diquat are 0.036, 0.045 and 0.057 ppm c.e. *Daphnia magna*. These values are well above the maximum expected exposure concentration (EEC) of 0.012 or 0.0087 ppm c.e. for 21 or 28 days, respectively. Therefore, there should be little or no impact to invertebrates.

  However, since the database is so small one cannot say that credible risk does not exist for chronic exposure of invertebrates to diquat. Further research on the chronic toxicity of diquat to aquatic invertebrates needs to be conducted to give the life-cycle NOECs greater credibility. Additional tests that should be conducted include life-cycle tests with *Ceriodaphnia dubia* and the mysid shrimp. These species are easy to rear in the laboratory and the procedures for conducting life-cycle studies are accepted by state and federal regulatory agencies.

  Chronic risk analysis for diquat is not necessary since it is not likely to be encountered on a chronic basis. However, it may is worthwhile to conduct a chronic risk assessment assuming that the acute to chronic ratio (19.0) for all invertebrates is the same as that determined for *Daphnia magna*. However, life-cycle tests conducted with species of known sensitivity like *Daphnia magna, Ceriodaphnia dubia* and mysid shrimp would improve the credibility of this risk analysis. A formal risk assessment in Section 4.3.2.5 supports the conclusion of this toxicity review.

4.3.2.3 Impacts of Single versus Multiple Applications

It would be extremely rare for lakes in Washington State to be treated with diquat products more than once in a season. Therefore, very little practical field knowledge is known on this subject. However, some field work has involved both multiple treatments at 0.25, 0.5 and 1.0 ppm and treatments at high concentrations (3.0 ppm). Treatment with diquat once or twice during a season had little effect on the survival, abundance, size structure, condition or movement of largemouth bass in Guntersville reservoir when the treatment rate was diquat plus Cutrine® at 0.3 ppm c.e. plus 0.3 ppm copper (Bain & Boltz, 1992).

A one-time treatment of a Wisconsin pond with 1.0 ppm diquat had no significant effect on adult and fingerling bluegills that could be attributed to the use of diquat. Survival was actually higher in treated ponds than in control ponds, the average weight and length were similar for fingerlings, and total harvest weight was similar in treated and control ponds. However, adult fish were distinctly smaller in treated ponds than in control ponds. Histopathological examination found no differences in tissues or major organs that could be attributed to diquat use. However, in similar ponds multiple (two to three) treatments with diquat spaced 8 weeks apart did not have obvious affects on survival or histopathology. Nevertheless, adults grew more slowly in the ponds that were treated with multiple applications of diquat with the final length and weight being at least

2.7-fold less in treated ponds (0.15 inches and 2.34 grams) than in control ponds (0.41 cm and 6.35 grams) (Gilderhaus, 1967).

Two treatments of Spooner Pond in Wisconsin at 0.25 ppm c.e. produced no ill effects to either vertebrates or invertebrates. The invertebrates involved were *Daphnia pulex*, ostracods, copepods and snails; the vertebrates involved included tadpoles, suckers, perch, bluegill and muskellunge fingerlings. The use of diquat has potential to prevent the entrance of northern pike and perch into Spooner Pond but currently this is just a hypothesis. The survival of *Daphnia pulex* was surprising in light of diquat's high toxicity to this species (Johnson, 1962).

Additional laboratory work indicates that chronic exposure at high concentrations of diquat (mimicking multiple exposures) may cause both pathological and biochemical signs of stress in the grass carp (El-Deen and Rogers, 1992 and 1993). This exposure was very high (53 ppm diquat = 29 ppm c.e. for 7 days). Furthermore, Holtz and Winter (1989) saw signs of stress including increased plasma hematocrit, plasma cortisol and plasma glucose levels in yellow perch exposed to concentrations of 0.1 to 50 ppm diquat (0.054 to 27 ppm c.e.) for at least one time period in 4, 24, 48 or 96 hours. Although an exposure of 53 ppm is clearly higher than would be encountered in the environment, 0.1 ppm diquat is a concentration that may have ecological impact on sensitive organisms. These environmental assaults have the potential to cause additional stress making sensitive species more susceptible to parasites, disease, predators, and other pesticides.

Studies conducted with fathead minnow, rainbow trout and yellow perch indicate that exposure to diquat can slow the speed of swimming, change the rheotaxic response or cause changes in respiratory function. Concentrations as low 0.5 ppm c.e. can decrease swimming speed in rainbow trout from 22.6 to 14.1 cm/sec and marginally effect the null response to rheotaxic stimuli (Dodson & Mayfield, 1979). In the fathead minnow, at least 9.2 ppm c.e. is necessary to decrease the swimming speed from 11.4 fish lengths/sec to 5.8 fish lengths/sec and to significantly increase the rheotaxic response from an average of 36 sec/2 min period to 82 sec/2 min period (de Peyster and Long, 1993). Exposure of yellow perch to 1.0 ppm diquat causes an increase in the coughing rate with as little as 6.0 hours of static exposure. Increases in coughing rate from ~2 coughs per hour in the control to ~30 per hour after 72 hours of static exposure to diquat have been seen (Bimber et al , 1976). Although concentrations as high as 9.2 ppm probably have low environmental relevance, concentrations as low as 0.5 to 1.0 ppm c.e. may be encountered in the environment. Furthermore, changes in respiratory effectiveness, swimming speed and rheotaxic response can influence escape from predators, migration patterns, feeding, mating and breeding, etc.

Effects of diquat on the time to maximum population development in *Daphnia pulex* has also been noted. For example, maximum population development of *Daphnia pulex* in Wisconsin typically occurs in mid-June but treatment one or three times at 1.0 ppm c.e. delays this maximum population development to mid-October or early August, respectively. One would normally expect the maximum bluegill sunfish fingerling population to occur in mid-June as well. Such differences in the development time between predator and prey species has a potential to produce adverse effects in wild populations. If the spawning time is early for the predator species and the development time is late for the prey species, the prey may not be developed to an appropriate size when the predator fish fry need nutritional input most (Gilderhaus, 1967).

Potential impacts on populations

Wilson (1967), Hilsenhoff (1966), Tatum and Blackburn (1962), Gilderhaus (1967) Berry et al (1975), and Shearer and Halter (1980 cite Newbold, 1975 and Water Investigations Branch, 1977) reviewed a number of field studies on the effects of diquat against aquatic invertebrates (mainly benthic invertebrates). For example, diquat appears to be extremely toxic to the amphipod *Hyalella azteca* in northwest waters. *Hyalella azteca*, when caged as sentinel organisms, were killed by exposure to diquat plus paraquat at concentrations of 0.5 plus 0.5 ppm.

Wilson (1967) and Berry et al (1975) found that *Hyalella azteca* decreased in numbers when ponds in Oregon and the Chickahominy Reservoir in Virginia were treated with 2.5 ppm diquat or 0.1 ppm diquat plus 0.17 ppm endothall. The population of *Hyallela azteca* in the Soap Creek ponds of Oregon decreased in absolute terms by 292 organisms/meter^2 in the treated plot and increased in absolute terms by 33 organisms/meter^2 in the control plot. This change in numbers took a total of 8 days to completion. However, two-thirds of this decrease occurred by the day after treatment. Similar decreases occurred in shallow water areas of the Chickahominy Reservoir. In the deep water areas, the numbers of *Hyalella azteca* actually increased from 51/unit area to 71 per unit area. This difference in survivorship was attributed to high concentrations of diquat in the shallow area (0.73 ppm 12 hr after treatment) and a much lower concentration (0.03 ppm) in the deeper area. While direct toxicity of diquat can account for the decrease in *Hyalella azteca* numbers, the noted decrease in plant numbers and dissolved oxygen content may have contributed to the decrease in amphipod numbers. Hilsenhoff (1966) found that treatment of a pond in Wisconsin at 1.0 ppm diquat resulted in marked decreases in the numbers of the amphipod *Hyalella azteca*, and four genera of pond snail collected per unit area. However, Hilsenhoff attributed this decrease in amphipod and gastropod numbers to the elimination of habitat due to the herbicidal effects on *Elodea canadensis* rather than to the acute effects of diquat.

A number of studies have noted changes in the number of cladoerans after treatment with diquat. Newbold (1975 in Shearer and Halter, 1980) found that *Simocephalus vetulus* was eliminated from ponds treated with 1.0 ppm for up to 64 days after treatment. It is likely that this extremely sensitive cladoeran species was killed by the direct toxic action of diquat. In another species of cladoeran (*Daphnia pulex*), the treatment of ponds in Wisconsin with 1.0 ppm had no adverse impact on numbers. However, the time of peak cladoeran abundance was shifted from mid-June to mid-October. Multiple treatments with diquat at 1.0 ppm separated by 8 weeks resulted in a marked decrease in the number of cladoerans relative to the control. Control ponds had an average number of cladoerans of 221 animals/L while ponds treated one, two, or three times had 602, 82 and 136 animals/L (Gilderhaus (1967).

Numbers of some species of invertebrates appeared to increase due to treatment. For example, glassworm (*Chaoborus* spp.) numbers appeared to almost double from 104 to 203 animals/unit area after treatment with diquat at 0.5 ppm. This effect was believed to be due to the increase in decaying plant (southern naiad) matter, which chaoborids used for foodstuff. When chaoborid populations increased significantly, there was a concurrent decrease in the number of oligochaete worms, which was hypothesized to be due to the effects of competition for food and possibly space (Berry et al, 1962).

Although a number of authors (Wilson, 1967, Hilsenhoff, 1966 and Berry et al, 1975) have monitored a wide diversity of species in the field, no adverse impact was noted on the numbers of these organisms. Any increases or decreases in the populations of these species were attributed to natural seasonal rise and fall seen in all animal populations. Some of these species that were not affected in the field included may flies (*Caenis* spp. and *Callibaetis* spp.), damselflies (*Enallagma* spp. and *Ischnura* spp.), true bugs including *Merrigata* spp., *Mesovleia* spp., *Gerris* spp., *Microvelia* spp., *Plea* spp. *Belostoma* spp. and *Sigara* spp., beetles including *Haliplus* spp. and *Hydrovatus* spp., flies including *Chironomus* spp., *Tanytarsus* spp. and *Tanypus* spp., various copepods and various ostracods. The fact that these species are not affected adversely in the field is not surprising in view of their typical low response to diquat (typical 48 to 96-hour LC50 = 6.8 to >100 ppm c.e.). However, as previously discussed *Hyalella azteca* and various snails and water fleas are typically affected adversely at concentrations that exceed their LC50s (0.048 to <1.0 ppm c.e.). Furthermore, field studies have been noted to adversely impact these species at treatment concentrations that range from ~0.11 to 2.5 ppm c.e.

There is not a large literature base concerning negative or positive impacts of diquat treatment on numbers of fish and invertebrates in natural ecosystems. However, Bain and Boltz (1992) noted that treatment of the Guntersville Reservoir in Alabama with diquat plus Cutrine®-Plus at 0.3 ppm c.e. plus 0.3 ppm copper for the removal of *Hydrilla verticillata* caused no changes in abundance, size structure, condition or movement of largemouth bass. Similarly, Gilderhaus (1967) found no significant differences in the number of fingerling or adult bluegills after treatment with 1.0 ppm c.e. diquat for the removal of American waterweed. Although their work on waterhyacinth removal does not apply to Northwest waters, Olalaye et al (1993) found that treatment of the Kofawei Creek in Nigeria with 2.0 Kg a.i./ha allowed for an increase in the number of fish collected per unit area is from 8 to 67 animals/unit area. Furthermore, there is data on the effect of failure to remove weeds when they become so dense they interfere with the action of indigenous fishes, but even this data is ambiguous. Klussmann et al (1988 in Bain and Boltz, 1992) found that catch rates for largemouth bass was greatest when the plant densities were highest, while Colle et al (1987 in Bain an Boltz, 1992) found that largemouth bass catches were unaffected by a reduction in plant density. Ideal plant cover of about 36% appears optimal for production of largemouth bass (Ware and Gasaway, 1978 in Bain & Boltz, 1992) and complete removal of aquatic plants can cause a major decline in forage fish and largemouth bass abundance (Moxley and Langford, 1985 in Bain & Boltz, 1992). There can be a decrease in the numbers of certain size classes (intermediate size largemouth bass) and not others (large largemouth bass) if foliage is entirely removed (Klussmann et al, 1988 in Bain & Boltz, 1992). Swingle (1950 and 1956) has found that while a certain amount of foliage cover and filamentous green algae (foodstuff) is necessary to achieve the ideal forage to predatory fish ratio of 3 to 6, the ideal amount of forage and foodstuff algae varies significantly between water bodies. Swingle did not specify what might cause those variations but it can be conjectured to be due to size and depth of the water body, and possibly due to other factors like transparency, and size distribution of predator and prey animals.

Negative impacts were seen due to the treatment of ponds in Wisconsin with diquat for the control of Eurasian watermilfoil. Treatment with diquat plus Cutrine® at concentrations of 3.0 ppm plus 2.2 ppm resulted in a complete kill of the resident fish population in field microcosms. It is unclear if this is due to synergistic effects or low dissolved oxygen concentrations or a combination of these two factors. Frank (1972) is of the opinion that fish-kills of this nature are usually due to anaerobiosis due to decaying vegetation and not to the effects of a registered aquatic herbicide.

- **Potential impacts on diversity**

  As described in the Nigerian work with waterhyacinth, high infestation rates with weeds can affect both fish numbers and fish diversity (Olaleye et al, 1993). Details on the effects of weed infestation on numbers can be found in potential impacts on numbers section.

  A detailed study on the numbers and diversity of fish conducted by Olaleye et al (1993) concluded that areas heavily infested with waterhyacinth contained a very low number (8 per unit area) of the *Ctenopoma kinglayae* (Anabantid), and no other fish species. However, if waterhyacinth had been removed by the application of diquat at 2.0 Kg a.i/ha, the numbers of fish went up to 67 per unit area and five other species of fish were seen. Many of these species were seen in relatively large numbers including *Gymnarchus niloticus*, *Channa obscura*, and *Heterotis niloticus*. Since this work was conducted in Nigeria, it may not be directly applicable to the State of Washington.

  Information on the changes in numbers of fish and invertebrates due to diquat treatment has been gathered, but the changes in diversity have not been addressed. As reported previously, by Berry et al (1975) and Hilsenhoff (1966), the numbers of individuals in any species may decrease due to the direct or indirect effects of diquat treatment, but diversity between treatment and non-treatment areas is often unaffected. Although the number of invertebrate taxa remained the same throughout these experiments it is unclear how diversity was determined. However, general inspection of the data in these two studies would indicate that the conclusion of no change in diversity between treated and untreated study areas is probably correct. This lack of change in diversity was seen at both low (0.11 ppm diquat plus 0.17 ppm endothall in the Chickahominy Reservoir) and at high treatment concentrations (1.0 ppm diquat in ponds located in Wisconsin).

- **Potential impacts on habitat use for spawning, rearing and growth**

  - **Effects on Trout and other salmonids**

    Diquat has been shown to have low acute and early life-stage toxicity to freshwater trout. Diquat has an acute toxicity (96-hour LC50) to trout of 6.1 to 18.7 ppm c.e. and a chronic toxicity (21-day LC50) of 2.9 ppm c.e. Since diquat is not likely to be encountered at concentrations higher than 0.224 to 0.75 ppm c.e., this product is unlikely to cause toxicity in freshwater trout. Due to diquat's very short half-life, salmonids are not likely to be exposed to expected environmental concentrations (EEC) as high as those given above. The typical 4-day geometric mean EEC under EPA's worst case scenario would not be higher than 0.059 ppm and experimental data indicate that the concentrations (0.0325 to 0.0912 ppm c.e.) of diquat in Northwest waters (Lake Steilacoom) would be similar in magnitude (Serdar, 1997). It is noteworthy that this EEC calculated by Shaw et al (1995) is slightly higher than the value that we have calculated. Modeling work by Campbell et al (2000) indicates that the 4-day geometric mean EEC for a spot treatment at 0.30 to 0.37 ppm c.e. would not be higher than 0.021 ppm and indications are that the concentrations (0.0006 to 0.0028 ppm c.e.) of diquat in Northwest waters (Gravelly Lake) spot treated with diquat would be somewhat less than this value (Serdar, 1997).

    Trout fry have not been noted to avoid diquat at concentrations that are typically environmentally relevant (0.1 to 1.0 ppm c.e.) (Folmar, 1976). Furthermore, no avoidance of diquat by trout at field concentrations of 26.7 ppm was noted. However, these trout did not display any adverse reaction to the presence of diquat for 24 hours. Some species of fish (carp suckers and other scrap fish) will avoid diquat in the field at extremely high concentrations (26.7 ppm). However, the environmental relevance of this observation is not entirely clear. Avoidance may not be possible in real field treatment situations. Fish driven from a habitat by avoidance behavior, may not be able to obtain necessary resources for survival in other habitats. These resources may include food, refuge, mates and appropriate egg-laying (substrate).

Probably the greatest concern is managing aquatic plants so that maximum breeding opportunities can occur. Although it typically takes concentrations of 0.25 to 1.0 ppm c.e diquat to effectively manage aquatic foliage, exposure (starting in June of any given year) of ~0.1 mg/L will control American waterweed and most species of pondweed in Northwest waters [(Lake Steilacoom and Gravelly Lake (Serdar, 1997)]. However, the degree of control was not discussed by Serdar. The consequences of eliminating such plants from a habitat can have tremendous consequences. Due to the effects of erosion by floods, the character of a habitat may be changed from one suitable for the reproduction of sunfish to one suitable for the reproduction of salmonids.

If water that contains diquat at effective concentrations passes from a lake or pond into a river or stream, rooted aquatic macrophytes may be destroyed. This can have a substantial impact during the next high water event. Normal spring floods in absence of rooted aquatic macrophytes can dig up and kill large numbers of benthic organisms while summer spates can completely denude streams of benthic biota. This damage to the benthic biota cannot be prevented if aquatic macrophytes are removed by the effects of herbicides.

It was noted that at 24 hours after application, the concentrations of diquat in Lake Steilacoom (Washington) was the highest at the outlet to Chambers Creek and that this concentration was higher than at any other station within the lake. Furthermore, it is estimated that the MCLG (0.02 ppm c.e.) would be exceeded for at least 10 days after treatment and that the concentrations that may affect the macrophyte (0.0014 to 0.02 ppm c.e.) and algal biota (0.001 ppm c.e.) would also be exceeded. Due to these high concentrations of diquat at the lake outlet into Chambers Creek, damage to the macrophyte biota is likely with subsequent damage by hydraulic action to the benthic biota and character of the habitat.

- **Effects on salmon smoltification**

  Evidence for effects on salmon smoltification is of great potential concern with herbicides that are applied in Northwestern waters. Three seawater challenges have been performed with salmon species exposed to ecologically relevant concentrations of diquat. The data appear to conflict both between species and within species. Lorz et al (1979 in Shearer and Halter, 1980) found that Coho salmon smolts exposed to 5.0 to 20 ppm c.e. for 144 hours experienced 15 to 87% mortality in a subsequent seawater challenge test. However, later tests by the Lorz et al (1979) showed that exposure of Coho salmon to 0.5 to 3.0 ppm diquat did not produce subsequent mortality in a sea water challenge test and that these concentrations did not affect the level of gill ATP-ase. Similar tests on Chinook salmon smolts exposed to 0.125 to 0.5 ppm c.e. diquat for 96 hours did not produce mortality in subsequent seawater challenge tests. Furthermore, Chinook salmon smolts exposed to diquat maintained plasma sodium (170 meq/L) at levels that were consistent with normal seawater adaptation by healthy Chinook salmon smolts (Meier, 1997).

  Nevertheless, the exposures of Coho salmon smolts to concentrations of diquat as low as 0.5 ppm for 96 or 285 hours inhibited the downstream migration of a significant portion of the exposed fish. Inhibition of downstream migration is dosage related with only 50% migrating downstream after exposure to 0.5 ppm c.e. diquat for 285 hours and only 20% migration downstream after exposure to 3.0 ppm c.e. diquat. Sixty to 70 % of typical control salmon smolts will migrate downstream.

  These results indicate diquat products used for control of aquatic weeds may impact the smoltification process primarily by interfering with downstream migration. Although few species relevant to the Northwest have been tested, Coho salmon smolts may be sensitive to diquat under concentrations which may be encountered in the environment.

  Relatively high concentrations of diquat can produce histopathological changes in the tissues and organs of Coho salmon. Concentrations of 3.0 ppm diquat produced histopathological effects on eyes, kidneys, gills, and liver (Lorz et al, 1979). These effects were not seen at lower concentrations. However, subtle histopathological effects that may be encountered in the environment at the lower rates could adversely impact the ability of these salmon to escape from predators, pursue prey, find mates, breed, and resist disease or environmental assault due to pollution from other pesticides or other anthropogenic sources.

- **Effects on sea-run cutthroat trout and sea-run steelhead trout**

  No work was found on the effect of diquat on sea-run cutthroat trout or sea run-steelhead. General acute toxicity data has been observed on a few andromonous species of fish including the American eel (*Anguilla rostra*), Coho salmon (*Oncorhynchus kisutch*), Chinook salmon (*Oncorhynchus tshawytsha*) and striped bass (*Morone saxatilis*). The LC50s for these species are 17 to 30, 21 to 30, 16 and 0.54 to 43 ppm c.e., respectively. The most sensitive stage of striped bass is sac-fry and the least sensitive stages are fingerlings and sub-adults. Based on the toxicity of diquat to other salmonids (LC50 = 6.1 to 30 ppm c.e.), diquat is likely to be only moderately toxic to slightly toxic to sea-run cutthroat trout. Since rainbow trout were unaffected by 26.7 ppm diquat in the field, the andromonous strain of this species (steelhead trout) are also unlikely to be affected by field exposure to this concentration (Hesser et al, 1972). However, additional information on the acute and chronic toxicity of diquat products to cutthroat trout, steelhead trout and other andromonous species would be useful to aid in risk assessment with either these or related species.

A potential complicating factor with sea-run cutthroat trout *(Oncorhynchus clarki*), and steelhead trout (*Oncorhynchus mykiss*) is similar to the parr to smolt metamorphosis except that these sea-run trout may go through this process several times in their life-time including each time the adults migrate to the sea and the initial parr to smolt metamorphosis. Based on the seawater challenge tests and migration inhibition tests with Coho salmon, this seawater to freshwater to seawater metamorphosis may be an issue with some strains and species of andromonus fish. Therefore, depending on one's confidence in the likelihood that environmental exposure of diquat will interfere with freshwater to saltwater to freshwater metamorphosis further research may be warranted.

- **Effects on other species (sunfish, minnow and catfish)**

The acute and chronic toxicities of diquat are very low in fish. The acute LC50s range from 0.54 to 70 ppm c.e. for most ecologically relevant stages and species. The most sensitive stage/species is the sac-fry of the striped bass. The less sensitive stages/species are subadults of striped bass (LC50 = 43 ppm c.e.), subadults of the largemouth bass (LC50 = 60 ppm c.e.) and subadults of the bluegill sunfish (LC50 = 52 ppm c.e.).

It is noteworthy that changes in water hardness can strongly affect the toxicity of diquat (Table 8). For example: the LC50 of diquat in soft water (22-45 ppm $CaCO_3$) is 7.6 and 14-39 ppm c.e. for fathead minnow and bluegill sunfish, respectively, but the LC50 of diquat in hard water (340-379 ppm $CaCO_3$) is 70 and 76 ppm c.e., respectively. While differences in toxicity are probably seen in other species due to the effects of water hardness, most reports did not indicate the degree of water hardness maintained during the test. Since the LC50 (0.54 ppm c.e.) for even the most sensitive stage/species of sunfish (striped bass sac-fry) is approximately 10-fold higher than the 4-day geometric mean EEC (0.059 ppm c.e) for EPA's worst case scenario, it is unlikely that these typical short-term EECs of diquat will adversely affect sunfish, minnow or catfish biota.

The chronic MATCs for diquat are greater than or equal to 0.2 ppm c.e. for all ecologically relevant fish species tested (Table 20). Since the 28-day geometric mean EEC (0.0087 ppm c.e.) is lower than this MATC value, it is unlikely that the typical long-term EECs of diquat will adversely affect sunfish, minnow or catfish biota. Since very few environmentally relevant species were tested for long-term toxicity, an accurate assessment of chronic affects is difficult. However, due to the short dissipation half-life [(0.75 days) under typical EPA worst case scenario)], it is unlikely that chronic exposure of fish will occur in the environment.

A potential concern is the effects of diquat on behavior and metabolic responses in wild fish. A relatively high concentration of diquat (9.18 to 35 ppm c.e.) impaired swimming performance (speed) in fathead minnow (*Pimephales promelas)* and caused an increase in rheotaxic response immediately after exposure (Peyester and Long (1993). In the rainbow trout, diquat at an environmentally relevant concentration (0.5 ppm c.e.) also impaired swimming performance (speed) and increased the number of null responses to rheotaxic stimuli (Dodson and Mayfield, 1979). Avoidance behavior was also seen in goldfish exposed to diquat concentrations as low as 1.10 ppm c.e. (Berry et al, 1984) and scrap fish (carp and suckers) were seen to avoid diquat applied in the field at 26.7 ppm (Hesser et al, 1972). Diquat at concentrations of 1.0 ppm induced respiratory distress in yellow perch; the degree of distress was proportional to the exposure time with 5 coughs/hour occurring after six hours after static exposure and 30 coughs/hour occurring 72 hours after static exposure (Bimber et al, 1976). Holtz and Winter (1989) found that yellow perch exhibited physiological (biochemical) stress responses when exposed to concentrations of 0.1 to 50 ppm a.i diquat. These biochemical changes included increases in the blood hematocrit, plasma cortisol and plasma glucose levels.

As discussed previously, Bain and Boltz (1992) found that treatment with diquat plus Cutrine®-Plus in areas of *Hydrilla verticillata* infestation had no significant effects on abundance, size structure, condition, or movement of largemouth bass within the Guntersville Reservoir, Alabama. This may not be the case for all species of fish and invertebrates since carp, goldfish and suckers will avoid diquat when they are exposed in the field (Hesser et al, 1972). However, since the concentrations used by Hesser et al were so much higher than those currently used in the field, it is unlikely that significant numbers of exposed fish or invertebrates would or could avoid exposure to diquat under current application rates.

Effects on the feeding behavior in fish have also been discussed. Although grass carp were only minimally affected by diquat (96 hour LC50 = 125 ppm c.e.), these fish would not eat at exposure concentrations as low 25 ppm c.e. Lower dosages were not tested, but Tooby et al (1980) contend that it is reasonable to expect that decreased feeding rates will be seen at concentrations typically found in the environment. Furthermore, Gilderhaus (1967) note that adult bluegill sunfish were significantly smaller in ponds treated with diquat at 1.0 and 3.0 ppm c.e. than adult fish in untreated ponds. This decrease in size was correlated with both dosage and number of treatments (Table 22).

- **Effects on invertebrates**

Data on the field effects of diquat against aquatic invertebrates is not extensive. From the laboratory data it is apparent that the free-swimming (cladoerans) species are very sensitive to diquat. Field concentrations ranging around 1.0 ppm c.e. will affect several species of daphnia including *Daphnia pulex* and *Simocephalus vetulus*. One application to ponds in Wisconsin at 1.0 or 3.0 ppm does not affect the average and maximum number of *Daphnia pulex* during the summer season. However, there is a shift in the time of peak cladoeran abundance from mid-June till early-August or mid-October. This shift in peak abundance correlates well with a decrease in concentrations of diquat below that level which is typically toxic to *Daphnia pulex*. It takes three to six weeks for the environmental concentration of diquat to drop below concentrations of diquat typically seen to be toxic to 1st instar *Daphnia pulex* (LC50 = 0.16 ppm c.e.) (Table 5) (Gilderhaus, 1967). In a similar study, *Simocephalus vetulus* (Newbold, 1975) was eliminated for 64 days from a pond treated with 1.0 ppm diquat. It is interesting to note that the recovery of this population occurred in a similar time frame to the recovery seen with *Daphnia pulex*. Multiple treatments with 1.0 or 3.0 ppm caused significant decreases in both average and maximum numbers of cladoerans but the time shift to peak abundance usually remained the same as single treatments.

Other species of invertebrate have also been adversely impacted in the field when treatment with diquat for aquatic weed control was necessary. However, only *Hyalella azteca* appears to be acutely affected by diquat in the field. Concentrations as high as 2.5 ppm and as low as 0.11 ppm have been observed to cause significant reductions in the numbers of *Hyalella azteca* found in shallow water (Wilson, 1967 and Berry et al, 1975). In shallow water, there appeared to be "hotspots" with concentrations as high as 0.73 ppm for 12 hours after treatment. These concentrations are considerably higher than the LC50 of 0.048 ppm c.e. for this species. Therefore, the reduction in numbers of *Hyalella azteca* seen in shallow water at this site is not unexpected. However, in deep water the maximum diquat concentration never exceeded 0.03 ppm and *Hyalella azteca* was observed to be unaffected by these concentrations of diquat. In Lake Steilacoom, average concentrations of ~0.05 ppm c.e. were seen for the first 3 days after treatment with 0.13 ppm diquat for the control of *Elodea canadensis* and *Potamogeton* spp. This concentration is also high relative to the LC50 of *Hyalella azteca*. Therefore, adverse impact to this species is likely.

Both *Hyalella azteca* and four genera of aquatic snail were found by Hilsenhoff (1966) to decrease after treatment with diquat at 1.0 ppm. However, in this case, Hilsenhoff believed this was a secondary effect. Hilsenhoff attributed the decline in the numbers of this species to be due to a decrease in the amount of habitat available for colonization. This loss of habitat may be mitigated by the use of conservation webbing as described by Berry et al (1975).

Other species previously described in the section on diversity did not decrease in numbers due to the effects from diquat treatment. Increases in *Chaoborus* spp. numbers occurred due to an increase in dead and dying southern naiad species which provided additional foodstuff for this *Chaoborus* spp. Decreased oligochaete worm populations may have been due to their inability to compete effectively for food with the chaoborids (Tatum and Blackburn, 1962). A detailed description of the animal species present after treatment with diquat to control *Myriophyllum spicatum*, *Egeria densa* and *Elodea canadensis* has been discussed previously. No decreases in diversity were seen in these cases although the absolute numbers of an individual species or at a specific site were noted to increase or decrease as is typical for the various seasons (Wilson, 1967; Berry et al, 1975 and Hilsenhoff, 1966).

The short half-life exhibited by diquat (typically for 0.75 to 1 or 2 days) makes it unlikely that most species of aquatic invertebrates will be chronically impacted by diquat. However, for cladoerans, the shift in the time to peak cladoeran abundance or their absence for up to 4 months after treatment is indicative of a chronic reproductive effect. These shifts in foodstuff organisms have the potential to cause nutritional harm in species that depend on these invertebrates as their primary food source. If enough invertebrates of the right size class are not available when peak fish fry numbers occur, population of young of the year fish could be adversely impacted.

Dissolved oxygen content (DO) can effect the suitability of breeding habitat. Daniel (1972) found that treatment with diquat plus Cutrine® at concentrations of 3.0 plus 2.2 ppm, respectively, for the control of Eurasian water milfoil cause the dissolved oxygen concentration to drop to less than 1.0 ppm for more than 4 days. The DO recovered in about 2 weeks to 1.5 months. Recovery of DO occurred more rapidly in surface waters than in 30 to 90 cm deep water. Such conditions of anaerobiosis may be the primary cause of fish-kill or invertebrate-kill (Frank, 1972) or it may contribute to fish-kill by increasing the toxicity of the herbicidal treatment (Daniel, 1972). Similar decreases in DO were observed in the Soap Creek Ponds in Oregon and Lake Marion in South Carolina due to treatment with diquat. However, no mortality of invertebrate populations was attributed to these DO decreases (Inabinet, 1976, Wilson, 1967).

- **Interaction of water quality with Diquat products and their commercial recommended adjutants**

Water quality encompasses many parameters, but the toxicity of chemicals to fish is most often influenced by water hardness, pH and the inter-related factors of temperature and dissolved oxygen.

Hardness is the only factor that has been investigated extensively as a contributor to the toxicity of diquat. As discussed previously, diquat exhibits much higher toxicity to fish in soft water than in hard water. Even when water hardness is not indicated, high toxicity of diquat to fish (LC50 = <10.0 ppm c.e.) probably correlates with soft water conditions (<50 ppm $CaCO_3$) and low toxicity (LC50 = >30 ppm) probably correlates with hard water conditions (>150 ppm $CaCO_3$). Details on this phenomenon can be found in Table 18 with more precise information found in Table 8.

In Washington State, hard water/higher pH conditions are generally found in eastern Washington lakes relative to western Washington lakes. Some mitigating factors for the protection of the more susceptible stages and species may include using diquat in hard water bodies and selecting another herbicide with a less adverse impact in soft water to treat soft water bodies.

Because very little work has been done on the effects of pesticide combinations it is unclear whether other pesticides, applied for other purposes, would typically enhance the effects of diquat. However, Cutrine®-Plus and diquat have been noted to have a synergistic toxicity on a number of fish species including brown trout (Simonin and Skea, 1977 and Daniel, 1967). However, Cutrine®-Plus or Komeen® combined with diquat is not synergistic against apple snail (Winger et al 1984). Diquat and parathion act in a synergistic manner against the mosquito fish (Shearer and Halter, 1980) and diquat plus the fungicide nabam behave in a synergistic manner against embryos and tadpoles of *Rana pipiens*. Details of these synergistic effects may be found in section 4.2.4.

In the State of Washington, diquat is not allowed to be mixed with other products. However, for the control of floating or emergent plant species, the use of nonionic surfactants is highly recommended (Reward® LA label, 2000). Also, the use of thickening agents or invert-emulsions is recommended for applications of diquat to control submerged aquatic macrophytes. A number of surfactants are registered for use with water-soluble herbicides like diquat for drift control such as R-11, X77 and LI700. Thickeners or invert-emulsions are often used with liquid products applied by subsurface injection to allow the treatment to sink more deeply into the water column where it can be most effective.

There are a number of adjuvants registered for aquatic use in Washington State. Most nonionic surfactants should be mixed at 0.25 to 0.5% by weight of application solution when diquat is being applied to floating (surface) or emergent aquatic macrophytes. The toxicity of these adjuvants to bluegill sunfish, rainbow trout and daphnia has been well Doumented. None of these aquatic adjuvants should be toxic to fish or aquatic invertebrates when applied at labeled rates. However, it has been noted by Watkins et al (1985) that some aquatic adjuvants have a potential to be toxic to aquatic organisms when applied to shallow water. For example: 1) If Spar-Mate® is applied at the labeled use rates to water with a depth of less than 1.5 meters, it can be toxic to bluegill sunfish. 2) If Cide-Kick®, X-77®, Formula 403®, or IVOD® are applied at the labeled use rate to water with a depth of less than 0.1 meters, they may be toxic to fish. Since the depths given are for concentrations of the adjuvant that will kill 50% of the treated animals an additional safety factor of ~10-fold would need to be added to assure safety of the adjuvant to the biota. Details of the toxicity and depth considerations for a number of aquatically applied adjuvants can be found in Table 9. Although adjuvants are typically considered to be "nearly inert", they are not entirely inert. Adjuvants can enhance, diminish, or have no effect on the activity of herbicides. Although acute aquatic testing has been done on a number of adjuvants, insufficient data exists to appropriately evaluate risk from the toxic effects of adjuvants when mixed with herbicides and applied to the aquatic ecosystem. Lee and Furtado (1977) also recommend this safety factor approach relative to depth when controlling *Salvelina molesta.*

All formulations of diquat may not be similar in toxicity on a cation equivalence (c.e.) basis. We know that most commercial diquat formulations have similar toxicity to fish and invertebrates. However, only one of these products (Reward® LA) is labeled for use in public waterways. It is unclear as to why most formulations may only be used in water impounds. However, those formulations, which are not labeled for use in public waterways, should not be used in public waterways. If new formulations are registered for use in public waterways, caution should be practiced since all inert materials are not equivalent. The "inert materials" and contaminants may interact with the pesticide to give antagonistic, additive, cumulative or synergistic effects against target aquatic plants and algae, and non-target fish and aquatic invertebrates.

4.3.2.4 Effects on Endangered Species

There are a number of aquatic species that have been classified as sensitive, endangered or threatened. These include several species of salmon, 13 species of rockfish, 2 species of herring, 2 species of dace, and 8 species of amphibians. A list of these species can be found in Appendix 5. However, the salmonid species including Coho salmon (*Oncorhynchus kisutch*) and Chinook salmon (*Oncorhynchus tshawytscha*) are not acutely affected by diquat at concentrations that will typically be found in the environment. For example, Chinook and Coho salmon have LC50 values that range from 16 to 30 ppm c.e. and these values greatly exceed the most typical 4-day mean EEC (0.059 ppm c.e.). Therefore, these species are unlikely to be acutely impacted by typical field rates of diquat. However, since diquat can influence migration of Coho salmon at concentrations as low as 0.5 ppm c.e., it may be advisable when treating at rates approaching this concentration to avoid treating at times of year when salmon smolts are present (Lorz et al, 1979. There is also evidence that Coho salmon may not survive seawater challenge tests after exposure with 5.0 to 20 ppm c.e. diquat. It is noteworthy that this concentration of diquat approaches the highest field rate of 0.75 ppm c.e., which is another reason to avoid treating salmon habitat with diquat when smolts are present.

These results indicate that diquat used for control of aquatic weeds may put Coho salmon at risk during the smoltification process. Although very few species relevant to the northwest have been tested, Coho salmon and Chinook salmon smolts have been tested in a seawater challenge test with diquat; and the effects of diquat treatment on migration has also been tested in Coho salmon smolts.

1.2

4.3.2.5 Risk Analysis for Aquatic Species

Summary: Based on the acute toxicity determined in the laboratory, diquat is unlikely to adversely impact the fish biota. However, the invertebrate biota may be adversely impacted since the most sensitive species of invertebrate are ~10-fold more sensitive to diquat than the most sensitive species of fish. Available field data tends to support the conclusion that was reached with laboratory data. Secondary effects of diquat may impact the aquatic biota in either a positive or negative manner. Reduced dissolved oxygen content may affect both numbers and dominant species present. Increases in nutrient levels can lead to an increase in the number of phytoplankton and an improvement in the condition of species that feed on phytoplankton. Short-term decreases in the number of phytoplankton present have also been observed; this decrease in the phytoplankton count can lead to decreases in biomass (yield) of planktovoric amphibians and fish. A great deal of data relevant to the risk analysis was discussed in Sections 4.2.3.2.1 Acute Effects and 4.2.3.2.2 Chronic effects. However, no formal risk analysis was actually performed in those sections.

A number of fish and invertebrate species are known to be able to avoid environmentally relevant concentrations of diquat in the laboratory. For example, rainbow trout fry can avoid the effects of diquat at concentrations of 0.5 ppm by drifting out of the treatment zone due to a decrease in swimming speed (Dodson and Mayfield, 1979). Goldfish can actively avoid diquat at concentrations as low as 1.1 ppm c.e. (Berry, 1984). Mayflies (*Ephemerella walkeri*) may also avoid diquat concentrations as low as 1.0 ppm with a trend seen down to 0.1 ppm c.e. (Folmar, 1978). However, even though these species can distinguish between polluted and unpolluted waters, the investigators conducting the work believed it was unlikely that they would or could avoid water polluted by pesticides. It was also noted by Hesser et al (1972) that certain scrap fish (carp and suckers) could avoid diquat when applied in the field at concentrations as high as 26.7 ppm. Even though these fish could avoid these higher than normal concentrations in the field, such concentrations were not noted to be toxic in the field or the laboratory.

There has been only one field report on the avoidance of diquat. Hesser et al (1972) found that carp, suckers and other scrap fish could avoid diquat in drainage ditches at the treatment concentration of 26.7 ppm. The maximum treatment rate is not higher than 0.75 c.e. ppm so it may not be relevant to current practice. This report and reports on the ability of goldfish to actively avoid 1.1 ppm diquat in the laboratory and the ability of rainbow trout fry to passively avoid diquat at 0.5 ppm have been repeatedly cited as evidence that fish may avoid diquat treated areas and thus reduce the potential for incurring adverse effects. However, in a much more sophisticated research program using radio-tracking of largemouth bass in treated and control areas, Bain and Boltz (1992) found that “movement of radio-marked largemouth bass in Guntersville Reservoir (Alabama) were limited and largely confined to study sites. Differences among sites and the effect of reduced vegetation coverage had no measurable effect on movement of adult largemouth bass.” However, the minimal amount of vegetation in the diquat plus Komeen® treated sites and the control sites made interpretation of the effects of plant cover difficult.

Typical concentrations of diquat can be predicted from EPA’s worst case scenario application of 0.224 ppm c.e. and a half life of 0.75 days to average around 0.059 ppm c.e. during the 4-days of acute exposure. Modeling work indicates that when between 0.30 to 0.37 ppm c.e. is used as a spot treatment the 4-day geometric mean EEC will not be higher than 0.021 ppm c.e. in northwest reservoirs, northwest ponds or southeast canals (Ritter et al, 2000). It is important to note that the highest mean 3 day EEC in Lake Steilacoom was typically ~0.05 ppm c.e. which is reasonably close to the 0.021-0.059 ppm c.e. that was modeled by Ritter et al (2000) and determined independently by CSI, based on the field work of Fujie (1987).

Since these results are fairly consistent, the expected environmental concentrations (EEC) for diquat have been estimated to be 0.021 to 0.059 ppm c.e. for short-term acute effects and 0.0087 ppm c.e. for long-term (28 day) chronic effects. However, since the dissipation of diquat is so rapid, another approach is to assume that no chronic exposure is possible and all aquatic animals will be protected from the chronic effects of diquat.

Due to non-linear dissipation, the dissipation rate of diquat in the field is difficult to determine. Therefore, the EECs used in these risk assessments are based on linear dissipation models of expected environmental concentrations for short periods (4-days) and for longer periods (28 days). These time frames correspond to those considered to be acute and chronic exposures, respectively. However, field data indicate that the concentration found in the field are similar to those generated by the predictive models. For details of concentrations found in the field see sections 4.2.2, 4.2.2.1, 4.2.2.2, and 4.2.2.4.

- **Acute risk assessment**

  In general, risk assessment for protection of the biota takes into consideration two factors.

  1) The typical environmental concentrations of the pesticide that the biota will be exposed for short periods of time (1 to 4 days):

     For diquat, the most typical time-weighted average expected environmental concentration (EEC) that fish and most invertebrates will be exposed to for 1, 2, 3 and 4 days has been estimated by us to be 0.15, 0.102, 0.076 and 0.059 ppm c.e., respectively. These time-weighted EEC values assume an initial worst case concentration of 0.224 ppm and a half-life of 0.75 days. However, modeling work by Ritter et al (2000) assuming a spot treatment of 0.30 to 0.37 ppm c.e. indicates that time-weighted average EEC will not be higher than 0.077, 0.042 and 0.021 ppm c.e. for 1, 2 and 4 days, respectively, in northwest waters.

     The work of Fujie (1987) gives the daily concentrations detected after application of 1 gallon formulation/acre. These values were 0.020 to 0.095, 0.009 to 0.063, 0.013 to 0.047, and <0.004 to 0.027 ppm c.e. at 1, 2, 3 and 5 days, respectively. The predicted values based on a 1 gallon formulation/acre treatment applied to 4-feet of water yield daily concentration values of 0.073, 0.029, 0.011 and 0.002 ppm c.e., respectively, which is in good agreement with experimental results.

  2) The 96-hour toxicity (LC50) of the pesticide to the most sensitive environmentally relevant species is as follows:

     For diquat, the most sensitive environmentally relevant fish size class and species are sac-fry of striped bass (*Morone saxatilis*) with a 96-hour LC50s of 0.54 ppm c.e. Other species of fish with similar sensitivity to diquat include 8 to 10 day old walleye

> (*Stizotedion vitreu*) (96-hour L50 = 0.75 ppm c.e.), sac-fry of the smallmouth bass (*Micopterus dolmieu*) (96-hour LC50 = 1.3 ppm c.e.), and 9 to 13 day old largemouth bass (*Micropeterus salmoides*) (96 hour LC50 = 4.9 ppm c.e.). The only other time frame at which these species may suffer adverse impact is after 24-hours of exposure; and the 24 hour LC50's for striped bass sac-fry and smallmouth bass sac fry is 0.54 and 1.29 ppm c.e., respectively.
>
> The most sensitive size class and species of aquatic invertebrates is the amphipod *Hyalella azteca* with a 96-hour LC50s of 0.048 mg/L. Other species of invertebrates with similar sensitivity to diquat include 1st instar *Daphnia pulex* (48 hour LC50 = 0.162 ppm c.e.), adult *Daphnia pulex* (48-hour LC50 = 0.22 ppm c.e.), apple snail (*Pomacea pludosa*) (96-hour LC50 = 0.34 ppm c.e.), pocket shrimp (*Mysidopsis bahia*) (96 hour LC50 = 0.42 ppm c.e.) and 1st instar *Daphnia magna* (48-hour LC50 = 0.79 ppm c.e.). The other time frames at which some of these species may experience adverse impact include both a 24-hour and 48-hour exposure. These 24-hour LC50's were shown to be 0.62 ppm c.e. for *Hyalella azteca*, 0.81 ppm c.e. for *Daphnia pulex* larvae and 0.702 ppm c.e. for *Daphnia pulex* adults. The only other invertebrate species likely to be adversely impacted after 48-hour of exposure are *Hyalella azteca* and the pocket shrimp with 48-hour L50s of 0.22 and 0.87 ppm c.e., respectively.

Diquat does not appear to have significant acute toxicity to the most sensitive fish species tested (Table 18). The risk quotient for the most sensitive fish species tested is approximately at the level of concern (0.1) for protecting the biota (RQ = 0.11 = 0.059 ppm c.e./ 0.54 ppm c.e.) (Table 23). Under a worst case scenario diquat would be classified as a restricted use compound since the generated RQ for the most sensitive species lies between 0.1 and 0.5. However, since an assumption of accuracy on a risk quotient of even one significant digit is likely excessive, the fish biota can be judged to be sufficiently protected from the adverse impact of diquat when it is applied according to EPA's worst case scenario. However, Hughes (1968) recommends that diquat not be used to control aquatic weeds when striped bass are present. This recommendation may have added weight when the one-day time-weighted EEC (0.15 ppm c.e. and LC50 (0.54 ppm c.e.) are considered. Under this scenario the risk quotient for striped bass exceeds the level of concern of 0.10 for low risk but does not exceed the level of concern (0.5) for high risk; (RQ = 0.27 = 0.15 ppm c.e. /0.54 ppm c.e.. Therefore, during the first 24-hours of exposure, striped bass sac-fry or similarly sensitive species in fish biota may suffer adverse impact from exposure to diquat.

In US EPA evaluation of pesticides under FIFRA, an acute Risk Quotient of higher than 0.1 is interpreted as exceeding the Level of Concern, and leads to the conclusion that the risk may be unacceptable unless further analysis shows otherwise (Urban and Cook, 1985). The general practice in risk assessment is to estimate the Expected Environmental Concentration (EEC) based on the highest expected initial concentration and the most representative half-life. Therefore, diquat should be safe to fish when used at or below the maximum use rate in a pond that is at least 200 cm deep.

However, the more sensitive species of invertebrates in the biota may be at risk. This conclusion is apparent since the 4-day EEC and the LC50 for the amphipod *Hyalella azteca* are 0.059 ppm and 0.048 ppm c.e., respectively (Table 19). This amphipod is important to the food web where it is found (Wilson, 1967) and related species in Washington water lilies may also be of importance to the food web. However, since this is the only species of invertebrate tested that can be judged to be at high risk, it is likely that fish using this amphipod species for food will be able to find other species of a similar size class and nutrition value. For example, various copepods like *Eucyclops* spp., *Diapotomus,* and the scud-amphipod *Gammarus fasciatus* are fairly tolerant of diquat with typical LC50s ranging from 5.4 to 25 ppm c.e. Since the Risk Quotient for *Hyalella azteca* is above the level of concern (0.10), benthic organisms within the biota may be adversely affected; RQ = EEC/LC50 = 1.2 = 0.059 ppm c.e./0.048 ppm c.e.). Since the presence of sediment decreases the toxicity of diquat (LC50 = 3.4 to 6.8 ppm c.e), when *Hyalella azteca* is associated with sediment, it may be protected from the affects of diquat (RQ = 0.017 = 0.059 ppm c.e./3.4 ppm c.e.). Although *Hyalella azteca* may be protected from the effects of diquat by the presence of sediment, other sensitive species may be adversely impacted at levels that yield short-term risk quotients between the level of concern for low and high risk (0.1 to 0.5); RQ = 0.37 = 0.059 ppm c.e./0.16 ppm for *Daphnia pulex* larvae; RQ = 0.27 = 0.059 ppm c.e./0.216 ppm c.e. for *Daphnia pulex* adults; RQ = 0.17 = 0.059 ppm c.e./0.34 ppm, for apple snail; RQ = 0.14 = 0.059 ppm c.e./0.42 ppm c.e for pocket shrimp. With the levels of RQ (0.14 to 1.2) exceeding the level of concern of 0.1, invertebrate species will probably experience adverse impact from exposure to diquat at concentrations that reflect EPA's worst case scenario of 0.224 ppm when the 4-day geometric mean EEC is 0.059 ppm. Spot treatment with 0.3 to 0.37 ppm, which allows for a modeled 4-day EEC of 0.021 ppm c.e., still creates an environment where the risk quotient will exceed the low risk level (0.1) of concern but will be less than the high risk level of concern (0.5); RQ = 0.44 = 0.021 ppm c.e./ 0.048 ppm c.e. for *Hyalella azteca*; RQ = 0.13 = 0.021 ppm c.e./0.16 ppm c.e. for *Daphnia pulex*); and RQ = 0.06 = 0.021 ppm c.e./ 0.34 ppm c.e for the apple snail. Therefore, at higher treatment concentrations applied to an entire water body, the invertebrate biota will be at high risk, which is unlikely to be mitigated by classification of diquat as a restricted use compound. If the more typical spot treatment scenario is used, the invertebrate biota will still be at risk; but mitigation will be possible by classifying diquat as a restricted use compound.

In determining these acute risk quotients for aquatic invertebrates, four-day time weighed average EECs were used in the calculation even if the standard LC50 test time was only two days. Historical data has shown that the 48- and 96-hour LC50s will be similar for these species, so acute risk quotients, assuming an exposure of 4 days, are typically conducted with 48-hour LC50 data for these species (primarily cladoerans). It is interesting to note that those species at risk after 4 days of acute exposure were usually also at risk after oneor two days of acute exposure providing that acute tests were conducted in those time frames (Tables 19 and 23).

For both treatments of entire water bodies and spot treatments, field studies in Washington support the laboratory risk assessment since the three-day mean EEC = ~0.05 ppm at Lake Steilacoom and <0.02 ppm at Gravelly Lake (Serdar, 1997). Field verification with specific organisms has also been achieved. *Hyalella azteca,* several snail species, and various cladoerans are adversely impacted in water bodies that have been treated with from 0.11 to 1.0 ppm diquat (Berry, 1975 Hilsenhoff, 1966 and Wilson, 1967). Other species of invertebrates do not appear to be adversely impacted in the field. For a discussion of field results for both fish and invertebrates, see section 4.3.2.3.

- **Chronic risk assessment**

  In general, chronic risk assessment for protection of the biota takes into consideration two factors.

  The typical environmental concentration of the pesticide that the biota will be exposed to for “chronic” periods of time is determined as follows: chronic exposure is typically considered to be 21 to 28 days. However, what is considered chronic exposure for a species depends in part on the length of its life cycle. For example, a chronic exposure for *Ceriodaphnia dubia* may be considered to be four to seven days since this organism is able to complete its full life cycle within this time frame. However, chronic exposure for *Daphnia magna* is generally considered to be 21 to 28 days since it takes this much time for *Daphnia magna* to go from a neonate to the completion of its reproductive period. Fish on the other hand, can take 30 to 90 days to go through the most sensitive portion of their life cycle, which is generally considered to be from egg to free-swimming fry.

  For diquat, the typical environmental concentration that fish and most invertebrates will be chronically exposed to is fairly low. Therefore, the 28-day geometric mean EEC = 0.0087 ppm c.e. assuming a maximum use rate application to a 200 cm deep pond and a dissipation half-life for diquat of 0.75 days. Even the most susceptible species of fish will not be adversely impacted by chronic 28-day exposure to diquat since the long-term level of concern is less than 1.0 as demonstrated by the chronic risk quotient; RQ = <1.0 = EEC/MATC = 0.51 = 0.0087 ppm c.e/0.017 ppm c.e. for killifish. This risk quotient, however, may not have great validity since the end-points measured are not lower than the chronic LC50 for this species, which is also not ecologically relevant. However, the only well run chronic test was conducted with the fathead minnow which yielded an MATC of 0.2 ppm c.e. Since this value is much greater than the chronic EEC, it allows one to conclude that fish are not at risk due to chronic exposure to diquat. If an estimate is made for the MATC of the most sensitive species based on the acute to chronic ratio (38) for fathead minnow, the MATC for bass would be 0.014 ppm c.e. (0.54 ppm c.e./38). Since the MATC is significantly higher than the 28-day EEC, one can conclude that fish will not be adversely impacted by diquat when it is applied at the maximum use rate to a 200 cm deep pond; RQ = <1.0 = 0.062 = EEC/MATC = 0.0087 ppm c.e./0.014 ppm c.e.). This conclusion can generally be assumed valid since the chronic tests on which it is based are longer than 28 days

  Chronic risk to diquat may be an issue with invertebrate species. The chronic (21-day) MATC (0.045 ppm) indicates that *Daphnia magna* will not be impacted when the predicted 21- or 28-day long-term EEC is 0.012 or 0.0087 ppm c.e., respectively. This lack of chronic impact is predicted because the chronic risk quotient is lower than the level of concern (1.0); RQ = EEC/MATC = 0.27 = 0.012 ppm c.e./0.045 ppm c.e. for 21 days; or RQ = 0.19 = 0.0087 ppm c.e./0.045 ppm c.e. for 28 days. However, the predicted MATC for *Hyalella azteca* is based on the LC50 divided by the acute to chronic ratio of 19 for *Daphnia magna*. Therefore, the predicted MATC for *Hyalella azteca* will be 0.0025 ppm c.e. (0.048 ppm c.e./19). With a predicted MATC that is lower than the chronic EEC, the long-term risk quotient exceeds the level of concern (1.0); RQ = EEC/MATC = 3.48 = 0.0087 ppm c.e./0.0025 ppm. Therefore, it is likely that chronic adverse impact will occur to the invertebrate biota. Only one other species (*Daphnia pulex*) also yields a predicted chronic MATCs that are below the chronic EEC and when this occurs, it can be expected that the chronic risk quotient will be above the level of concern of 1.0; RQ = 1.42 =0.012 ppm c.e./0.0084 ppm c.e. for 21 days; RQ =1.03 = 0.0087 ppm c.e./0.0084 ppm c.e. for 28 days.

Other species like apple snail (predicted MATC = 0.017 ppm c.e.) and pocket shrimp (predicted MATC = 0.022) do not appear to be at chronic risk to diquat.

These data are confirmed by results from field studies that indicate that populations of *Hyalella azteca* and the cladoerans *Simochephalus vetulus* and *Daphnia pulex* may be disrupted for 1 to 4 months after treatment with diquat at concentrations that range from 1.0 to 2.5 ppm. Although the higher dosages are not likely to be encountered in the field, 1.0 ppm c.e. approximates the maximum field application of 0.75 ppm c.e. and therefore has ecological relevance. It is therefore likely that *Hyalella azteca* and the cladoerans will be adversely impacted when exposed to diquat at rates that mimic EPA's worst case scenario. Depression of critical foodstuff organisms for more than a month has the potential to have long-term impact on juvenile fish that depend on them as a food source. However, it is likely that these species are not the only invertebrates that are of the proper size class and nutritional quality to satisfy the needs of newly hatched fry (Wilson, 1967). For example, *Gammarus fasciatus* (LC50 = 18.7 ppm), and the copepods and ostracods (LC50 = 5.4 to 25) are of a proper size to also serve as foodstuffs for fry and fingerling fish. Gilderhaus (1967) found that fingerling fish appeared to be unaffected by changes in cladoeran peak population times that had been altered by the use of diquat to control American waterweed.

Summaries of the Risk Assessments used in this section are provided in Table 23.

4.3.3 Diquat Potential Impacts to Terrestrial Wildlife and Plants

The goal of this portion of the document is to discuss the effects of single applications/exposures and chronic applications/exposures to terrestrial wildlife (birds and mammals) and terrestrial plants exposed to aquatic herbicides containing diquat (i.e. Reward®LA). In addition, possible effects on the food chain and threatened and endangered species will be discussed as well as ways to mitigate exposure of these organisms to the aquatic uses of diquat. The information presented summarizes toxicological studies to determine the effects of diquat containing products on plant and animal species. Much of this information has already been discussed in sections 4.2.2.4 and 4.2.5.

4.3.3.1 Effects on Amphibians

Acute data for diquat are not available. However, chronic data and field data is available for several species of amphibians (Table 20). For the leopard frog (*Rana pipiens*) and the African clawed toad (*Xenopus laevis*), the MATC for development is 1.7 and 0.64 ppm c.e., respectively (Dial and Dial, 1987, Anderson and Prahlad, 1976). While the chronic LC50 for leopard frog was >5.4 ppm c.e., the chronic LC50 for African clawed toad was ~0.41 ppm c.e.

Diqaut at field applied concentrations of 1.0 ppm did not appear to have long term adverse impacts to the frog (*Rana temporia*) or the toad (*Bufo bufo*). Although reduction in growth rate was seen with these two species for a few days after application, they had recovered their weight by 18 days after treatment and by 32 days after treatment both species from treated areas weighed more than their control cohorts. This recovery in weight was believed to be due to an algal bloom of *Mougeotia* spp., which the tadpoles ate in great quantity.

The predatory smooth newt (*Triturus vulgaris*) bred in all treated ponds in the two years following treatment. Significantly fewer newt larvae were caught in treated ponds than control ponds in the year following treatment. However, it could not be determined if this was due to the direct effects of diquat or some other undermined environmental factors.

4.3.3.2 Effects on Terrestrial Animals (Birds, Mammals and Insects)

Studies have been conducted to assess the toxicity of technical grade diquat and the diquat containing product Reward®LA on various animal groups. Acute oral (LD50), acute dietary (LC50) and chronic dietary studies are presented below.

4.3.3.2.1 Acute Effects on Birds

Acute oral data are available for the mallard duck (Table 25). The acute oral LD50 for mallard ducks ranges from 60.6 ppm to 31 ppm (expressed as cation equivalents). These data indicate that diquat is moderately toxic to the mallard duck when orally dosed.

Acute dietary (LC50) data is available for diquat for several different species of birds. The lowest LC50 for diquat (expressed as cation equivalents) is 264 ppm for Japanese quail and the highest LC50 is listed as 575 ppm for bobwhite quail. These data indicate that diquat is moderately toxic to birds when consumed in the diet.

4.3.3.2.2 Chronic Effects on Birds

One generation reproduction (in feed) studies were performed on both bobwhite quail and mallard duck. The no observable effect level (NOEL) for diquat in bobwhite quail is >19.6 ppm c.e. (no reproductive impairment at the highest dose level tested). The NOEL for mallard ducks ppm (expressed as cation equivalents) was 5 ppm c.e. (based on reduction in numbers of eggs laid, hatchlings and 14-day old survivors seen at 25 ppm dose level).

4.3.3.3 Acute Effects on Mammals

Acute oral data is available for more than one mammalian species for diquat dibromide and LD50 values range from 120 mg/kg in rats to 233 mg/kg in mice (Table 26). This data indicates that diquat is moderately toxic. However, the reported LD50 for cows is listed as 30 to 56 mg/kg which indicates that diquat is highly toxic to cows.

4.3.3.3.1 Subchronic and Chronic effects on Mammals

A 20-day repeated dose dermal toxicity study with rabbits was conducted using technical diquat dibromide. The no observable effect level (NOEL) was 20 mg cation diquat/kg/day based on weight loss, unsteadiness, muscular weakness and inability to stand at the next highest dose level (EPA 1995 RED). A three-week repeated dose rat dermal toxicity study was also conducted using technical diquat dibromide (EPA 1995 RED). The NOEL for this study was 5 mg diquat cation/kg/day based on a number of symptoms seen at the next highest dose level. In a repeated dose rat inhalation toxicity study (EPA 1995 RED) using technical diquat the NOEL was found to be < 0.49 ug c.e./L . In a second repeated dose rat inhalation study using technical diquat (EPA 1995 RED) the NOEL was found to be 0.10 ug c.e./L.

In a rat multigenerational study (Hodge, 1990) the NOEL for toxicity was 16 ppm c.e. and the NOEL for reproductive effects was 240 ppm based on decreased body weight gains and other signs of toxicity at the next highest dose level. In a chronic (104-week) rat feeding study the systemic NOEL was 0.58 mg/kg/day for males and 0.72 mg/kg/day for females [expressed as cation equivalents (RED)] based on extralenticular lesions at the next highest dose level (cataracts). A one-year dog study feeding study (Hopkins, 1990) had a 0.5 mg c.e. diquat/kg/day based on unilateral focal lenticular opacity (cataracts) at the next highest dose level (12.5 mg c.e. diquat/kg/day).

4.3.3.4 Mitigation of Effects on Birds and Mammals

- **Mitigation measures specific to diquat products**

  There are two common routes of exposure for livestock and terrestrial wildlife to aquatic applications of diquat products. The two routes are exposure through drinking water treated with products containing diquat or eating aquatic plants, fish or other aquatic organisms from the treatment site. Based on the acute and chronic studies listed above, diquat and its products, that are used as aquatic herbicides, do not pose a significant acute or chronic risk to wild birds or terrestrial mammals. However, in order to mitigate possible problems with the watering of livestock, the Reward® LA label restricts the use of treated water for watering livestock for one day following treatment. Many studies have been run on diquat products to ensure their safety to wildlife and the label directions and warnings reflect the results of these studies. Therefore, if the chemicals are applied according to the label the effect on terrestrial wildlife should be minimal.

- **General mitigation measures**

  Although diquat products used as aquatic herbicides do not pose a significant risk to terrestrial wildlife, the following measures should be considered prior to all aquatic herbicide applications. One possible mitigation measure would be not allowing applications if large populations of birds use shorelines or islands in the water body to be treated until after nesting is complete. Another mitigation measure would be to time applications to avoid migratory waterfowl and other bird species that use certain water bodies during migration. Efforts to avoid effects on migratory and nesting birds would best be coordinated between the permit writer and the Washington State Department of Fish and Wildlife (WDFW) prior to granting the permit.

4.3.3.5 Possible Effects on the Food Chain

The potential of diquat to bioaccumulate in birds and mammals has not been well studied. However, it is unlikely that bioaccumulation will occur due to diquat's low Kow and its rapid binding to soil/sediment. Adverse effects on the food chain are also unlikely because of the relatively high LC50 and LD50 values reported and the low no observable effect levels seen in chronic studies with birds and mammals.

4.3.3.6 Effects on Endangered Terrestrial Plants, Birds and Mammals

A list of endangered terrestrial plants, birds and mammals is located in Table 27. Minimal effects to these organisms are expected from application of aquatic herbicides containing diquat. It should also be noted that Reward® LA appears to be of similar toxicity to terrestrial organisms.

Other mitigation measures involve the contact of WDFW by the issuer of the permit to ascertain if any endangered species may be affected by the application of the chemical to the water body in question. Questions asked by the permit granter would ascertain if any resident endangered bird or animal species are known to use the water body in question or its shorelines or islands as breeding or forage areas, or if the application coincides with the migration of any endangered species. If endangered species are present, mitigation measures may involve postponing application until after the breeding season or postponement of application until after migration of the species in question. If the risk is determined to be too great to the species in question, use of an alternate means of control (i.e. mechanical) may also be an option.

4.3.3.7 Effects on Terrestrial Plants

- **Acute effects of diquat on terrestrial plants**

  Diquat dibromide is a nonselective, quick-acting herbicide and plant growth regulator, causing injury only to the parts of the plant to which it is applied (Extoxnet). Diquat dibromide is also used to desiccate potato vines and seed crops, to control flowering of sugarcane, and for industrial and aquatic weed control. Adverse effects to terrestrial plants depend on a wide variety of factors including application rate and number of applications, soil absorption and the susceptibility of the species in question.

  As discussed in section 4.2.5, diquat is toxic to many non-target broadleaf plants, including both crops and native vegetation. However, if the water use restrictions on the label are followed, minimal damage to non-target terrestrial plants should occur.

- **Chronic effects of diquat on terrestrial plants**

  As noted in section 4.2.5, many plants can survive repeated exposure to diquat if key periods where they are more susceptible are avoided. However, due to the use pattern for diquat products in Washington State (typically no more than two applications per water body per year) chronic effects on terrestrial plants are not expected.

- **Mitigation of the effects on terrestrial plants**

  While diquat products are labeled for use on potatoes, hops, seed alfalfa and cotton, many terrestrial plants are susceptible to the effects of diquat. The main routes of exposure for terrestrial plants from aquatic herbicides are over spray/drift and the use of treated water as irrigation. In order to mitigate problems with agriculture, all labels for aquatic herbicides containing diquat have wording prohibiting the use of Reward® LA treated water for irrigation for three days on ornamental crops and 5 days on agricultural crops. In addition, the label specifically states that contact or drift (over spray) to other plants or crops should be avoided as injury to the plants may occur. If these label directions are followed, adverse effects to terrestrial plants following application of diquat containing aquatic herbicides will be minimal.

4.3.3.7.1 Acute Effects on Honey Bees

An acute five-day contact and oral toxicity honey bee study was performed by Gough et al (1987). They found that the contact LD50 of Reglone® (2.0 lb/gal diquat cation) fell from 337 ug/bee on day one to 60 ug/bee on day five. The acute oral LD50 fell from 81 ug/bee on day one to 13 ug/bee on day five. This paper reports that the lowest LD50 values reported fall within the International Commission for Bee Botany rating of slightly toxic. The 1995 EPA RED indicates that the diquat LD50 to honey bees is 100 ug/bee and therefore, diquat is practically non-toxic to bees. Therefore, diquat in either its recommended terrestrial or aquatic uses should present little hazard to honey bees.

4.4 ADDITIONAL POTENTIAL DIRECT AND INDIRECT IMPACTS OF HERBICIDE USE ON WET LAND ENVIRONMENTS

Summary: The presence of diquat products at concentrations effective against weeds in wetland environments may adversely affect these environments. Dilution should mitigate the effects of diquat so that it does not affect aquatic plants or non-target animals in marsh, bank and estuarine areas. The presence of diquat in the lotic environment, due to outflow from a lake or pond, may cause the destruction of aquatic plants that are favorable to the production of habitat for sunfish, minnows and bass.

The estuarine environment may be affected by the use of diquat. ***.*** *The more susceptible species of invertebrates are the pocket shrimp (*Mysidopsis bahia*) and the euryhaline amphipod (*Hyalella azteca*).* Hyalella azteca *is the most susceptible species tested and may be impacted adversely by diquat at concentrations typically found in the field.*

Failure to control emersed, floating, marginal and bank weeds can cause the native vegetation to be crowded out producing dense monoculture stands of noxious and invasive weeds leading to the degradation of natural habitats and an economic burden for residents who must keep water flowing or navigable.

Most crop, ornamental, and pasture land plants should be able to withstand concentrations of diquat in irrigation water that greatly exceed the maximum concentration typically used to control aquatic weeds. If the water use restrictions noted on the label are followed or if the MCLG (0.02 ppm c.e.) is not exceeded, damage from irrigation water treated with diquat should not be a substantial issue. Furthermore, residues that exceed the tolerances set by the EPA/FDA should not be exceeded. However, multiple irrigation of crops, ornamentals or pastureland may have adverse impact. The toxicity of diquat to various aquatic and terrestrial plants is noted in Tables 11 and 13.

Due to the manner in which diquat products are applied, significant impact to other wetland environments is unlikely. There may be tendency for drift into other wetland environments or a flow of water into estuarine, palustrine, riparian, lentic or lotic environments. However, due to dilution effects as treated ponds, lakes, and canals normally flow into streams and rivers and ultimately into estuaries, it is not anticipated that the impact would be measurable.

4.4.1 Estuarine (Intertidal) Environments

Water from a stream or river containing diquat may flow into an estuary. Dilution effects from the water already present in the estuary and diurnal tides should dilute diquat to levels where it is not significant in the water column. However, since some estuaries contain considerable amounts of sediment and diquat binds very tightly to sediment, diquat has the potential to build up in sediment where it may be stable for a long period of time. It has been demonstrated that diquat in sediment at concentrations up to 250 ppm c.e. usually is not released from sediment, but that higher concentrations of diquat may be released from sediment depending on sediment characteristics (Hiltibran et al, 1972). Diquat in aerobic or anaerobic conditions does not degrade readily (EPA RED, 1995). There has not been an adverse impact from diquat due to its release from sediment in over 30 years of its use for the control of aquatic weeds (Dyson and Takacs, 2000). However, Birmingham and Colman (1983) believe that only about 7% of the maximum binding capacity of sediment can be utilized before release of diquat from the sediment may occur. They estimated that under conditions of total binding to the sediment and no degradation it would take 10 to 50 years for diquat to accumulate to a level where a release would occur. However, since only a portion of diquat is bound to the sediment and slow degradation does occur, modeling data indicate that typical sediment diquat concentrations will not exceed 25 ppm c.e. With this hypothesis, it is estimated that it would take hundreds of years of yearly treatment before any potential of exceeding the binding capacity of the sediment is likely to occur (Dyson an Takacs, 2000).

A number of species of bacteria have been noted to degrade diquat including *Pseudomonas fluorescens*, *Rhizobium* spp., and *Aerobacter aerogenes* (Calderbank, 1968 cited by Simsiman et al, 1976). In addition, the fungus *Psedoaegerita matsushimae* has been noted to reduce the toxicity of foliage in diquat-treated media. It is apparent that these species can degrade diquat into at least three water-soluble metabolites but the identification of these metabolites has not been determined. Furthermore, there are indications that when diquat is adsorbed to kaolinite clay, it is more susceptible to degradation than diquat adsorbed to montmorillonite clay (Weber and Coble, 1968 in Simsiman et al, 1976). Although diquat is degraded on dead and dying plant surfaces by the resident microbes, it is not understood to what degree microbial degradation of diquat occurs after adsorption to the sediment. However, most authors believe that sediment-bound diquat is inactivated and not biologically available to microbes for degradation.

For diquat, the estuarine/marine/euryhaline organisms had LC50 or EC50s values that ranged from 0.048 ppm c.e. for the amphipod *Hyalella azteca* to 54 - 141 ppm c.e. for the Eastern oyster [(*Crassostrea virginica*) Table 18 and 19)]. Species that may be found in estuaries include the *Hyalella azteca* (96-hour LC50 = 0.048 ppm c.e.), *Mysidopsis bahia* (96-hour LC50 = 0.42 ppm c.e.), pink shrimp (*Paneus durarum* with a 48-hour LC50 = 8.48 ppm c.e.), white shrimp (*Paneus setriferus* with a 48-hour LC50 = > 1.0 ppm), and the sheepshead minnow (96-hour LC50 = 48 ppm c.e.). Other species of fish, which are found in either fresh or saltwater may, at times, be found in estuary environments. These species include the American eel (96-hour LC50:22 ppm c.e.), coho salmon (96-hour LC50 = 20.5 to 30 ppm c.e.), Chinook salmon (48-hour LC50 = 16 ppm c.e.) and striped bass (96-hour LC50 = 0.54, 5.4 and 43 ppm c.e. for sac-fry, fingerlings and subadults, respectively). Estuarine shrimp and other small estuarine species are often tested for toxicity because of their importance in the aquatic food web, great abundance and sensitivity to pollution and pesticides.

Some estuarine/marine species are more susceptible than the freshwater species. Because of its high susceptibility to diquat relative to potential environmental concentrations, *Hyalella azteca* is classified as an organism at relatively high risk from the effects of diquat (RQ = 1.2). *Hyalella azteca*, which may be found in both freshwater and estuarine environments, is the most sensitive species tested. The pocket shrimp is also a very sensitive species (RQ = 0.14); it is important that pesticides destined for use in and near estuaries be tested to determine their toxicity to crustaceans and the capacity of crustaceans to avoid them. This is particularly important since some species of estuarine shrimp are important in the food web and abundant in the ecosystem. However, to date, no avoidance tests with crustaceans have been conducted with diquat.

Even with extensive dilution, the more sensitive species may be adversely affected if they inhabit an estuarine environment where the sediment is capable of accumulating large amounts of diquat. Although the presence of sediment decreases the toxicity of diquat to *Hyalella azteca* from a 96-hour LC50 of 0.048 ppm c.e. in water only to 6.8 ppm c.e. in the presence of sediment, it is probably insufficient to protect this species from field impacts of diquat applied at concentrations ranging from 0.1 to 2.5 ppm c.e. (Wilson, 1967; Wilson and Bond, 1969; Hilsenhoff, 1966; and

Berry et al, 1975). All of the field studies conducted with *Hyalella azteca* have been conducted in freshwater and no estuarine field studies were conducted.

4.4.2 Palustrine (Marshy) Environments

Extensive growth of rooted aquatic macrophytes such as cattails may effectively dam a marsh and increase the depth of the palustrine system by several-fold. In this way, aquatic macrophytes assist in spreading waters onto the surrounding land to increase its fertility and provide additional areas for fish and amphibians to feed and spawn (Goldman & Horne, 1983). Even without flooding, these plants may have an effect on habitat suitability for wild birds, mammals and other terrestrial organisms.

The dominant plants found in palustrine environments are emersed (emerged). While most emersed species are not significantly affected by diquat, cattail and water pennywort can be severely affected. Other species that may be affected include water paspalum (*Pasplum fluitans*), water primrose (*Ludwigia uruguayensis*), watershield (*Brasenia schreberi*) and waterwillow (*Justica americana*) (Westerdahl and Getsinger, 1988). Most emersed plants are not likely to be adversely impacted at the concentrations of diquat used to control submersed aquatic weeds. However, floating and rooted submersed plants typically found in a palustrine environment may be impacted by treated water that enters these areas from lakes and ponds. For example, treated water that entered Chambers Creek from Lake Steilacoom contained concentrations of diquat that were as high or higher than any point measured within the lake even though a 400 foot buffer zone was provided between the point of application and the outlet to Chambers Creek. If these rooted macrophytes are destroyed, potential habitat will be lost to fish and amphibians. Also, loss of suitable habitat for wild birds and mammals may occur.

4.4.3 Riparian (Margin and Bank) Environments

Diquat products may be used to treat the submerged margins of lakes and ponds to eliminate weeds and algae that interfere with recreational use. Diquat is used to control grass and broadleaf weeds along rights-of-way, ditch banks, and floating, emersed, marginal and bank weeds of ponds and lakes.

The application rates of these herbicides have the potential to impact any non-target grass and broadleaf species that they come in direct contact with. Typically, the rate of application as a spot spray to non-aquatic weeds is to apply 1 to 2 quarts of Reward® LA in 100 gallons of water. If the treatment is to be applied as a broadcast treatment, 1 to 2 pints of Reward® LA should be applied in at least 125 gallons of water/acre. A nonionic surfactant like X-77 should be added to the spray mixture at the rates recommended on the label. The Reward® LA label does not specify which species of non-aquatic weeds to control, but does specify that for best results the target weeds should be young, actively growing grass or other undesirable growth. Difficult weeds (such as perennial or deeply rooted weeds) can often be controlled by tank mixing Reward® LA with other systemic type herbicides. Diquat has particular utility in controlling cattails and pennywort and may be effective against a variety of other emergent species which proliferate in wetland habitats, including wet meadows, pasture wetlands, cattail marshes, stream and river banks, lake shores, irrigation ditches, drainage ditches and storm water retention basins. Non-native riparian weeds may grow in dense monoculture stands that provide poor habitat for native wildlife. In producing thick stands, these weeds may choke out waterways, slowing flow and promoting siltation. Invasion of these noxious weeds results in degradation of natural habitats for native vegetation and wildlife as well as causing economic burden on farmers who must keep irrigation water flowing.

Diquat, under a wide range of environmental conditions, is used primarily along rights-of-way, railroad beds/yards, highways, roads, dividers and medians, parking lots, pipelines, pumping stations, public utility lines, transformer stations and substations, electric utilities, storage yards and other noncrop areas. Reward ® LA may also be used to control broadleaf and grassy weeds in landscape beds, lawns, golf courses and roadsides as well as on the edges of non-flooded ponds, lakes and ditches. As discussed before, Reward® LA may be applied directly to the water of ponds, lakes, reservoirs, marshes, bayous, drainage ditches, canals, streams, rivers and other quiescent bodies of water for control of aquatic weeds at rates ranging from 0.5 to 2 gallons formulation/acre, which can lead to typical concentrations of ~0.1 to 0.75 ppm c.e. (depending on rate of application and depth of water). In water that is less than two feet deep, Reward® LA should not be applied at rates of more than one-gallon formulation/acre. In the state of Washington, diquat has been used in the recent past at Lake Steilacoom and Gravelly Lake for the control of *Elodea canadensis* and various *Potamogeton* spp. However, Reward® LA is not labeled for control of *Potamogeton richardsonii* and may not be effective against *Potamogeton robbinsii* except in the spring and early summer when the plants are still in an early stage of growth. Other species of aquatic weeds, which may be controlled by diquat are discussed in Section 1.0; a few of these are coontail (*Ceratophyllum demersum*), watermilfoil (*Myriophyllum* spp.), *Hydrilla verticillata*, *Salvinia* spp., Duckweed (*Lemna* spp.) and cattails (*Typha* spp.). For the latest information on the use of diquat products, the current labels and permits must be consulted.

Diquat products (Syngenta, formerly Zeneca® Diquat) may also be used for other plant control situations including applications for preharvest desiccation of seed alfalfa, seed clover, seed sorghum, seed soybeans and potatoes. It may also be used as a directed spray for the control of weeds in crop sites that are being prepared for planting with tree, vine, small fruit and vegetable crops. Furthermore, diquat may be used for control of weeds where nonbearing trees, and vines are grown. Treated sites should not be used for food or feed for 1-year after application. For the control of weeds, diquat acts as an inhibitor of photosynthesis. At rates typically used for the control of weeds, diquat is not generally considered to be a systemic herbicide, although at higher rates some systemic action may be seen.

Non-target plants and animals have the potential to be impacted by diquat products as described in Sections 4.2.3.1, 4.2.3.2, 4.3.3.1, 4.3.3.2 and 4.3.3.4. Please review these sections for information on diquat's acute (Table 2, 14, 15, 16, 17 18, 19, 23, 24, 25 and 26, Appendix 1 and Appendix 2) and chronic (Table 2, 20 and 21, Appendix 3 and Appendix 4) effects in non-target aquatic plants and animals.

4.4.4 Other Wetland Environments

Pasture, which is completely or partially flooded, may be impacted by diquat-treated waters that caused the flooding. Although, many species of terrestrial grasses and broadleaf weeds can tolerate exposure to diquat for one time at concentrations that exceed the rates typically found in treated water (Table 10), multiple irrigations or floodings may have adverse impact on terrestrial plants (Ecology, 1992). Many sensitive plants are not affected by irrigation, agricultural spray or flood waters that have their origin in treated water bodies**.** Section 4.2.5 (Table 10) deals with the effects of irrigation water, agricultural sprays and floodwater. Most of the tested crop plants are not effected by water containing normal concentrations (up to at least 0.45 ppm c.e.) of diquat.

These irrigation events can occur up to three times during the season without having unacceptable residue in the crop or adversely impacting growth (Gangstad, 1986). However, more susceptible crop species may be affected if more than one irrigation event occurs with treated water. If the diquat concentrations are less than the MCLG (0.02 ppm c.e.), adverse impact should not occur on irrigated crops. The crops that were investigated included corn, wheat, carrots, lettuce, potatoes, sorghum, soybeans and onions. Not all crops were irrigated with the highest rates of diquat containing water but were irrigated at concentrations believed to be the maximum that would typically be encountered in irrigation water (Gangstad, 1986; White, 1994). Phytotoxicity damage is unlikely in crops irrigated at 0.5 ppm diquat (Calderbank, 1972 in White, 1994).

4.4.4.1 Lentic Environment

Potential impacts on lentic and lotic environments as to the chemical ecology were discussed in Section 4.2.3.1. Effects on the biota in these environments were discussed in Section 4.3.

4.4.4.2 Lotic Environment

The lotic environment can be influenced by the presence of diquat in water from a lake or pond outlet. If diquat is present at levels that control weeds and the outlet gate is closed, a type of habitat favorable to sunfish and amphibians will develop. If the outlet gate is open, another type of habitat more favorable to salmonids may develop. Also, if protracted (seasonal) contact with water containing diquat at concentrations of as low as 0.11 to 0.13 ppm c.e. occurs, *Elodea canadensis* and *Potamogeton* spp. may be controlled (Serdar, 1997). Diquat has been used to control these species in Lake Steilacoom and Gravelly Lake during the 1996 season. During treatment season, concentrations high enough to control these species were seen at the outlet from Lake Steilacoom to Chambers Creek.

- **Closed outlet gate or absence of diquat**

  If the outlet gate from a pond or lake to a river or stream remains closed, dense growths of rooted aquatic macrophytes may effectively dam rivers and streams and increase the depth of the lotic system by several-fold. Similar effects may occur if the lake or pond is not treated with diquat.

- **Open outlet gate in presence of diquat**

  If water containing diquat at effective concentrations passes through the outlet gate of a lake or pond into a river or stream, the rooted aquatic macrophytes may be destroyed. This can have a substantial impact during the next freshet or high water event. Normal spring high water flows, in absence of rooted aquatic macrophytes, can dig up and kill large numbers of benthic organisms while summer spates (uncommon in Washington) can completely denude streams of benthic biota.

  Most biota avoid spates either by migrating to calm backwaters or by having life cycles which are terrestrial or aerial at these times. However, when floods occur at unusual times the fauna may be severely depleted and require several years to recover (Goldman & Horne 1983).

Larger organisms, like salmonids, normally choose to ascend rivers or streams during spates or high water because there are fewer shallow water barriers. Severe floods are detrimental to smaller, particularly invertebrate biota if they leave only rocks and gravel. However, these increased water flows may improve fish migration by removing major obstacles. Adequate water levels can improve the environment for salmonid egg laying and survival by removing excessive silt. These benefits cannot occur if the lotic system has been dammed by aquatic weeds.

4.5 UNCERTAINTY ANALYSIS

Summary: The uncertainty analysis indicates that field studies often reflect the risk analysis that has been used to generate the label. Models that have been used since 1975 indicate that an acute risk quotient of <0.1 or a chronic risk quotient of <1.0 reflects the safety of the product to exposed aquatic animals under field situations. An acute risk quotient is generated by dividing the short-term predicted environmental concentration (short-term EEC) by the acute LC50 of the most sensitive species of concern within the ecosystem. Providing a 10-fold safety factor will insure that only 0.01 to 4% of the animals with similar sensitivity will be adversely affected. Dividing the long-term EEC by the chronic MATC or chronic predicted NOEC for the most sensitive species generates a chronic risk quotient. The MATC is currently the preferred value, as indicated in the Diquat RED (EPA RED, 1995). The MATC is the value that will be used if development, growth or reproductive effects are the end-points to be measured while the NOEC is used when mortality is the measured end-point. A safety factor is not necessary in chronic risk assessment since all animals with a similar sensitivity will not be affected by exposure to chronic EECs for the compound being evaluated.

Diquat has been evaluated as having potential adverse impact on Hyalella azteca *(RQ =1.22), pocket shrimp (RQ 0.144) apple snail (RQ = 0.17) and* Daphnia pulex *(RQ = 0.27 to 0.65). Since the level of concern (0.1) for the risk quotient was exceeded, the invertebrate biota were determined to potentially be at risk. This risk was verified by the adverse impacts seen in the field. In field tests conducted with these species, adverse impact on* Hyallela azteca *was noted at concentrations as low as 0.11 ppm c.e, 1.0 ppm c.e for* Daphnia pulex, *1.0 ppm c.e for four species of snail and 1.0 ppm for the water scorpion (Wilson, 1967, EPA Red, 1995, Gilderhaus, 1967 and Cooke, 1977).*

However, even the most sensitive species of fish (striped bass, walleye and small-mouth bass) did not yield risk quotients that exceeded the level of concern. Since the level of concern (0.1) was not exceeded, the fish biota was determined to be at low potential risk from exposure to diquat, which was verified by the lack of adverse impact from diquat seen in the field. In field tests with bluegill sunfish, largemouth bass and several native Nigerian species, no adverse impact was observed. The laboratory risk assessments, which were verified by field studies, indicate the utility of the method and the fair degree of certainty in indicating adverse impact or the lack thereof to the biota.

There have been very few chronic tests conducted with diquat and the data could be judged to be insufficient for chronic risk assessment. However, since diquat is rapidly dissipated, many authors feel that chronic exposure is not likely. Nevertheless, the chronic risk quotient based on estimated MATCs exceeds the level of concern (1.0) for Hyalella azteca *(RQ = 3.48) and* Daphnia pulex *(RQ = 1.01 to 1.35). Therefore, a few invertebrate species will be adversely impacted until the concentration of diquat falls below the level that is toxic (Wilson, 1967; Hilsenhoff, 1966; Berry et al, 1975, and Gilderhaus, 1967). Field studies show that both* Hyalella azteca, Daphnia pulex and Simocephalus vetulus *are adversely impacted and that these adverse impacts last from 1.5 to 4 months (Wilson, 1967; Newbold, 1975 in Shearer and Halter, 1980 and Gilderhaus, 1967). These time periods are sufficient to determine that the adverse impact to the invertebrate biota indicated by laboratory studies are confirmed by field test with invertebrates.*

In no case did the chronic risk quotient based on estimated MATCs exceed the level of concern (1.0) for fish. Therefore, it is unlikely that fish will be adversely impacted by chronic exposure to diquat. Field studies indicate that exposure to typical use rates of diquat in the field will not directly effect the survivorship, numbers (fish-catch), diversity, physical condition, movement, size structure or reproduction in various warm water fish (Bain and Boltz, 1992; Olaleye et al, 1993 and Gilderhaus, 1967). However, it was noted that the growth of adult blue-gills was less than in the control but that total harvest appeared to be unaffected indicating that weight decreased as over-all survivorship went up.

Classic laboratory risk assessment to determine the field safety of diquat was very effective as a predictive tool for diquat risk management. The acute risk quotient with diquat is >0.1 for the most sensitive invertebrate species and $\leq$0.1 *for all fish species that were tested for 96 hours. Treatment of ponds and creeks indicates that the impact of diquat on fish and amphibians is minimal and that only a few species of invertebrates are affected by the treatment with diquat.*

Fish-kills, increases in chaoborid numbers, increases in the weight of amphibians, and claDOeran numbers have all been noted as results of secondary effects. However, these effects appear to be due to reduction of DO to lethal levels, increases in foodstuff due to the death and decay of treated plants, and algal blooms respecitively. In general, similar effects were observed with chronic risk quotients. Chronic risk quotients of <1.0 for diquat for environmentally relevant fish predict the chronic safety of this herbicide to fish. However, chronic risk quotients of >1.0 for a few of the invertebrates indicates that this segment of the biota is at risk from exposure to diquat.

Furthermore, while chronic risk quotients of <1.0 for diquat generally predict chronic safety to fish, accurate prediction of chronic safety or lack of safety for aquatic invertebrates from exposure to treated sediment is not possible without an understanding of bioavailability factors that could mitigate the toxic effects of diquat sorbed to sediment. These factors could include maximum binding capacity of the sediment, bioavailability and likelihood of degradation on particular sediment types.

The assumptions of risk analysis contain specific safety factors that are discussed by Urban and Cook (1986). The model discussed by Urban and Cook has been used since 1975 and was designed to provide a safety factor that would allow for differential variability and sensitivity among fish and wildlife species.

It was assumed that the slope of the dose response curve for the effects of a pesticide on most fish and wildlife species would be unknown. Since it is impossible to test every non-target-species that might be exposed, the following factors influence whether a correct risk management decision will be made:

1) Does the model predict risk so that the biota will be protected? Statistical analysis of the effects of slope on estimating the acute LC50 indicates that an expected environmental concentration (EEC) value 10-times less than the acute LC50 would lead to 0.01 to 4% mortality.

2) Terrestrial organisms are believed to be less susceptible to environmental assault than aquatic species. Therefore, a less stringent EEC is used to designate unacceptable risk in these species. The less stringent EEC is 5-times less than the acute LC50 or LD50, which would lead to field mortality of approximately 10%, is used as a level of unacceptable risk in birds and mammals. The higher safety factor (listed in item 1) is believed to be necessary since aquatic organisms are less likely to be able to limit their exposure through behavioral modifications such as moving out of the treated area or switching to an alternative food source.

3) Larger safety factors are warranted for the protection of threatened and endangered species where a factor of 10-fold is insufficient to protect that segment of the biota. E.g. in cases where no mortality is acceptable, an EEC of 20 times less than the acute LC50 should be sufficient to ensure protection of species in which even a single death is of special concern.

4) For chronic effects, an EEC equal to the maximum allowable toxic concentration (MATC) or no observed effect level (NOEC) is believed to be sufficient to reduce risk to a minimum level, since statistical analysis indicates that if the EEC is less than the MATC or NOEC there is a 95% probability that no adverse impact to long-term survival, growth or reproduction will occur.

5) The above precautions will adequately protect any species that is acutely exposed to residues 10-fold lower than the EEC. However, to protect the entire biota or a segment of the biota, the acute EEC must be 10-fold lower than the LC50 for the most sensitive species that you wish to protect and the chronic EEC must be less than the chronic MATC of the most sensitive species that you wish to protect.

The above criteria are considered rough estimates of potential risk to non-target organisms. The model used for ecological risk assessment does not provide a mechanism for estimating absolute uncertainty or an unchangeable probability of safety to the biota.

If the tested species are representative of the biota and are sufficient in number, uncertainty can be reduced to a minimum. With very few exceptions, diquat herbicide is ranked as being moderately toxic to practically non-toxic to most environmentally relevant fish and invertebrate species. However, crustaceans may be significantly more susceptible to diquat than other species of aquatic invertebrate. Even with this observation, not all crustaceans have a similar response to diquat. For example, while *Hyalella azteca* and *Daphnia pulex* are very sensitive to diquat (LC50 = 0.048 ppm to 0.216 ppm c.e.), other species like copepods, ostracods and scuds appear to be more than 10-fold more tolerant to diquat than the aforementioned species (LC50 = ~5.4 to 25 ppm c.e.). Diquat has been tested with at least 33 species of fish and 26 species of invertebrate. Such a relatively large database provides confidence that risk assessment and risk management decisions will be adequate for protection of the more sensitive species in the biota. Nevertheless, a number of these invertebrate species (6) may not be relevant to this assessment since they are estuarine or marine species. However, since these estuarine and marine species are often more sensitive than their freshwater equivalents, additional sensitivity may be added to the risk assessment through their inclusion.

Observations of fish or invertebrate-kill due to the effects of diquat have been rare. Most cases of fish or invertebrate kill have been due to anaerobiosis caused by rotting, dead and dying vegetation (Frank, 1972). In some cases where death of sensitive species like *Hyalella azteca* and snails occurred, the adverse impact was believed to be due to changes in habitat resulting from the death of *Elodea canadensis* (Hilsenhoff (1966) or *Egeria densa* (Berry, 1975). The effects of these invertebrate-kills can be mitigated by the inclusion of an artificial substrate such as conservation webbing (Berry et al, 1975).

The Expected Environmental Concentrations (EEC), as presented in Section 4.3.2.5 (Risk Analysis in Aquatic Species), are believed to be fairly accurate based on many years of successful risk management. However, field data for individual water bodies, indicating both acute and chronic time-weighted average concentrations, could improve the ability to assess and manage risk, particularly for sensitive species.

The conclusion that diquat appears to cause adverse impact to only 15% of tested invertebrate species and virtually none of the tested fish species indicates that the small number of species that may be adversely impacted may be substituted in the food web by species of similar size and nutritional quality. Although studies on the effects of growth on fingerling bluegill sunfish indicate that this hypothesis is true, further investigation is necessary to discover whether or not this hypothesis is universally true. Mitigating factors that are inherent to the nature of the herbicidal product, like high solubility, low octanol/water partition coefficient and high sediment binding must be considered in order to make a risk assessment that is of value to the user. Also, the possibility of making an incorrect risk management decision must be weighed carefully, particularly if limited field data is available. Fortunately, long-term use of diquat allows for practical field experience to mitigate an adverse laboratory risk assessment with diquat.

4.6 ADDITIONAL INFORMATION NEEDS

Summary: The importance of the role of sediment in removing diquat from the environment should be investigated along with the effects of diquat in sediment on benthic organisms. Levels of diquat in the sediment are particularly important since under some circumstances, there is a potential for diquat to accumulate to very high levels. However, modeling work shows that typical diquat levels will not exceed 25 ppm c.e. in the field and actual field monitoring studies have shown that diquat may accumulate on sediment up to concentrations as high as 37 ppm c.e. when it is applied at typical application rates (Dyson and Takacs, 2000 and Frank and Comes, 1967). It is unlikely that these concentrations are high enough to be toxic to sediment organisms. Diquat in sediment at concentrations below ~ 250 ppm is believed to be irreversibly bound and biologically unavailable. Therefore, diquat that is bound to the sediment is unlikely to be toxic to either animals or plants (Wilson, 196;, Birmingham and Colman, 1983; Coats et al, 1966 and Daniel, 1972).

The effects of post-treatment plantings of native aquatic plants need to be investigated to determine if this is a practical approach to revegetation after the elimination of watermilfoil. However, wheat, duckweed, and Eurasian water milfoil that have been planted in soil/sediment continue to grow on soils that have been treated with 170 ppm c.e. to 5,000 ppm c.e. diquat. However, growth was somewhat inhibited with more sensitive plants and soils with relatively low adsorptive capacity. Further investigations with varying treatment rates and conditions should be conducted to determine which rates and conditions cause the greatest destruction of watermilfoil and the least damage to native aquatic plants.

The toxicity of diquat to sensitive fish and invertebrate species is well enough understood to adequately manage the risk associated with aquatic weed control. However, further investigations need to be conducted to determine what levels of diquat are safe to sensitive, threatened and endangered species (particularly salmon and sea-run trout). Additional studies emphasizing species indigenous to the Northwest should be conducted so that risk due to exposure can be managed more effectively. This is of particular concern for benthic organisms since regulators, registrants, the applicator community and the general public have recently expressed great concern over this issue.

4.6.1 Soil and Sediment

The concentrations of diquat in sediment, after application of diquat for the control of weeds, have been adequately investigated. Diquat has been used for 30 years without a release from the sediment into the water column that was high enough to adversely affect aquatic organisms (Dyson and Takacs, 2000). As discussed previously, use of diquat at normal field rates would require hundreds of treatments before the diquat concentrations exceeded the capacity of the sediment to adsorb it. The effects of partitioning (Kd) between soils and water with different soils/sediments has been adequately investigated. Typical Kd values on sand-gravel sediment, sand sediment, and silty clay loam sediment are 30 to 1,000, 1,000 to 10,000, and 10,000 to 60,000, respectively (Ritter et al, 1995). These extremely high Kd values classify diquat as immobile. Desorption is unlikely as long as 7% of the maximum sorptive capacity is not exceeded (Birmingham and Colman, 1983). Although diquat is persistent on sediment, it binds nearly irreversibly to soil/sediment. It does not appear to be biologically available to aquatic organisms in surface water. Therefore, EPA and other agencies have not required that surface water advisories be issued. Also, since diquat is immobile, it is unlikely to leach into ground water and there have been no credible reports of ground water contamination with the proper use of diquat to control aquatic weeds. (EPA RED, 1995).

The concentration of diquat in sediment has not exceeded 37 ppm c.e. when applied at typical maximum use rates (Frank and Comes, 1967). Although diquat has been seen to be stable on sediment for at least 180 days, modeling work indicates that the concentration of diquat on sediment should never exceed 25 ppm c.e. (Dyson and Takacs, 2000). Since these values do not exceed the sorptive capacity of sediment, effects of diquat on sediment organisms should be minimal. This has already been seen in sediment organisms like the aquatic isopod *Asellus communis* and *Hyalella azteca* where the toxicity of diquat in the presence of sediment is much lower than the toxicity of diquat when sediment is absent (Williams, 1984 and Wilson, 1967). However, the apple snail appears to be sensitive to diquat even in the presence of sediment (LC50 = 0.34 ppm c.e.)(EPA, RED, 1995).

With the current knowledge of the concentration of diquat that typically occurs in sediment and a fairly good understanding of the toxicity of diquat to sediment organisms, risk can probably be managed effectively to protect sediment species from the toxic effects of diquat. Only a few species of free-swimming and sediment invertebrates have been tested that are affected by exposure to diquat for 4 days or less. Eighty-five percent of these species are not affected by diquat. Since diquat only affects a few organisms that are important in the food web, the food web will probably not be disrupted by the proper use of diquat. Other species of similar size and nutritional value should be able to substitute for those species temporarily lost from the water column due to treatment with diquat for aquatic weed control (Wilson, 1967). The effects of diquat on invertebrates and the dependency of fish on the invertebrates are discussed extensively in section 4.3.2.

4.6.2 Water

The effects on water quality of diquat products have been adequately investigated. It has been demonstrated that diquat products can cause depletion of dissolved oxygen concentrations to nearly zero due to decay of treated foliage and that ammonia, nitrite and nitrate, phosphate and pH affect the toxicity and secondary effects of diquat (Frank, 1972; Daniel, 1972; Inabinet, 1976; Peverly and Johnson, 1979; Surber and Pickering, 1962; Wilson, 1967; Tooby et al, 1980, El-Deen and Rogers, 1992 and Cooke, 1977). These water quality parameters have been known to influence algal blooms and consequently the growth of zooplankton, fish, and amphibians.

4.6.3 Plants

Daniel (1972) and Peverly and Johnson (1962) adequately demonstrated that dead and dying plants release nitrogen and phosphorous. These nutrient releases, especially the subtle forms of phospherous, are utilized rapidly by phytoplankton, and secondarily by zooplankton, fish and amphibian larva (Peverly and Johnson, 1962; Inabiniet, 1972; Gilderhaus, 1967 and Cook, 1977). In the case of diquat, field data indicates that the levels of nitrogen and phosphate can change significantly after treatment with diquat (Peverly and Johnson, 1962 and Daniel, 1972), but at lower diquat concentration (~0.1 ppm c.e.) this nutrient enrichment may not occur (Sedar, 1997).

The need to plant desirable vegetation in the aquatic environment after treatment with diquat has yet to receive serious investigation. In the laboratory, it has been shown that duckweed will grow when sediment is present that contains 170 ppm c.e. diquat, which corresponds to a water column concentration of 3.4 ppm c.e. (Birmingham and Colman, 1983). At this concentration, the growth of *Lemna minor* is inhibited fairly strongly when the amount of diquat approaches about 7% of the adsorption capacity. However, Daniel (1972) found that transplanted Eurasian watermilfoil grew without signs of diquat toxicity in sediments that had been treated in the previous year for the control of watermilfoil. The growth of these milfoil transplants was not as robust as had been anticipated. This was probably due to competition from *Potamogeton* spp. that dominated the water body after the elimination of the previous year's stand of Eurasian watermilfoil.

However, Frank (1972) noted that post-treatment plantings of native non-noxious and non-invasive plants could increase diversity and decrease the numbers of the less desirable plants through competition. This would improve habitat since a more diverse plant community would attract a more diverse animal community. The practicality and utility of post-treatment plantings of native plants, and when they are best used, should be evaluated on a case-by-case basis.

4.6.4 Acute and Chronic Animal Studies

Very few well-designed chronic toxicity studies have been conducted with diquat products. For an ideal understanding of chronic effects, early life-stage (ELS) studies may need to be conducted on all end-use products or their technical equivalent with rainbow trout, fathead and sheepshead minnows. Salmon, steelhead and trout are important in the Northwest. Therefore, ELS studies may also need to be conducted with these species. To have a better understanding of the chronic effects of diquat products on invertebrates, life cycle studies may need to be conducted with all end-use products or their technical equivalent on *Daphnia magna*, *Ceriodaphnia dubia* and *Mysidopsis bahia*. In order for proper comparisons to be made, additional acute studies should be conducted in the same time frame as the new early life-stage and life cycle studies. Ideally, the acute and chronic studies should be conducted with fish from the same parental line, or at least of the same known and specified seriotypes or germ lines.

Ideally, additional acute and chronic work needs to be done on fully aquatic and water associated animal species. These species include aquatic reptiles (turtles), amphibians (salamanders, toads, and frogs), lepidoptera and other insects associated with wetland communities or used as biocontrol agents on aquatic plants. Additional work with amphibians will be important because the endangered leopard frog and African clawed toad have been shown to be acutely sensitive to diquat. The chronic MATCs for these species is 1.7 and 0.64 ppm c.e., respectively. Chronic toxicity test information (page 16) for rainbow trout is "not considered adequate." Therefore this test and identical tests need to be conducted on indigenous endangered salmonids.

Great concern has recently been expressed concerning the effects of pesticides on benthic (sediment) organisms. In light of the high toxicity of diquat products to a few benthic organisms, additional testing needs to be conducted to determine the extent of the toxicity caused by diquat in both acute and chronic tests. Furthermore, although the few studies conducted in the field indicate that benthic invertebrate species are (for the most part) not affected directly by diquat, additional field studies, emphasizing the species that have demonstrated sensitivity in the laboratory to diquat products, are warranted. These species include, but should not be confined to, *Hyalella azteca,* various claDOerans like *Daphnia pulex, Daphnia magna, Simocephalus* spp., various mollusks like *Pomacea pludosa*, *Limnea stagnalis, Helisoma* spp., *Graulis* spp. and *Physa* spp. Some of the more sensitive species of estuarine and freshwater shrimp may also need to be investigated for the effects of diquat in field situations. A few of these important species are the pocket shrim*p (Mysidopsis bahia*), and the *Palaemonetes* spp.*Palaemonetes* spp. is another genus of shrimp that includes the grass shrimp *(Palaemonetes pugio)* and the grass shrimp *(Palaemonetes kadiakensis*), which appear to be important prey organisms for fish that are commercially and recreationally important. Species which are resistant to diquat in laboratory and field situations may be used as negative controls; e.g., if an unexpected response occurs with these species, then there may be problems with the test design or there may be factors other than diquat which are influencing survival of the test organism. A few of the species that would make good negative controls include *Gammarus fasciatus*, various odonates, and bloodworms.

4.7 MITIGATION MEASURES

Summary: Methods to lower levels of released phosphates during post-treatment aquatic plant decay could be useful.

In emergency situations such as when excessive levels of diquat have been released into the water, diquat may be scavenged by dusting bentonite or montmorillonite clays onto the surface of the water body containing excess diquat. This approach is probably not practical in large water bodies but has been shown to be effective for removing paraquat from small pools (Coats et al, 1955). The use of charcoal to remove excess diquat from water may also have some utility (Shearer & Halter, 1980).

The use of diquat itself may be considered a mitigation measure when noxious floating and submersed aquatic macrophytes are out of control. Treatment with appropriate concentrations of diquat may improve habitat for fish, pelagic aquatic invertebrates (zooplankton) and benthic organisms (catfish, common carp and sediment dwelling organisms).

However, treatment with diquat can produce side effects that may need to be mitigated, i.e., release of too much phosphate due to the decay of treated plants. Also, if anaerobic conditions develop, phosphate and iron may be released and stimulate algal growth. In order for these releases from the anerobic sediment to be available to photosynthetic organisms, the water must be shallow, transparent, and mixable enough for algae blooms to occur. Removal of excess phosphate may be achieved by the addition of ferric iron, metals in fly ash, or salts of aluminum or zirconium. However, these remediation techniques may have an adverse impact on sensitive aquatic animals. Therefore, the negative impact of excess phosphate must be weighed against the possible negative affects of these chelating metals on the resident biota. This method is occasionally used to clean up the phosphate from eutrophic lakes and it could be used as a remedial measure when high phosphate levels are noticed due to the decay of herbicide treated aquatic plants. However, by the time high phosphate levels are noticed, it may already be too late to prevent an algal or bacterial bloom (Goldman and Horne, 1983).

Excess concentrations of diquat that need remediation are unlikely to occur except in cases where there has been a spill. For example, if a treatment boat sinks, concentrations near the boat will be high enough to cause an extensive fish-kill. There is some evidence that carp and suckers may avoid areas in the field where the concentration of diquat is very high (Hesser et al, 1972), and laboratory studies have shown that goldfish and mayfly larvae can avoid diquat at concentrations of approximately 1.0 ppm (Berry, 1984 and Folmar, 1978). Laboratory studies indicate that rainbow trout may passively avoid diquat at concentrations of 0.5 ppm or higher by drifting out of areas that have been exposed to diquat (Dodson and Mayfield, 1979). However, unless the concentration is very high, the ability of these species to avoid pesticides has been largely rejected by investigators conducting avoidance studies. Furthermore, since the ability of threatened and endangered species to avoid exposure to diquat is unknown, extra caution should be taken to allow for a level of concern of 0.05 rather than the more typical value of <0.10 for non-endangered species. Restrictions on seasonal applications are warranted to protect sensitive salmon smolts from the affects of diquat; similar seasonal restrictions may be applied to protect fish and fisheries, particularly when early life-stage (sac-fry), striped bass, smallmouth bass, largemouth bass, and walleye (*Stizostedion vitreu*) are present.

When diquat products are being used for control of aquatic weeds, the lowest effective concentration should be used. The use of these herbicides in open waterways where a lot of lateral mixing and dilution occur will also decrease the dissipation time (Ritter et al, 1995, Ritter et al, 2000).

In cases where sediment or water becomes seriously contaminated, dissipation rates may be improved by adding sediment with high levels of bentonite or montmorillonite clay content (Coats et al, 1966 and Shearer and Halter, 1980). It is also conjecture by this author that the addition of plants that are tolerant to diquat may assist in the removal of diquat from the waterway by direct adsorption. Some of these more tolerant species include *Nuphar* spp., *Nymphaea* spp., *Potamogeton nodosus and Potamogeton richardsonii*. However, no experimental evidence exists to prove the efficacy of this approach. The use of chemostats or biofilms containing species of bacteria that are known to degrade diquat also has potential for mitigating cases of diquat water pollution. Species of bacteria known to degrade diquat include *Pseudomonas fluorescens*, *Rhizobium* spp., and *Aerobacter aerogenes*. Nevertheless, this approach is only a hypothesis and no experimental evidence exists to support the use of chemostats and biofilms to remove diquat from water.

4.8 SUMMARY AND CONCLUSIONS

Summary: In general, diquat does not appear to adversely affect tested fish species. However, early life-stage striped bass, and smallmouth bass appear to be susceptible to the acute affects of *diquat (Table 28); but chronic exposure risk is not likely to be an issue (predicted MATC ranges from 0.017 to 0.2 ppm c.e.). It is noted that the 28-day time-weighted average EEC (0.0087 ppm) is less than the chronic MATC for the most sensitive species.*

Aquatic invertebrates are susceptible to the adverse impacts of diquat. Hyalella azteca*,* Daphnia pulex*,* Daphnia magna*, pocket shrimp (*Mysidopsis bahia*) and the apple snail (*Pomacea pludosa*) are especially susceptible to the effects of diquat in the laboratory. The acute risk quotient for these species is >0.1 which is higher than the level of concern for protection of the biota. The short-term EEC is 0.059 ppm. While this concentration is higher than the LC50 (0.048 ppm) only in* Hyalella azteca*, it is greater than one-tenth of the LC50 for the other species discussed here. Invertebrate biota that are chronically exposed to diquat may also be at risk, as indicated by the predicted or empirical laboratory risk quotients for* Hyalella azteca *(predicted Chronic RQ = 3.5 to 4.4) and* Daphnia pulex *(predicted Chronic RQ = 0.79 to 1.3). When the chronic risk quotient exceeds the level of concern (1.0), the invertebrate biota is considered to be at risk.*

The results from both fish and invertebrate laboratory studies have been verified in the field. No significant long-term adverse impact has been noted on various fish species from aquatic weed control practices. Numbers, size structure, condition, and movement within the treated area have been observed to be unaffected by the use of diquat at labeled use rates. However, the size of adult bluegills has been noted to be reduced when ponds are treated one- to three-times with 1.0 to 3.0 ppm c.e. However, under these situations, total survivorship is improved in treated ponds. The growth rate of fingerling bluegills and the total weight of the adult harvest appear to be unaffected (Gilderhaus, 1967). Both the number, siz,e and diversity of native Nigerian fish populations was improved when diquat was used to control waterhyacinth at concentrations up to 2 kg a.i./ha. (Olaleye et al, 1993).

Sensitive invertebrate species have been noted to be adversely impacted after field treatments with concentrations of diquat that ranged from 0.1 to 2.5 ppm. The numbers of Hyalella azteca *were reduced to very low population levels for up to 1.5 months after treatment with diquat at 2.5 ppm. Minimal concentration treatment with diquat at 0.11 ppm plus endothall at 0.17 ppm temporarily reduced the survivorship of* Hyalella azteca *in shallow water containing 0.73 ppm diquat after 12 hours of exposure but did not impact* Hyallela azteca *in deep water where the concentration of diquat was not higher than 0.03 ppm. Similarly, treatment of water bodies with 1.0 ppm diquat caused a reduction in the numbers of cladoerans (*Daphnia pulex *and* Simocephalus vetulus*) for as long as several months after treatment. Populations of these species recovered after diquat had dissipated to concentrations that have been shown to be non-toxic in laboratory experiments. Other field studies (Hilsenhoff, 1967) have shown that the removal of* Elodea canadensis *causes a reduction in the numbers of* Hyalella azteca *and four genera of aquatic snails. However, this appears to be a secondary effect due to habitat loss, and does not appear to be due to the direct action of diquat. The inclusion of artificial substrates like conservation webbing appears to mitigate the effects of habitat loss due to the reduction in American waterweed and Brazilian elodea stands.*

Although diquat adversely impacts the invertebrate biota, 85% of the invertebrate species tested will probably not be affected by diquat when it is used according to label instructions. Reduction in Hyalella azteca *and cladoerans populations at times when fry and fingerling fish are actively feeding could cause reduction survival and growth. However, the limited data available indicates that survival and growth of bluegill sunfish fingerlings is not affected by reductions of amphipod and cladoeran populations. In some cases ostracods and copepods, which are of a* *similar size and nutritional value, may be of values for these important fish-food species if n8umbers ofostracods and cladocerans are high enough.*

Although fish species and 85 % of the tested invertebrate species should survive the whole water body treatment with diquat at typical concentrations of 0.224 and spot treatments at 0.30 to 0.37 ppm, it is apparent that very few macrophytes will survive the higher dosages of diquat. The risk to aquatic macrophytes from exposure to diquat at concentrations of 0.224 ppm c.e. is high (RQ = 0.5 to 1.0) to very high (RQ =>1.0) even if the 4-day time-weighted average of 0.059 ppm c.e. is considered as the most typical exposure scenario. Even if the EPA's assumption that only an RQ of >1.0 exceeds the level of concern, the non-target macrophytes like Potamogeton natans and Elodea canadensis *will be adversely impacted by typical time-weighted average concentrations. Other species such as waterhyacinth, waterlettuce,* Hydrilla verticillata*, and Eurasian watermilfoil may survive, but these exotic noxious weeds are less desirable than other species. In general, diquat is considered to be ineffective in the control of* Nuphar *spp.,* Nymphaea *spp., and* Chara *spp. (Ecology, 1992); and only temporary control is achieved with duckweed (*Lemna *spp.), curlyleaf pondweed and watermilfoil. Typical time periods for control of these species are typically less than two months, which may make re-treatment necessary within a single season (Hulbert, 1987 and Shearer and Halter, 1980 cite Water Investigations Branch, 1977).*

*Although diquat claims efficacy for only two genera of filamentous algae (*Spirogyra *spp. and* Pithophora *spp.), other species of algae may be adversely impacted since the risk quotient exceeds the level of concern (1.0) when the time weighed average EEC is 0.059 ppm c.e. or higher. The risk to blue-green algae, diatoms and pyrrophytes is generally high. However, the unicellular green and filamentous green algae do not appear to be at risk since the RQ is generally less than the level of concern for these species. In microcosm and field studies, green algae are affected for only a short period of time and in general appear to be resistant to the affects of diquat and growth may be stimulated by concentrations of 1 to 10 ppm diquat. However, the most sensitive species of pyrrophyte (*Peridinium cinctum*) and floating leaf pondweed are extremely sensitive and will be affected even if they are located adjacent to areas that are directly sprayed with diquat. However, after spot treatment with 0.30 to 0.37 ppm c.e. diquat, all tested species of plant and algae, located at least 375 meters from the treatment site, should not be adversely impacted by treatment with diquat; it is noteworthy that under this scenario the risk quotient does not exceed the level of concern of 1.0. Therefore, the plant biota should be protected if this buffer zone of 375 meters is provided (Ritter et al, 2000). Singh and Williams (1997) found that treatment of 50 acres within a reservoir requires a 1,400 foot buffer to assure that the concentration of diquat applied at the maximum use rate will not exceed the MCLG (0.02 ppm c.e.). The comparison of a safe buffer distance, as determined by Ritter et al 2000 and Singh and Williams (1997), appears to be similar; e.g., 375 meters vs ~430 meters.*

Diquat was observed to have no adverse impact on fish species at application concentrations of less than 1.0 ppm when used in the field (Gilderhaus, 1967, Olaleye et al, 1993 and Bain and Boltz, 1992). Diquat was also only observed to affect Hyalella azteca *and cladoerans (water fleas) adversely when used in the field at these rates (Berry et al, 1975; Gilderhaus, 1967 and Newbold, 1975 in Shearer and Halter, 1980). Diquat may also affect several genera of pond snails when applied at 1.0 ppm c.e. (Hilsenhoff, 1966). However, these effects on snails were believed to be due to the secondary effect of habitat reduction due to the reduction in numbers of the target weed. Field applications of diquat at concentrations as low as 0.25 ppm control most aquatic macrophytes listed on the label (Johnson, 1962) and will adversely impact most the algal and macrophyte biota (Peterson, 1994). Diquat has been observed to stimulate the growth of* *unicellular and filamentous green algae in both the laboratory and field after initially suppressing growth slightly (Melendez et al, 1993 and Cook, 1977). These increases in green algae growth have been observed to stimulate the growth of frog and toad tadpoles as well as the cladoeran* Simocephahlus vetulus*, which is likely to be due to increases in available foodstuffs. An increase in available foodstuffs, due to the death of southern naiad, after treatment with diquat at 0.5 ppm has been observed to cause an explosion in the population of* Chaeoborous *spp. (Tatum and Blackburn, 1962).*

The risk to aquatic life from the use of diquat was assessed using two methodologies. One of the methods was designed to compare chemicals for toxicity and the other was designed to determine whether or not the chemical was safe to the biota.

The first method is the U.S. EPA ecotoxicological risk categories for mammals, birds, and aquatic organisms. A summary of its criterion can be found in Table 1. For fish, birds and mammals these categories are very highly toxic, highly toxic, moderately toxic, slightly toxic, and practically non-toxic. The exact quantitative values vary considerably depending on species and exposure route [(EPA, 1982, Brooks 1973 in Ebasco, 1993) (Table 1)]. This method classifies diquat as moderately toxic (LC50 = >1 to 10 ppm) to practically non-toxic (LC50 = > 100 ppm c.e.) for most species of fish. However, the toxicity of striped bass sac-fry (96-hour LC50 = 0.54 ppm c.e.) and walleye sac-fry (*Stizostedion vitreu*; 96-hour LC50 = 0.75 ppm c.e.) classify diquat as highly toxic (LC50 >0.1to 1.0 ppm). While the toxicity of diquat to 85% of the tested invertebrates classifies diquat as moderately toxic to practically nontoxic, 5 species are more sensitive to diquat. *Hyalella azteca* (96 hour LC50 = 0.048 ppm c.e.) is the most sensitive species and its response to diquat leads to a classification of very highly toxic (LC50 = <0.1 ppm). The other sensitive species are *Daphnia pulex* (48-hour LC50 = 0.16 to 0.22 ppm c.e.), apple snail (96-hour LC50 = 0.34 ppm c.e.), pocket shrimp (96-hour LC50 = 0.42 ppm c.e.), *Daphnia magna* (48-hour LC50 = 0.79 ppm c.e.) and the pond snail (*Limnea stagnalis* (8-day LC50 = >1.0 ppm). The response of these species to the toxicity of diquat, classifies this herbicide as highly toxic (LC50 = >0.1 to 1.0 ppm). This indicates that compared to other pesticides, the acute toxicity of diquat ranges from one of the more toxic pesticides to sensitive invertebrates to practically non-toxic to the least sensitive invertebrates and fish. In general, diquat should be classified as moderately toxic relative to other commercial pesticides. Moderate toxicity is verified by field studies with both fish and aquatic invertebrates. There has been only one credible report of fish-kill due to the affects of diquat at 3.0 ppm. However, it is unclear if this was due to the accompanying low dissolved oxygen content (DO $\leq$1.0 ppm) or a synergistic action from the combined effects of Curtrine® at 2.2 ppm plus diquat at 3.0 ppm (Daniel, 1972). Frank (1972) reports that it is unusual for registered aquatic herbicides like diquat, 2,4-D, endothall or dichlobenil to cause fish kills by the direct toxic action of the herbicide; and it is more usual when fish–kills are due to the secondary effect of anaerobiosis.

For the test substance to be considered safe to the biota, according to the second method (Urban and Cook, 1986), the acute LC50 must be at least 10 times greater than the 4-day time weighed expected environmental concentration (EEC). For threatened and endangered species, the acute LC50 must be 20 times greater than the 4-day weighted expected environmental concentration (EEC) for the test substance to be considered safe. The chronic NOEC or MATC must also be equal to, or greater than, the 28-day weighted EEC, for the test substance to be considered safe. Currently, the US EPA RED (1995) for diquat is using the MATC to determine this level of concern.

In many cases, risk assessments indicate that diquat may be acutely and chronically safe to fish. The acute 96-hour LC50 for the most sensitive species in this segment of the biota is 0.54 ppm c.e. (Table 18, Table 23 and Appendix 1) and the short-term time-weighted average EEC is typically 0.059 ppm when a whole water body is treated [according to EPA's worst case scenario of 4.48 Kg c.e./ha in 200 cm of water (0.224 ppm c.e.)]. Therefore, the risk quotient will be approximately equal to the level of concern of 0.1 for the protection of fish (RQ = 0.11). Diquat also has a low chronic toxicity with predicted or empirical chronic toxicity ranging from 0.014 ppm c.e. for striped bass to 0.79 ppm c.e. for Coho salmon, yellow perch, and other environmentally relevant species of fish (Table 23). Since the long-term EEC is 0.0087 ppm c.e. for typical treatment levels, where initial environmental concentration is 0.224 ppm c.e., the risk quotient is less than the chronic level of concern of 1.0 (RQ = 0.62) for fish.

Although the method is not standard for the EPA, very short-term exposures (1-2 days) to diquat at initial treatment rates of 0.224 ppm c.e. yield time-weighted average EECs of 0.146 to 0.102 ppm c.e. and 24- and 48-hour LC50s of 0.54 ppm c.e. for striped bass sac-fry. These toxicity and environmental values yield a risk quotient of 0.19 - 0.27, which is greater than the low level of concern (0.1) but lower than the high level of concern (0.5) for protection of fish biota. Therefore, fish may be adversely impacted. Use of diquat should not be permitted during spawning and when sac-fry and fry are present. Nevertheless, since the LC50s at all short-term exposure periods (1 to 4 days) is $\geq$ four-times the time-weighted average EEC, it is the opinion of risk assessors using the ECOFRAM approach that diquat is unlikely to cause adverse impact fish (Campbell et al, 2000). This approach uses the following logic: "In situations in which the 90th percentile for exposure is greater than the 10th percentile for toxicity, the margin of safety is less than 1.0 and indicates a significant risk to the aquatic animal communities. Margins of safety greater than 1.0 indicate that the distributions do not overlap (minimal risk to aquatic organisms)."

Field-tests with diquat generally support the conclusion that its use to control aquatic weeds does not harm the studied fish. Bain and Boltz (1992) found that treatment with diquat at 0.3 ppm c.e. plus Komeen® at 0.3 ppm copper did not adversely impact abundance size structure, condition or movement of largemouth bass within the treatment area. Cooke (1977) found that treatment with diquat at 1.0 ppm temporarily slowed the growth of the frog (*Rana temporia*) and the toad (*Bufo bufo*). However, these animals had recovered within 18 to 32 days, and were larger than frogs and toads in the control ponds. Gilderhaus (1967) found that single or multiple treatments at 1.0 to 3.0 ppm c.e. had no adverse impact on survival and no effect on the growth of bluegill sunfish fingerlings; but adults from treated ponds were significantly smaller than adults in control ponds although total adult harvest by weight was not significantly different. Furthermore, reproduction in adults from treated ponds appeared to remain high (100,000 fry/acre produced by 100 adults/acre). Cooke (1977) found that the smooth newt continued to reproduce in ponds that had been treated with diquat; but it could not be determined if reduction in total catch was due to the effects of diquat or other unrelated environmental effects. Olaleye et al (1993) found that a Nigerian creek treated with diquat at 2 Kg a.i./ha had significantly greater numbers of fish (67 fish/unit area) than the control area (8 fish/unit area), greater maximum size of the resident fish population and an increase in diversity (6 versus one species) in the treated areas versus the control areas.

Risk assessments indicate that diquat is neither acutely or chronically safe to invertebrates. The acute 96-hour LC50 for the most sensitive species in this segment of the biota is 0.048 ppm c.e. (Tables 19 & 23, and Appendices 2 & 4); and the short-term time-weighted average EEC is typically 0.059 ppm when a whole water body is treated according to EPA's worst case scenario of 4.48 Kg c.e./ha in 200 cm of water (0.224 ppm c.e.). Therefore, the risk quotient will be much greater than the level of concern of 0.1 for the protection of this segment of the biota (RQ = 1.2). Diquat also has a high chronic toxicity for invertebrates with predicted or empirical chronic toxicity ranging from 0.0025 ppm c.e. for *Hyalella azteca* to >5.2 ppm c.e. for dragonfly (*Libellula* spp.), bloodworms (polychaetes) and damselfly (*Enallagma* spp.). The long-term EEC is 0.011 or 0.0087 ppm c.e. for typical treatment levels where initial environmental concentration is 0.224 ppm c.e. and the dissipation time is 21 or 28 days, the risk quotient is significantly greater than the chronic level of concern of 1.0 for *Hyalella azteca* (RQ = 3.5 to 4.4) and *Daphnia pulex* (RQ = 1.0 to 1.30) for protection of this segment of the biota. Therefore, the invertebrate biota is likely to be at both acute and chronic risk from the treatment of diquat to control aquatic weeds. While 85% of the invertebrate species tested are not highly susceptible to diquat (acute LC50 = ≥1.0 ppm c.e. and predicted or measured Chronic MATC =>0.011 ppm c.e.), four or five species are sensitive enough to be at acute and or chronic risk from the effects of diquat. These species are *Hyalella azteca* (acute RQ = 0.46 to 1.2; chronic RQ = 3.5 to 4.4*), Daphnia pulex*, (acute RQ = 0.62 to 0.68; chronic RQ =1.0 to 1.3), apple snail (acute RQ = 0.17; chronic RQ = 0.48-0.61), pocket shrimp (acute RQ = 0.14; chronic RQ =0.4 to 0.5), *Daphnia magna* (acute RQ = 0.13; chronic RQ = 0.14 to 0.26) and pond snail (Acute RQ = <0.059; chronic RQ = <0.16 to <0.21). Furthermore, the acute risk may be apparent at both typical exposure periods of 48- to 96-hours and at a shorter exposure period of 24 hours. Fortunately, risk quotients generated from both very short-term and short-term exposure led to the same conclusion. The invertebrate biota is probably at risk from diquat exposure when it is used to control aquatic macrophytes.

It is the contention of some risk assessors using the ECOFRAM approach that the estimated time-weighted average EEC is too high and that typical spot treatment applications of 0.3 to 0.37 ppm will have 4-day time-weighted average EEC values of not higher than 0.021 ppm c.e. in Northwest waters. Using these EEC values yields a risk quotient of 0.44 for the most sensitive species, which exceeds the level of concern for low-risk (0.1) but does not exceed the level of concern for high-risk (0.5). Furthermore, since the ECOFRAM approach takes the position that if the margin of safety is greater than one for the most sensitive 10% of the organisms that minimal risk to aquatic organisms is likely (Campbell et al, 2000).

However, in Lake Steilacoom the three-day EEC exceeds 0.05 ppm c.e. (Serdar, 1997). Therefore, the level of concern is likely to be exceeded for invertebrates in this water body. However, at Gravelly Lake, where the maximum use rate was used only for spot treatments, the 1 to three-day EEC was always less than 0.003 ppm c.e., and the level of concern was not likely to be exceeded. Thus, invertebrate biota would be at risk at Lake Steilacoom but not at Gravelly Lake.

Field data support the risk assessment conclusions given above for invertebrates. However, populations of *Hyalella azteca* were unaffected in deeper water where the concentration of diquat never exceeded 0.03 ppm (Berry et al, 1975). Higher concentrations (2.5 ppm) of diquat largely eliminated *Hyalella azteca* from Soap Ponds in Oregon for up to 1.5 months (Wilson, 1967). Single or multiple treatments with 1.0 ppm diquat has been observed to significantly reduce the numbers of cladoerans (*Daphnia pulex* and *Simocephalus vetulus*) for two to four months. Recovery of these cladoeran populations did not occur until the levels of diquat dissipated to levels below the acute toxicity values (Gilderhaus, 1967 and Newbold, 1975 in Shearer and Halter, 1980). However, Hilsenhoff (1966) contends that the reduction in numbers of *Hyalella azteca* and four genera of pond snail is due to reduction in habitat rather than due to the direct toxic effects of diquat. These adverse impacts to *Hyalella azteca* and pond snails may be mitigated by the use of an artificial substrate such as conservation's webbing (Berry et al, 1975).

Other potential adverse impacts of diquat include adverse respiratory impacts on yellow perch exposed to diquat at concentrations of 1.0 ppm (Bimber et al, 1976). Concentrations higher than 0.1 ppm (0.054 ppm c.e.) appear to elevate the haematocrit, and increase plasma cortisol and plasma glucose levels in yellow perch (Holtz and Winter, 1989). Concentrations of 0.5 ppm c.e. impacted the swimming ability of rainbow trout by decreasing swimming speed by nearly one-half (Dodson and Mayfield, 1979). The ability of Coho salmon to migrate downstream was affected by concentrations as low as 0.5 ppm c.e., which may impact long term survivorship of this species. Very high concentrations of diquat cause avoidance reactions in fathead minnows, carp and suckers (de Peyester and Long, 1973, Hesser et al, 1972). El-Deen and Rogers (1992 and 1993) found that 53 ppm (29 ppm c.e.) diquat concentrations causes increases in the haematocrit levels, decreases in muscle and liver protein levels, and increases in muscle and liver transaminase levels. Although these effects do not lead to lethal acute toxicity, concentrations of diquat that may be found in the environment could influence the ability of fish to feed, migrate, find mates, breed, and resist disease which could lead to more subtle and as yet unverified chronic exposure effects.

With plants, close to 100% control would be expected in the area directly treated with diquat. The risk quotients generated under the worst case scenario described above would be considered very high (>1.0) for at least one species of algae and three species of aquatic macrophyte at time-weighted average EECs of 0.059 ppm c.e. and 0.021 ppm c.e. (Tables 13 and 17). These four-day EEC values were generated from two assumptions: 1) Treatment of a whole water body with 0.224 ppm c.e. and assuming a linear dissipation with a 0.75 hour half-life (Shaw et al, 1995); or 2) Spot treatment at 0.30 to 0.37 ppm with a dissipation that is typical of northwest reservoirs (Ritter et al, 2000). Therefore, there is likely to be a significant impact on non-target algae and plants in the treatment zone from treatment with diquat. If non-target algae and plants are located adjacent to the treatment zone and spot treatments are used to control weeds one can expect the time-weighted average EEC to be 0.008 ppm c.e. during the first four days of exposure. In this case, the algae *Peridinium cinctum* and the aquatic macrophyte *Potamogeton natans* are susceptible enough to generate risk quotients that are higher than the level of concern (1.0) for adverse impact on algae and macrophytes. Therefore, even if algae and macrophytes are located in areas not directly treated with diquat, they will be at risk from treatment with diquat to control aquatic weeds.

If the treatment site is located 375 meters from the location of non-target algae and plants, algae and macrophytes will probably be protected from the adverse impact of diquat application to control algae and aquatic weeds.

The four-day time-weighted average EEC (0.00028 ppm c.e.) is significantly less than the EC50 for the most sensitive algae (0.001 ppm c.e.) and the most sensitive plant species (0.0072 ppm c.e.) (Campbell et al, 2000). Field data on plants indicates that the above risk assessment is valid. At initial concentrations as low as 0.25 ppm, 95 percent of the treated vegetation was controlled. This included the following species: *Potamogeton richardsonii*, *P. pectinatus*, *Eleocharis acicularis*, *Lemna minor*, *Callitriche* spp. and the filamentous algae (*Mougeotia* spp. and *Zygnema* spp.). Other species that were controlled at this concentration included *Potamogeton crispus* and *Elodea canadensis* (Johnson, 1992 and Hulbert, 1987). Field tests with diquat at 0.13 to 0.11 ppm c.e. at Lake Steilacoom and Gravelly Lake provided effective control of *Elodea canadensis* and *Potamogeton* spp. (Serdar, 1997).

In conclusion, diquat is generally safe to use for control of nuisance aquatic vegetation at labeled use rates and provides a large safety factor for protection of most fish. However, early life-stage (sac-fry) striped bass, smallmouth bass, and similarly sensitive species may be at risk when exposed to diquat at labeled use rates. Also, impacts on salmonids life histories need to be evaluated.

Aquatic invertebrate biota are at risk from acute and chronic effects of diquat treatment at concentrations ranging from 0.1 to 1.0 ppm under whole pond treatment scenarios and reservoir spot treatments of 0.30 to 0.37 ppm c.e. Cladoerans (*Daphnia magna* and *Simochephalus vetulus*) have been reduced in numbers for up to several months after application of 1.0 ppm c.e. diquat. Furthermore, concentrations of 1.0 ppm c.e. diquat have been observed to sharply decrease the numbers of *Hyalella azteca* and four genera of snails. However, these effects were believed to be due to the destruction of native plant species that served as habitat. In the field, one other species of aquatic invertebrate (water scorpion) has been observed to cease reproduction after exposure to diquat at 1.0 ppm.

Not all invertebrates respond negatively to treatment with diquat. In a different case as reported in the previous paragraph, *Simocephalus* spp. was observed to grow in numbers after the initiation of an algal bloom brought on by nutrient release from decaying aquatic macrophytes. Also *Chaoborus* spp. were observed to almost double in numbers shortly after treatment with 0.5 ppm diquat due to an increase in decaying southern naiad which is used as a foodstuff.

Although a few of the tested invertebrate species are sensitive to diquat at labeled use rates, 85% of the tested species are not adversely impacted by diquat.

Use of diquat at labeled rates is likely to adversely impact nontarget species of algae and aquatic plants when the non-target species are located at the site of application or adjacent to the site of application. Non-target species of algae and aquatic plant will probably be unaffected if they are located 375 meters from the site of application.

Reward® LA may be used at applications rates of up to 4 .0 lbs. c.e./acre in water that is more than two feet deep. If the water body is less than two feet deep, the maximum use rate indicated on the label is 2 lbs c.e./acre. Reward® LA will control Eurasian watermilfoil at 1 to 2 gallons formulation/acre and is likely to adversely impact most species of native aquatic vegetation. Since control of Eurasians watermilfoil is often lost two months after treatment with diquat, it may be necessary to treat more than one time per season. As described above, diquat is effective in controlling a large number of aquatic macrophytes, although some of the species specified on the label may not be controlled with one application for the entire treatment season. Field data indicated the use of diquat products designated for aquatic use should be safe to fish and 85% of the invertebrates.

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LIST OF TABLES

Table 1: U.S. EPA Eotoxicological Catagories[1] for Mammals, Birds and Aquatic Organisms

Acute Oral Toxicity in Mammals (mg/Kg body wt)	Toxicity in Birds		Acute Toxicity in Fish and Invertebrates mg/L test solution	Toxicity Ranking
	Acute Oral (mg/Kg body weight)	Dietary mg/Kg feed		
<10	<10	<50	<0.1	Very Highly Toxic
10-50	10-50	50-500	0.1-1.0	Highly Toxic
>50-100	>50-500	>50-1000	>1-10	Moderately Toxic
>500-2000	>500-2000	>1000-5000	>10-100	Slightly Toxic
>2000	>2000	>5000	>100	Practically Non-Toxic

[1] Elizabeth Zucker, 1985. Hazard Evaluation Division, Standard Evaluation Procedure, Acute Toxicity Test for Freshwater Fish. PB86-129277. EPA-540/9-85-006

Table 2: Toxicity of Diquat to Different Aquatic Species; and Sensitive Species and Stages Tested

Species Name	Common Name	Test Type	Age of Organism	Test Duration	Test Chemicals – Diquat Products LC50 or EC50 & (MATC) in mg a.i./L			
					Hardness not Specified	Low Hardness (<50 ppm)	Intermediate Hardness (50 to 150 ppm)	High Hardness (>150 ppm)
Algae								
Anabaena flos-aquae	Blue-green algae	Static Acute	Log growth phase	0.9-days	0.089	NA	NA	NA
Anabaena flos-aquae	Blue-green algae	Static Acute	Log growth phase	3-days	0.047[a]	NA	NA	NA
Anabaena flos-aquae	Blue-green algae	Static Acute	Log growth phase	7-days	0.096[a]	NA	NA	NA
Anabaena inaequalis	Blue-green algae	Static Acute	Log growth phase	0.9-days	0.005	NA	NA	NA
Microcystis aeruginosa	Blue-green algae	Static Acute	Log growth phase	0.9-days	0.1	NA	NA	NA
Microcystis aeruginosa	Blue-green algae	Static Acute	Log growth phase	3-days	0.065	NA	NA	NA
Microcystis aeruginosa	Blue-green algae	Static Acute	Log growth phase	7-days	0.070[a]	NA	NA	NA
Lyngbya wollei	Blue-green algae	Static Acute	Log growth phase	3-days	0.145	NA	NA	NA
Lyngbya wollei	Blue-green algae	Static Acute	Log growth phase	7-days	0.175	NA	NA	NA
Pseudoanabaena spp.	Blue-green algae	Static Acute	Log growth phase	0.9-days	0.063	NA	NA	NA
Oscillatoria spp.	Blue-green algae	Static Acute	Log growth phase	0.9-days	0.082	NA	NA	NA

Table 2: Toxicity of Diquat to Different Aquatic Species; and Sensitive Species and Stages Tested (continued)

Species Name	Common Name	Test Type	Age of Organism	Test Duration	Test Chemicals – Diquat Products LC50 or EC50 & (MATC) in ppm c.e.			
					Hardness not Specified	Low Hardness (<50 ppm)	Intermediate Hardness (50 to 150 ppm)	High Hardness (>150 ppm)
Selenastrum capricornutum	Green algae	Static Acute	Log growth phase	0.9-days	0.492	NA	NA	NA
Selenastrum capricornutum	Green algae	Static Acute	Log growth phase	3-days	0.083[a]	NA	NA	NA
Selenastrum capricornutum	Green algae	Static Acute	Log growth phase	4-days	0.039[a]	NA	NA	NA
Selenastrum capricornutum	Green algae	Static Acute	Log growth phase	7-days	0.084[a]	NA	NA	NA
Scenedesmus quadricauda	Green algae	Static Acute	Log growth phase	0.9-days	0.652	NA	NA	NA
Chlorella vulgaris	Green algae	Static Acute	Log growth phase	3-days	>2.94	NA	NA	NA
Chlorella vulgaris	Green algae	Static Acute	Log growth phase	3-days	0.37[a]	NA	NA	NA
Chlorococcum spp.	Marine Green algae	Static Acute	Log growth phase	10-days	43	NA	NA	NA
Dunaliella tertiolecta	Marine Green algae	Static Acute	Log growth phase	10-days	6.5	NA	NA	NA
Navicula pelliculosa	Freshwater Diatom	Static Acute	Log growth Phase	3-days	0.019	NA	NA	NA
Navicula pelliculosa	Freshwater Diatom	Static Acute	Log growth phase	7-days	0.096[a]	NA	NA	NA

Cycotella meneghiana	Diatom	Static Acute	Log growth phase	0.9-days	0.092	NA	NA	NA
Nitzshcia spp.	Diatom	Static Acute	Log growth phase	0.9-days	0.065	NA	NA	NA

Table 2: Toxicity of Diquat to Different Aquatic Species; and Sensitive Species and Stages Tested (continued)

Species Name	Common Name	Test Type	Age of Organism	Test Duration	Test Chemicals – Diquat Products LC50 or EC50 & (MATC) in ppm c.e.			
					Hardness not Specified	Low Hardness (<50 ppm)	Intermediate Hardness (50 to 150 ppm)	High Hardness (>150 ppm)
Ochromonas danica	Diatom	Static Acute	Log growth phase	3-days	0.056[a]	NA	NA	NA
Ochromonas danica	Diatom	Static Acute	Log growth phase	7-days	0.059[a]	NA	NA	NA
Chryptomonas ovata	Diatom	Static Acute	Log growth phase	7-Days	0.046	NA	NA	NA
Chryptomonas ozolini	Diatom	Static Acute	Log growth phase	3-Days	0.035	NA	NA	NA
Chryptomonas ozolini	Diatom	Static Acute	Log growth phase	7-Days	0.038[a]	NA	NA	NA
Skeletonema costatum	Marine Diatom	Static Acute	Log growth phase	3-days	>2.94	NA	NA	NA
Skeletonema costatum	Marine Diatom	Static Acute	Log growth phase	7-days	>2.94	NA	NA	NA
Isochrysis galbana	Marine Diatom	Static Acute	Log growth phase	10-days	3.2	NA	NA	NA
Phaeodactylum tricornutum	Marine Diatom	Static Acute	Log growth phase	10-days	3.2	NA	NA	NA
Peridinium cinctum	Pyrrophyte	Static Acute	Log growth phase	7-days	0.001	NA	NA	NA
Macrophytes								
Lemna minor	Common duckweed	Flow-Through Acute	3-4 leaf stage	0.9-Days	0.004	NA	NA	NA

Table 2: Toxicity of Diquat to Different Aquatic Species; and Sensitive Species and Stages Tested (continued)

Species Name	Common Name	Test Type	Age of Organism	Test Duration	Test Chemicals – Diquat Products LC50 or EC50 & (MATC) in ppm c.e.			
					Hardness not Specified	Low Hardness (<50 ppm)	Intermediate Hardness (50 to 150 ppm)	High Hardness (>150 ppm)
Lemna minor	Common duckweed	Static acute	3-4 leaf stage	4-Days	0.018	NA	NA	NA
Lemna minor	Common duckweed	Static Acute	3-4 leaf stage	11-Days	0.0089[a]	NA	NA	NA
Lemna minor	Common duckweed	Flow-Through Acute	3-4 leaf stage	7-Days	0.0019[a]	NA	NA	NA
Lemna minor	Common duckweed	Static Acute	3-4 leaf stage	11-Days	0.0089	NA	NA	NA
Lemna minor	Common duckweed	Static Acute	3-4 leaf stage	14-Days	0.02	NA	NA	NA
Spirodela polyrhyza	Giant duckweed	Static Acute	NR	7	0.005	NA	NA	NA
Spirodela polyrhyza	Giant duckweed	Static Acute	3-4 leaf stage	16-days	0.01	NA	NA	NA
Spirodela polyrhyza	Giant duckweed	Static Acute	3-4 leaf stage	NR	0.00075	NA	NA	NA
Eichhornia crassipes	Waterhyacinth	Static Acute	NR	NR	0.1	NA	NA	NA
Eichhornia crassipes	Waterhyacinth	Static Acute	NR	16	0.114	NA	NA	NA
Hydrilla verticillata	Hydrilla	Static Acute	NR	NR	0.0099	NA	NA	NA

Hydrilla verticillata	Hydrilla	Static Acute	NR	16	0.08	NA	NA	NA

Table 2: Toxicity of Diquat to Different Aquatic Species; and Sensitive Species and Stages Tested (continued)

Species Name	Common Name	Test Type	Age of Organism	Test Duration	Test Chemicals – Diquat Products LC50 or EC50 & (MATC) in ppm c.e.			
					Hardness not Specified	Low Hardness (≤50 ppm)	Intermediate Hardness (>50 to 150 ppm)	High Hardness (>150 ppm)
Pistia stratiotes	Waterlettuce	Static Acute	NR	14	0.11	NA	NA	NA
Potamogeton natans	Floating leaf pondweed	Static acute	NR	14	0.0072	NA	NA	NA
Elodea canadensis	American waterweed	Static acute	NR	14	0.023	NA	NA	NA
Myriophyllum spicatum	Eurasian watermilfoil	Static acute	NR	14	0.046	NA	NA	NA
Wolfiella floridana	NR	Static acute	NR	7	~0.005	NA	NA	NA
Azolla caroliniana	Azolla	Static acute	NR	7	~0.01	NA	NA	NA
Azolla caroliniana	Azolla	Static acute	NR	14	0.02	NA	NA	NA
Azolla caroliniana	Azolla	Static acute	NR	16	0.12	NA	NA	NA
Wolffia columbiana	Watermeal	Static acute	NR	7	~0.1	NA	NA	NA
Fish and Amphibians								
Anguilla rostra	American eel	Static acute	Glass eel stage	4-days	NA	17	NA	NA

Anguilla rostra	American eel	Static acute	Black eel stage	4-days	NA	29.7	NA	NA
Brachdanio rario	Zebra fish	Static acute	NR	4 days	13	NA	NA	NA
Carrasius auratus	Goldfish	Static acute	NR	4-days	35	NA	NA	NA
Carrasius auratus	Goldfish	Static acute	390-600mm	4-days		85	NA	NA
Cirrhina mrigala	Hamilton's carp	Static acute	40-50mm	4-days	75	NA	NA	NA
Ctenopharyngodon idella	Grass carp	Static acute	95 to 235mm	4-days	NA	29	NA	125
Cyprinodon variegatus	Sheepshead minnow	Static acute	25mm	4-days	48	NA	NA	NA

Table 2: Toxicity of Diquat to Different Aquatic Species; and Sensitive Species and Stages Tested (continued)

Species Name	Common Name	Test Type	Age of Organism	Test Duration	Test Chemicals – Diquat Products LC50 or EC50 & (MATC) in ppm c.e.			
					Hardness not Specified	Low Hardness (≤50 ppm)	Intermediate Hardness (>50 to 150 ppm)	High Hardness (>150 ppm)
Cyprinus carpio	Common carp	Static acute	Various	4-days	<141	43[a] 27-67[b]	NA	35
Esox lusius	Northern pike	Static acute	NR	4-days	16	NA	NA	NA
Esox masquinongy	Muskellunge	Static acute	Sac-fry & fingerlings	4-days	NA	NA	5.2[a] 4.6-5.8[b]	NA
Erimyzon succeta	Lake chubsucker	Static acute	Sac-fry	2-days	1.8[a] 1.3-2.5	NA	NA	NA
Fundulus similis	Longnosed killifish	Static acute	NR	2-days	>1.0	NA	NA	NA
Fundulus heteroclitis	Killifish	Renewal Chronic	Egg to fry	32-days	<0.054 (0.017)	NA	NA	NA
Gambusia affinis	Mosquito fish	Static acute	31mm	4-days	NA	156	NA	NA
Ictalurus melas	Black bullhead	Static acute	0.8g	4-days	4.6	87	NA	NA
Ictalurus punctatus	Channel catfish	Static acute	13mm sac-fry & 50mm fingerlings	4-days	10[c]	NA	5.4[d]	NA
Ictalurus punctatus	Channel catfish	NR chronic	Egg to fry	NR	(>1.0)	NA	NA	NA
Lepomis macrochirus	Bluegill sunfish	Static acute	Various	4-days	28	31[a] 14-125[b]	12.8[a] 12.1-13.5[b]	76

Lepomis macrochirus	Bluegill sunfish	Static chronic	Sac-fry	12-days	(>10)	NA	NA	NA
Micropterus dolomieu	Smallmouth bass	Static acute	Sac-fry	4-days	1.3	NA	3.9	NA
Micropterus salmoides	Largemouth bass	Static acute	Fry & Fingerlings	4-days	NA	4.2	4.9	NA
Micropterus salmoides	Largemouth bass	Static acute	100mm subadults	4-days	NA	NA	60	NA

Table 2: Toxicity of Diquat to Different Aquatic Species; and Sensitive Species and Stages Tested (continued)

Species Name	Common Name	Test Type	Age of Organism	Test Duration	Test Chemicals – Diquat Products LC50 or EC50 & (MATC) in ppm c.e.			
					Hardness not Specified	Low Hardness (<50 ppm)	Intermediate Hardness (>50 to 150 ppm)	High Hardness (>150 ppm)
Morone saxatilis	Striped bass	Static acute	Sac-fry	4-days	NA	0.54	NA	NA
Morone saxatilis	Striped bass	Static acute	1-month Fry	4-days	NA	5.4	NA	NA
Morone saxatilis	Striped bass	Static acute	Fingerlings & Subadults	4-days	NA	43	NA	NA
Notoemigonus crysoleucas	Golden Shiner	Static acute	74mm	4-days	NA	NA	33	NA
Notropis atherinoides	Emerald Shiner	Static acute	55mm	4-days	NA	NA	13.9	NA
Oncorhynchus kisutch	Coho salmon	Static acute	45mm yearling & NR	4-days	25.1[a] 20.5-30[b]	NA	NA	NA
Oncorhynchus mykiss	Rainbow trout	Static acute	Fingerlings	4-days	9.5[a] 8-11.2	15	14.9[a] 6.1-26[b]	NA
Oncorhynchus mykiss	Rainbow trout	Flow-through acute	48.6mm Fingerlings	4-days	NA	NA	16	NA
Oncorhynchus mykiss	Rainbow trout	Flow-through Chronic	48.6mm Fingerlings	21-days	NA	NA	2.9 (<1.4)	NA
Oncorhynchus tshawytscha	Chinook salmon	Static acute	58-96mm Fingerlings	2-days	NA	NA	6.1	NA

Perca flavecens	Yellow perch	Static acute	0.6grams and NR	4-days	16.2[a] 4.4-60[b]	30.6	NA	NA
Pimephales promelas	Fathead minnow	Static acute	45 to 50mm	4-days	24	7.6	NA	70

Table 2: Toxicity of Diquat to Different Aquatic Species; and Sensitive Species and Stages Tested (continued)

Species Name	Common Name	Test Type	Age of Organism	Test Duration	Test Chemicals – Diquat Products LC50 or EC50 & (MATC) in ppm c.e.			
					Hardness not Specified	Low Hardness (≤50 ppm)	Intermediate Hardness (>50 to 150 ppm)	High Hardness (>150 ppm)
Pimephales promelas	Fathead minnow	Flow-through acute	Egg to fry	34-days	NA	(0.2)	NA	NA
Poecilla mexicana	Mexican molly	Static acute	NR	4-days	9.0	NA	NA	NA
Poecilla reticualta	Guppy	Static acute	10mm	4-days	4.4	NA	NA	NA
Poecilla reticualta	Guppy	Static acute	25mm adults	4-days	50	NA	NA	NA
Rasbora heteromorpha	Harlequin fish	Static Acute	20mm juvenile	2-days	18[a] 16-20[b]	NA	NA	NA
Rasbora trillineata	Scissor-tail rasbora	Static acute	30mm adults	4-days	30	NA	NA	NA
Rana pipiens	Leopard frog	Static chronic	Eggs & sperm to tadpoles	21-days	<54 (<54)	NA	NA	NA
Rana pipiens	Leopard frog	Static chronic	Eggs & sperm to tadpoles	21-days	NA	NA	NA	>5.4 (1.7)
Salmo trutta	Brown trout	Static acute	Fingerlings & NR	4-days	19[a] 17.8-20[b]	NA	NA	NA
Salvelinus fontinalis	Eastern brook trout	Static acute	NR	1-days	>11	NA	NA	NA

Salvelinus fontinalis	Eastern brook trout	Static chronic	Egg to sac-fry	NR	(3.2)	NA	NA	NA
Stizostedion vitreu	Walleye	Static acute	8 to 10 day sac-fry	4-days	NA	NA	0.75	NA
Stizostedion vitreu	Walleye	Static acute	41 to 43 day Fry	4-days	NA	NA	1.5	NA
Stizostedion vitreu	Walleye	Static acute	84-46-day Fry & NR	4-days	2.1	NA	4.9	NA
Xenopus laevis	African clawed toad	Static chronic	Embryo to tadpole	10-days	~0.41 (<0.27)	NA	NA	NA

Table 2: Toxicity of Diquat to Different Aquatic Species; and Sensitive Species and Stages Tested (continued)

Species Name	**Common Name**	**Test Type**	**Age of Organism**	**Test Duration**	**Test Chemicals – Diquat Products LC50 or EC50 & (MATC) in ppm c.e.**			
					Hardness not Specified	**Low Hardness (≤50 ppm)**	**Intermediate Hardness (>50 to 150 ppm)**	**High Hardness (>150 ppm)**
Invertebrates								
Alonella spp.	Water flea	Static acute	NR	2-days	<5.4	NA	NA	NA
Asellus communis	Isopod	Static acute in sediment	NR	5-days	>10	NA	NA	NA
Callibaetis spp.	Mayfly	Static acute	NR	4-days		NA	16.4	NA
Cardina nilotica	Freshwater shrimp	Static acute	Adult	1-day	108.8	NA	NA	NA
Cardium edule	Cockle	NR	NR	2-days	5.4	NA	NA	NA
Cragnon cragnon	Shrimp	NR	NR	2-days	>10	NA	NA	NA
Crassostrea virginica	Easter oyster	Flow-through	Spats	4-days	>54.9	NA	NA	NA
Crassostrea virginica	Easter oyster	Flow-through	Juveniles	4-days	141.5	NA	NA	NA
Cypria spp.	Ostracod	Static acute	NR	2-days	NA	7.5	NA	NA
Diapotomus spp.	Copepod	Static acute	NR	NA	NA	5.4	NA	NA
Diapotomus spp *& Eucyclops spp.*	Copepod	Static acute	NR	NA	NA	19	NA	NA

Eucyclops spp.	Cyclops	Static acute	NR	2-days	NA	25.2	NA	NA
Daphnia pulex	Water flea	Static acute	Newborns	2-days	0.162	NA	NA	NA
Daphnia pulex	Water flea	Static acute	Adults	2-days	0.216	NA	NA	NA
Daphnia magna	Water flea	Static acute	Newborns	2-days	0.64[a] 0.324-1.03[b]	1.2	NA	1.62
Daphnia magna	Water flea	Static acute	Adults	2-days	1.6	NA	NA	NA
Daphnia magna	Water flea	Flow-through chronic	1-day to adult	21-days	(0.045)	NA	NA	NA

Table 2: Toxicity of Diquat to Different Aquatic Species; and Sensitive Species and Stages Tested (continued)

Species Name	**Common Name**	**Test Type**	**Age of Organism**	**Test Duration**	**Test Chemicals – Diquat Products LC50 or EC50 & (MATC) in ppm c.e.**			
					Hardness not Specified	**Low Hardness (<u><</u>50 ppm)**	**Intermediate Hardness (>50 to 150 ppm)**	**High Hardness (>150 ppm)**
Enallagma spp.	Damselfly	Static Acute	Nymph	4-days	NA	NA	>100	NA
Gammarus fasciatus	Lined scud	NR & Static Acute	NR & Mature	4-days	18.7	51	54	NA
Hyalella azteca	Amphipod	Static acute	4-8mm	4-days	NA	NA	0.048	NA
Hyalella azteca	Amphipod	Static acute in sediment	4-8mm	4-days	NA	NA	6.8	NA
Libellua spp.	Dragonfly	Static acute	Nymphs	4-days	NA	NA	>100	NA
Limnephilus spp.	Caddisfly	Static acute	Larvae	2-days	NA	NA	33	NA
					NA	NA	NA	NA
Limnea stagnalis	Pond snail	Static acute	NR	8-days	>1.0	NA	NA	NA
Mysidopsis bahia	Pocket shrimp	Static acute	New born	4-days	0.42	NA	NA	NA
Nymphula resopnsalis	Aquatic moth (Salvinia biocontrol agent)	Dip	Larvae	4-days	86.94	NA	NA	NA

Panaeus duroraum	Pink shrimp	Static acute	Juvenile	2-days	8.48	NA	NA	NA
Paneus setriferus	White shrimp	Flow-through acute	NR	2-days	>1.0	NA	NA	NA
Pomacea pludosa Apple snail	Apple Snail	Static acute	7-days	4-days		0.34		
Streptocephalus seali	Fairy Shrimp	Static acute	Mature	2-days			>2.0	
Tendipedinae	Bloodworms	Static acute	Larvae	4-days			>100	

[a] Geometric mean
[b] Range
[c] Sac-fry
[d] Fingerlings
NA = Not Applicable

Table 3: Soil Erodability Factors

Surface Layer Texture	Estimated K
Clay, Clay Loam, Loam, Silty Loam	0.32
Fine Sandy Loam, Loamy very Fine Sand, Sand Loam	0.24
Loamy Fine Sand, Loamy Sand	0.17
Sand	0.15
Silt Loam, Silty Clay Loam, Very Fine Sandy Loam	0.37

Source: Barfield et al., 1981in EBASCO, 1993 and USDA. 1978a in EBASCO, 1993

Table 4: Classification of Pesticides Based on Bioconcentration Factor

Classification	Bioaccumulation Factor[1]	Characteristics
Non-accumulative	≤10	Pesticide readily decreases when organism is removed from exposure
Slightly accumulative	60-700	Pesticide is gradually lost when organism is removed from exposure or pesticide is degraded by organisms
Moderately accumulative	700-8,000	Pesticide is gradually lost when organism is removed from exposure, or pesticide is slowly degraded by organism
Highly accumulative	≥8,000	Pesticide is not lost when an organism is removed from exposure, or pesticide is not significantly degraded by organism

Source: Weber, 1977 in EBACO, 1993

[1] As defined by Weber, 1977 in EBACO, bioaccumulation = concentration in aquatic organism/ concentration in water.

Table 5: Concentration of Diquat in Water, Hydrosoil and Fish for Diquat Applied to Ponds in Wisconsin

Time after Application (Days)	Application Rate (ppm c.e.)	Water (ppm c.e.)	Hydrosoil (ppm c.e.)	Fish (ppm c.e.)	Concentration Factor Soil	Concentration Factor Fish
3	1.0	0.57	NR[1]	0.14	NR	0.25
10	1.0	0.16	NR	0.49	NR	3.1
21	1.0	<0.01	NR	0.09	NR	>9.0
42	1.0	<0.01	2.74	0.03	>274	>3.0
84	1.0	<0.01	5.02	<0.01	>502	ND[2]
168	1.0	<0.01	6.47	<0.01	>647	ND
3	1.0	0.65	NR	0.13	NR	0.2
10	1.0	0.13	NR	0.41	NR	3.2
21	1.0	<0.01	NR	0.16	NR	0.01
42	1.0	<0.01	2.53	0.06	>253	>6.0
84	1.0	<0.01	11.4	<0.01	>1,140	ND
168	1.0	<0.01	10.1	<0.01	>1.010	ND
3	3.0	2.52	NR	0.40	NR	0.16
10	3.0	1.03	NR	1.45	NR	1.4
21	3.0	0.12	NR	0.35	NR	2.9
42	3.0	<0.01	3.98	0.20	>3,980	>20
84	3.0	<0.01	22.8	<0.01	>2,280	ND
168	3.0	<0.01	20.2	<0.01	>2,020	ND
3	3.0	2.39	NR	0.56	NR	0.23
10	3.0	0.76	NR	1.12	NR	1.5
21	3.0	0.07	NR	0.35	NR	5.0
42	3.0	0.01	19.50	0.06	1,950	6.0
84	3.0	<0.01	45.8	0.03	>4,580	>3.0
3 Mean[3]	1.0	0.61	NR	0.13	NR	0.21
10 Mean	1.0	0.14	NR	0.45	NR	3.2
21 Mean	1.0	<0.01	NR	0.12	NR	>12.0
42 Mean	1.0	<0.01	2.63	0.04	>263	>4.0
68 Mean	1.0	<0.01	7.56	<0.01	>756	ND
168 Mean	1.0	<0.01	8.08	<0.01	>808	ND
3 Mean	3.0	2.45	NR	0.40	NR	0.16
10 Mean	3.0	0.88	NR	1.27	NR	1.44
21 Mean	3.0	0.09	NR	0.40	NR	4.4
42 Mean	3.0	<0.01	8.80	0.11	>880	>11
84 Mean	3.0	<0.01	32.3	<0.02	>3,230	ND
168 Mean	3.0	<0.01	31.94	<0.01	>3194	ND

[1] NR = Not Reported

[2] ND = Not Determined

[3] Geometric mean of two samplings

Table 6: Dissolved Oxygen Concentration (mg/L) at Different Temperatures

Temperature in Degrees Centigrade	Dissolved Oxygen Concentration in mg/L
0	14.2
1	13.9
2	13.5
3	13.1
4	12.7
5	12.4
6	12.1
7	11.7
8	11.5
9	11.2
10	10.9
11	10.7
12	10.5
13	10.2
14	10.0
15	9.8
16	9.6
17	9.4
18	9.1
19	9.0
20	8.9
21	8.6
22	8.5
23	8.4
24	8.3
25	8.2

Table 7: Relationship of pH and Temperature to the Percent of Unionized Ammonia [NH4OH + NH3 (dissolved)] in Freshwater

pH	**Temperature (°C)**				
	5°	**10°**	**15°**	**20°**	**25°**
6.5	0.04%	0.06%	0.09%	0.13%	0.18%
7.0	0.12%	0.19%	0.27%	0.40%	0.55%
7.5	0.39%	0.59%	0.85%	1.24%	1.73%
8.0	1.22%	1.83%	2.65%	3.83%	5.28%
8.5	3.77%	5.55%	7.98%	11.2%	15.0%
9.0	11.0%	15.7%	21.4%	28.5%	35.8%

Table 8: Effects of pH and Hardness on the Toxicity of Diquat[1]

Hardness (ppm)	pH	Fish Species	96 hour LC50 (ppm)
			Diquat c.e.
Soft 21-22	7.4	Bluegill sunfish	39[1]
Soft 20-22	7.1-7.5	Fathead minnow	7.6[1]
Soft 21	7.0-7.6	Grass carp[2]	29[2]
Hard 310-312	8.1-8.2	Bluegill sunfish	76[1]
Hard 275-299	8.2	Fathead minnow	70[1]
Hard 270	8.1	Grass carp	125[3]

[1] Surber and Pickering (1962)
[2] El-Deen & Rogers (1992)
[3] Tooby et al, (1980)

Table 9: Toxicity of Adjuvants Registered for Aquatic Use to Aquatic Animals*

Adjuvant	**Use**	**Use Rate L/ha**	**Depth for LC50 to be Achieved**	**96 hr LC50 (mg/L)**		
				Bluegill	**Rainbow Trout**	**Daphnia magna**
Spar-Mate®	Surfactant	140	1.5	0.96	NA	NA
R-11®	Surfactant	NA	NA	4.2-5.5	3.8	19
X77®	Surfactant	4.7	0.1	4.3	4.2	2.0
Cide-Kick II®	Surfactant	7.0	0.1	4.3-5.2		
Widespread®	Surfactant	NA	NA	7.0	6.6	16
Induce®	Surfactant/ Accelerant	NA	NA	7.3	8.3	18
Super Spread 200®	Surfactant	NA	NA	9.3	NA	NA
Liqua Wet®	Surfactant	NA	NA	11.0	13	7.2
Spreader Sticker®	Surfactant/ Sticker	NA	NA	35	36	48
Formula 403	NA	18.7	0.1	37	NA	NA
IVOD®	NA	18.7	0.1	37	NA	NA
Passage®	NA	NA	NA	52	75	17
Big Sur®	NA	4.7	<0.1	112	NA	NA
Nalquatic®	Thickener	9.3	<0.1	200	NA	NA
LI-700®	NA	NA	NA	210	130	170
Agri Dex®	Surfactant/ Accelerant	NA	NA	>1000	>1000	>1000
Polysar®	Thickener	4.7	<0.1	3600	NA	NA
Herbex®	NA	2.3	<0.1	8000	NA	NA
Foamer®	Anti-Foam	NA	NA	NA	NA	NA
No Foam A®	Anti-Foam	NA	NA	NA	NA	NA
Dyne Amic®	Surfactant	NA	NA	NA	NA	NA
Penetrator®	Surfactant/ Accelerant	NA	NA	NA	NA	NA

NA = Not Available
* Only R-11, X77 and LI700 are currently allowed for aquatic use in Washington State.

Table 10: Effects of Diquat Treated Irrigation Water on Various Crops

Crop	Concentration in Water (ppm)	Mode of Irrigation	Phytotoxic Effect	Effects on Yield	Residue in Crop (Mg diquat/Kg crop)
Corn	125	NR[1]	None	None	NR[1]
Field Beans	5	NR	None	None	NR
Wheat	0.01 at ¼, ½ and ¾ maturity	Sprinkler (0.16 acre-inch)	Plants grow effectively	Plants grow effectively	<0.01
Carrots	0.01 at ¼, ½ and ¾ maturity	Sprinkler (0.16 acre-inch)	Plants grow effectively	Plants grow effectively	<0.01
Lettuce	0.01 at ¼, ½ and ¾ maturity	Sprinkler (0.16 acre-inch)	Plants grow effectively	Plants grow effectively	<0.01
Wheat	0.01 at ¼, ½ and ¾ maturity	Furrow (0.16 acre-inch)	Plants grow effectively	Plants grow effectively	<0.01
Carrots	0.01 at ¼, ½ and ¾ maturity	Furrow (0.16 acre-inch)	Plants grow effectively	Plants grow effectively	<0.01
Lettuce	0.01 at ¼, ½ and ¾ maturity	Furrow (0.16 acre-inch)	Plants grow effectively	Plants grow effectively	<0.01
Potatoes	0.67 at Late Stage of Maturity	Sprinkler (1 acre-inch)	No phytotoxic effects were mentioned	No yield effects were mentioned	<0.05
Sorghum	0.45 at Early Stage of Maturity	Sprinkler (1 acre-inch)	No phytotoxic effects were mentioned	No yield effects were mentioned	<0.10
Sorghum	0.45 at Late Stage of Maturity	Sprinkler (1 acre-inch)	No phytotoxic effects were mentioned	No yield effects were mentioned	<0.10
Soybeans	0.45 at Early Stage of Maturity	Sprinkler (1 acre-inch)	No phytotoxic effects were mentioned	No yield effects were mentioned	<0.10

Table 10: Effects of Diquat Treated Irrigation Water on Various Crops (continued)

Crop	Concentration in Water (ppm)	Mode of Irrigation	Phytotoxic Effect	Effects on Yield	Residue in Crop (Mg diquat/Kg crop)
Soybeans	0.45 at Late Stage of Maturity	Sprinkler (1 acre-inch)	No phytotoxic effects were mentioned	No yield effects were mentioned	<0.10
Carrots	0.09 at Early Stage of Maturity	Sprinkler (1 acre-inch)	No phytotoxic effects were mentioned	No yield effects were mentioned	<0.05
Carrots	0.45 or 2.25 at Late Stage of Maturity	Sprinkler (1 acre-inch)	No phytotoxic effects were mentioned	No yield effects were mentioned	<0.05
Lettuce	0.09 or 0.45 at Late Stage of Maturity	Sprinkler (1 acre-inch)	No phytotoxic effects were mentioned	No yield effects were mentioned	<0.05
Onions	0.45 or 2.25 at Late Stage of Maturity	Sprinkler (1 acre-inch)	No phytotoxic effects were mentioned	No yield effects were mentioned	<0.05
Potatoes	0.67 at Late Stage of Maturity	Soil (1 acre-inch)	No phytotoxic effects were mentioned	No yield effects were mentioned	<0.05
Sorghum	0.45 at Early Stage of Maturity	Soil (1 acre-inch)	No phytotoxic effects were mentioned	No yield effects were mentioned	<0.10
Sorghum	0.45 at Late Stage of Maturity	Soil (1 acre-inch)	No phytotoxic effects were mentioned	No yield effects were mentioned	<0.10
Soybeans	0.45 at Early Stage of Maturity	Soil (1 acre-inch)	No phytotoxic effects were mentioned	No yield effects were mentioned	<0.10
Soybeans	0.09 or 0.45 at Late Stage of Maturity	Soil (1 acre-inch)	No phytotoxic effects were mentioned	No yield effects were mentioned	<0.10
Carrots	2.25 at Late Stage of Maturity	Soil (1 acre-inch)	No phytotoxic effects were mentioned	No yield effects were mentioned	<0.05
Lettuce	0.09 or 0.45 at Late Stage of Maturity	Soil (1 acre-inch)	No phytotoxic effects were mentioned	No yield effects were mentioned	<0.05
Onions	0.45 or 2.25 at Late Stage of Maturity	Soil (1 acre-inch)	No phytotoxic effects were mentioned	No yield effects were mentioned	<0.05

[1] NR = Not reported.

Table 11: Plant Susceptibility to Diquat

Weeds – Killed by application of Diquat		
Easily Killed Aquatic Plants (EPA RED, 1995):		
Plant	EC50 (lbs cation/acre)	EC50 (ppm c.e.)
Giant duckweed	0.0036	0.00075
Waterhyacinth	0.0198	0.114
Azolla	0.0277	0.0116
Hydrilla	NR	0.0099
Aquatic Plants Controlled in the Field (Johnson, 1962 and Hulbert, 1987)		
Plant	EC95 (ppm c.e.)	EC100 (ppm c.e.)
Richardson’s pondweed	0.25	0.5
Sago pondweed	0.25	0.5
Eleocharis acicularis	0.25	0.5
Lemna minor	0.25	0.5
Callitriche sp.	0.25	0.5
Filamentous algae (*Mougeotia*	0.25	0.5
and *Zygnema*)	0.25	0.5
Aquatic Plants and Algae Controlled at Maximum Field use Rate (~0.75 ppm c.e.) and % inhibition CO_2 uptake Growth (Peterson et al, 1994):		
Plant or Algae	% Inhibition of CO_2 uptake	
Cyclotella meneghiana	99	
Nitzschia sp.	100	
Scenedesmus quadricauda	53	
Selenastrum capricornutum	69	
Microcystis aeruginosa	100	
Oscillatoria sp.	100	
Pseudoanabaena sp.	100	
Anabaena inaequalis	100	
Aphanizomenon flos-aquae	100	
Duckweed *(Lemna minor)*	100	
Crops (EPA RED, 1995):		
Easily killed:		
Crop	EC25 (lbs/acre)	
Corn	0.011 to 0.016	
Sweet Corn	0.016	
Wheat	0.016	
Cotton	0.0047	
Soybeans	0.0074	

Table 11: Plant Susceptibility to Diquat (continued)

Aquatic Plants Controlled at application rates of 2 lbs /acre in 9-10 inches of water (2.5 ppm) (Wilson, 1967)	
Plant or Algae	Observation
Brazilian elodea	Total regrowth within 9 months.
American water weed	Total regrowth within 9 months.
Ludwigia palustris	Total regrowth within 9 months.
Potamogeton foliosus	Total regrowth within 9 months.
P. natans	Total regrowth within 9 months.
Eleocharis macrostachya	Regrowth over larger area than before treatment.

Table 12: Acute to Chronic Ratio for Aquatic Organisms

Species	Length of Chronic Test	Diquat Formulation	Acute Toxicity (LC50 ppm c.e)	Chronic Toxicity MATC[1] (ppm c.e.)	Ratio[2]	References
Bluegill sunfish	12 day chronic	Diquat	14	>10	>1.4	Thompson, 1980; Hiltibran, 1967
Eastern brook trout	Early life stage	Diquat	>11.0	3.8	>2.9	Eadey & Reaney, 1965
Channel catfish	NR (life cycle)	Diquat	>5.4->10.0 (>7.3)[3]	>1.0	>7.3	Jones, 1965; Lawrence et al, 1965; EPA RED, 1995
Fathead minnow	34 days (life cycle)	Diquat	7.6[5]	0.2[5]	38	Surber & Pickering , 1962; Suprenenat, 1987
Rainbow trout	21-day chronic (fry)	Diquat	15.0	MATC = <1.4 LC50 = 2.9	>11 5.2	Tapp & Caunter, 1989; Thompson et al, 1980
Daphnia magna	21 days (Life cycle)	Diquat	0.32-1.6[6] (0.87)	0.045[6]	19	4

[1] Chronic values are MATC values unless otherwise noted.

[2] Geometric mean and of all pertinent data = 26.9 (16.5 to 43.8) where item in parenthesis is the range based of $10^{\text{Mean-st.dev)}}$ to $10^{\text{(Mean+St.dev.)}}$.

Greater than values were not used to calculate the geometric mean of the acute/chronic ratio and chronic LC50s are not used to calculated geometric mean of the acute/chronic ratio.

[3] Values in parenthesis are geometric means of all valid individual values.

[4] Benjit-Clause & Persoone, 1975, EPA RED, 1995, Wheeler, 1978, EPA RED, 1995, Bishop and Perry, 1961 and Suprenant, 1987.

[5] Test conducted in soft water.

[6] *Daphnia magna* grows best in hard water.

Table 13: Acute Toxicity of Diquat to Algae and Aquatic Macrophytes

Species	Test Media	End Point	Length of Test (days)	%A.I.	EC50 (ppm c.e)	Source
Lemna minor (Common duckweed)	Peterson's solution	$^{14}CO_2$ uptake	0.9	NR	0.004	Peterson et al, 1997
Lemna minor (Common duckweed)	NR[1]	Growth of biomass	4	Technical (~100%)	0.018	Fairchild et al, 1997
Lemna minor (Common duckweed)	NR	Chlorosis	7	NR	~0.005	Blackburn & Weldon, 1965
Lemna minor (Common duckweed)	Hunter's Solution	Frond count	7	35.3	0.0017	Bishop and Perry, 1981
Lemna minor (Common duckweed)	Hunter's Solution	Dry weight	7	35.3	0.0014	Bishop and Perry, 1981
Lemna minor (Common duckweed)	Hunter's Solution	Root Length	7	35.3	0.00081	Bishop and Perry, 1981
Lemna minor (Common duckweed)	Hunter's Solution	Growth Rate	7	35.3	0.0025	Bishop and Perry, 1981
Lemna minor (Common duckweed)	NR	Chlorosis	11	NR	0.008-0.01	Funderburk and Lawrence, 1963
Lemna minor (Common duckweed)	NR	Damage	14	NR	0.02	Adams and Schulz, 1987 in Campbell et al, 2000
Spirodela polyrhyza (Giant duckweed)	NR	NR	NR	35.3	0.00075	EPA, RED, 1995
Spirodela polyrhyza (Giant duckweed)	NR	Chlorosis	7		~0.005	Blackburn & Weldon, 1965
Spirodela polyrhyza (Giant duckweed)	NR	Damage	16	NR	0.01	Haller et al, 1988 in Campbell et al, 2000
Eichhornia crassipes (Waterhyacinth)	NR	NR	NR	35.3	0.114	EPA, RED, 1995

Table 13: Acute Toxicity of Diquat to Algae and Aquatic Macrophytes (continued)

Species	Test Media	End Point	Length of Test (days)	%A.I.	EC50 (ppm c.e)	Source
Eichhornia crassipes (Waterhyacinth)	NR	Damage	16	NR	0.1	Haller et al, 1988 in Campbell et al, 2000
Pistia stratiotes (Waterlettuce)	NR	Damage	16	NR	0.11	Haller et al, 1988 in Campbell et al, 2000
Hydrilla verticillata (Hydrilla)	NR	NR	NR	35.3	0.0099	EPA RED, 1995
Hydrilla verticillata (Hydrilla)	NR	Damage	16	NR	0.08	Haller et al, 1988 in Campbell et al, 2000
P*otamogeton natans* (American pondweed)	NR	Damage	14	NR	0.0072	Adams and Schulz, 1987 in Campbell et al, 2000
Elodea canadensis (American waterweed)	NR	Damage	14	NR	0.023	Adams and Schulz, 1987 in Campbell et al, 2000
Myriophyllum spicatum (Eurasian watermilfoil)	NR	Damage	14	NR	0.046	Adams and Schulz, 1987 in Campbell et al, 2000
Wolifiella floridana	NR	Chlorosis	7		~0.005	Blackburn & Weldon, 1965
Azolla caroliniana (Azolla)	NR	Chlorosis	7		~0.01	Blackburn & Weldon, 1965
Azolla caroliniana (Azolla)	NR	Damage	7	NR	0.02	Adams and Schulz, 1987 in Campbell et al, 2000
Azolla caroliniana (Azolla)	NR	Damage	16	NR	0.12	Haller et al, 1988 in Campbell et al, 2000
Azolla caroliniana (Azolla)	NR	NR	NR	35.3	0.0116	EPA RED, 1995

Table 13: Acute Toxicity of Diquat to Algae and Aquatic Macrophytes (continued)

Species	Test Media	End Point	Length of Test (days)	%A.I.	EC50 (ppm c.e)	Source
Wolffia columbiana (Watermeal)	NR	Chlorosis	7	NR	~0.1	Blackburn & Weldon, 1965
Microcystis aeruginosa (Unicellular Blue-green algae)	Peterson's Media	$^{14}CO_2$ uptake	0.9	NR	0.1	Peterson, 1997
Microcystis aeruginosa (Unicellular Blue-green algae)	Hoagland's Solution	Chlorophyll levels	3	NR	0.065	Phlips et al, 1992
Microcystis aeruginosa (Unicellular Blue-green algae)	NR	Growth rate	7	NR	0.093	Graham et al, 1987 in Campbell et al, 2000
Microcystis aeruginosa (Unicellular Blue-green algae)	Hoagland's Solution	Chlorophyll levels	7	NR	0.053	Phlips et al, 1992
Anabaena flos-aquae (Filamentous N-Fixing Blue-green algae)	Peterson's Media	$^{14}CO_2$ uptake	0.9	NR	0.089	Peterson, 1997
Anabaena flos-aquae (Filamentous N-Fixing Blue-green algae)	Hoagland's Solution	Cell density	3	NR	0.053	Phlips et al, 1992
Anabaena flos-aquae (Filamentous N-Fixing Blue-green algae)	Hoagland's Solution	Chlorophyll levels	3	NR	0.042	Phlips et al, 1992
Anabaena flos-aquae (Filamentous N-Fixing Blue-green algae)	NR	Growth rate	7	NR	0.05	Graham et al, 1987 in Campbell et al, 2000
Anabaena flos-aquae (Filamentous N-Fixing Blue-green algae)	Hoagland's Solution	Cell density	7	NR	0.138	Phlips et al, 1992

Table 13: Acute Toxicity of Diquat to Algae and Aquatic Macrophytes (continued)

Species	Test Media	End Point	Length of Test (days)	%A.I.	EC50 (ppm c.e)	Source
Anabaena flos-aquae (Filamentous N-Fixing Blue-green algae)	Hoagland's Solution	Cell density	7	NR	0.130	Phlips et al, 1992
Anabaena inaequalis (Filamentous N-Fixing Blue-green algae)	Peterson's Media	$^{14}CO_2$ uptake	0.9	NR	0.005	Peterson, 1997
Lyngbya wollei (Blue-green algae)	Hoagland's Solution	Chlorophyll levels	3	NR	0.145	Phlips et al, 1992
Lyngbya wollei (Blue-green algae)	Hoagland's Solution	Chlorophyll levels	7	NR	0.175	Phlips et al, 1992
Oscillatoria sp. (Filamentous blue-green algae)	Peterson's Media	$^{14}CO_2$ uptake	0.9	NR	0.082	Peterson, 1997
Pseudoanabaena sp. (Filamentous blue-green algae)	Peterson's Media	$^{14}CO_2$ uptake	0.9	NR	0.063	Peterson, 1997
Selenastrum capricornutum (Green algae)	Peterson's Media	$^{14}CO_2$ uptake	0.9	NR	0.492	Peterson et al, 1997
Selenastrum capricornutum (Green algae)	Hoagland's Solution	Cell density	3	NR	0.096	Phlips et al, 1992
Selenastrum capricornutum (Green algae)	Hoagland's Solution	Chlorophyll levels	3	NR	0.073	Phlips et al, 1992
Selenastrum capricornutum (Green algae)	NR	Growth rate	4	NR	0.019	Smyth & Tapp, 1988 in Campbell, 2000
Selenastrum capricornutum (Green algae)	NR	Growth of biomass	4	Technical (~100%)	0.08	Fairchild et al, 1997

Table 13: Acute Toxicity of Diquat to Algae and Aquatic Macrophytes (continued)

Species	Test Media	End Point	Length of Test (days)	%A.I.	EC50 (ppm c.e)	Source
Selenastrum capricornutum (Green algae)	NR	Growth rate	7	NR	0.48	Graham et al, 1987 in Campbell et al, 2000
Selenastrum capricornutum (Green algae)	Hoagland's Solution	Cell density	7	NR	0.047	Phlips et al, 1992
Selenastrum capricornutum (Green algae)	Hoagland's Solution	Chlorophyll levels	7	NR	0.026	Phlips et al, 1992
Scenedesmus quadricauda (Green algae)	Peterson's Media	$^{14}CO_2$ uptake	0.9	NR	0.652	Peterson et al, 1997
Chlorella vulgaris (Green algae)	Hoagland's Solution	Cell density	3	NR	>2.94	Phlips et al, 1992
Chlorella vulgaris (Green algae)	Hoagland's Solution	Chlorophyll levels	3	NR	>2.94	Phlips et al, 1992
Chlorella vulgaris (Green algae)	NR	Growth rate	7	NR	0.395	Graham et al, 1987 in Campbell et al, 2000
Chlorella vulgaris (Green algae)	Hoagland's Solution	Cell density	7	NR	0.307	Phlips et al, 1992
Chlorella vulgaris (Green algae)	Hoagland's Solution	Chlorophyll levels	7	NR	0.41	Phlips et al, 1992
Chlorococcum sp. (Marine green algae)	Artificial seawater	Growth	10	40	43	Walsh, , 1972
Dunaliella tertiolecta (Marine green algae)	Artificial seawater	Growth	10	40	6.5	Walsh, , 1972
Euglena gracilis (Euglenophyte)	Hoagland's Solution	Cell density	3	NR	>2.94	Phlips et al, 1992
Euglena gracilis (Euglenophyte)	Hoagland's Solution	Chlorophyll levels	3	NR	>2.94	Phlips et al, 1992

Table 13: Acute Toxicity of Diquat to Algae and Aquatic Macrophytes (continued)

Species	Test Media	End Point	Length of Test (days)	%A.I.	EC50 (ppm c.e)	Source
Euglena gracilis (Euglenophyte)	NR	Growth rate	7	NR	2.94	Graham et al, 1987 in Campbell et al, 2000
Euglena gracilis (Euglenophyte)	NR	Growth rate	7	NR	2.94	Graham et al, 1987 in Campbell et al, 2000
Euglena gracilis (Euglenophyte)	Hoagland's Solution	Cell density	7	NR	>2.94	Phlips et al, 1992
Euglena gracilis (Euglenophyte)	Hoagland's Solution	Chlorophyll levels	7	NR	>2.94	Phlips et al, 1992
Peridinium cinctum (Pyrrophyte)	NR	Growth rate	7	NR	0.001	Graham et al, 1987 in Campbell et al, 2000
Navicula pelliculosa (Chrysophyte = diatom)	Hoagland's Solution	Chlorophyll levels	3	NR	0.019	Phlips et al, 1992
Navicula pelliculosa (Chrysophyte = diatom)	NR	Growth rate	7	NR	0.065	Graham et al, 1987 in Campbell et al, 2000
Navicula pelliculosa (Chrysophyte = diatom)	Hoagland's Solution	Cell density	7	NR	0.127	Phlips et al, 1992
Navicula pelliculosa (Chrysophyte = diatom)	Hoagland's Solution	Chlorophyll levels	7	NR	0.106	Phlips et al, 1992
Cyclotella meneghiana (Chrysophyte = diatom)	Peterson's Media	$^{14}CO_2$ uptake	0.9	NR	0.092	Peterson et al, 1997
Nitzshcia sp. (Chrysophyte = diatom)	Peterson's Media	$^{14}CO_2$ uptake	0.9	NR	0.065	Peterson et al, 1997
Isochrysis galbana (Marine Chrysophyte = diatom)	Artificial seawater	Growth	10	40	3.2	Walsh, , 1972

Table 13: Acute Toxicity of Diquat to Algae and Aquatic Macrophytes (continued)

Species	Test Media	End Point	Length of Test (days)	%A.I.	EC50 (ppm c.e)	Source
Phaeodactylum tricornutum (Maine Chrysophyte = diatom)	Artificial seawater	Growth	10	40	3.2	Walsh, , 1972
Cryptomonas ovata (Chrysophyte = diatom)	NR	Growth rate	7	NR	0.046	Graham et al, 1987 in Campbell et al, 2000
Skeletonema costatum (marine diatom)	Enriched Artificial Seawater	Chlorophyll levels	3	NR	>2.94	Phlips et al, 1992
Skeletonema costatum (marine diatom)	Enriched Artificial Seawater	Chlorophyll levels	7	NR	>2.94	Phlips et al, 1992
Ochromonas danica (Chrysophyte = diatom)	Species Specific Media	Cell density	3	NR	0.136	Phlips et al, 1992
Ochromonas danica (Chrysophyte = diatom)	Species Specific Media	Chlorophyll levels	3	NR	0.023	Phlips et al, 1992
Ochromonas danica (Chrysophyte = diatom)	NR	Growth rate	7	NR	0.022	Graham et al, 1987 in Campbell et al, 2000
Ochromonas danica (Chrysophyte = diatom)	Species Specific Media	Cell density	7	NR	0.124	Phlips et al, 1992
Ochromonas danica (Chrysophyte = diatom)	Species Specific Media	Chlorophyll levels	7	NR	0.076	Phlips et al, 1992
Cryptomonas ozolini (Chryptophyceae)	Hoagland's Solution	Chlorophyll levels	3	NR	0.035	Phlips et al, 1992

Table 13: Acute Toxicity of Diquat to Algae and Aquatic Macrophytes (continued)

Species	Test Media	End Point	Length of Test (days)	%A.I.	EC50 (ppm c.e)	Source
Cryptomonas ozolini (Chryptophyceae)	Hoagland's Solution	Cell density	7	NR	0.075	Phlips et al, 1992
Cryptomonas ozolini (Chryptophyceae)	Hoagland's Solution	Chlorophyll levels	7	NR	0.019	Phlips et al, 1992

Table 14: Potential Hazard Ratios[1] for Eleven Species of Non-Target Aquatic Green Algae, Blue-Green Algae and Duckweed (Peterson et al, 1994)

Hazard Rating and Percent Inhibition of Growth when Exposed to an EEC of 0.75 ppm c.e. of Diquat	
	Very High (RQ >1.0; EEC Causes >50% Reduction in Growth)
Lemna minor (Duckweed)	100(0)[2]
Green Algae	
Cyclotella meneghiana	99(1)
Nitzschia sp.,F110-D	100(0)
Scenedesmus quadricauda, F11	53(13)
Selenastrum capricornutum, U1648	69(8)
Blue-Green Algae	
Microcystis aeruginosa, PCC7820	100(0)
Microcystis aeruginosa, U2063	100(0)
Oscillatoria sp.,T129	100(0)
Pseudoanabaena, F63	100(0)
Anabaena inaequalis, U381	100(0)
Aphanizomenon flos-aquae, F107	100(0)

[1] Peterson's Hazard Ratings: Very high if EEC causes >50% reduction in growth and RQ = >1.0; High if EEC causes 25 to 50% reduction in growth and RQ is much greater than 0.1 (0.5 to <1.0); Moderate if EEC causes 5 to 25% reduction in growth and RQ ≥0.1 (>0.1 to <0.5); Potentially low if EEC causes <5% reduction in growth and RQ ≤0.1.

[2] Values in parenthesis are standard deviations.

Table 15: Laboratory Effects of Diquat Formulations on Phytoplankton

Results	References[1]
<0.3 ppm c.e. inhibits the growth of blue-green algae species after three to seven days of exposure.	Birmingham and Colman, 1983
No metabolism of diquat by algae or other aquatic plants. However up to 32% of diquat may be metabolized by bacteria associated with dead and dying vegetation.	Simsiman and Chesters, 1976
20 ppm inhibits nitrogen fixation in blue-green algae. However, this concentration, which is higher than would be encountered in the field probably has no real environmental impact.	DaSilva et al, 1975
Unicellular green algae including *Oocystis*, *Characium*, *Ankistrodesmus*, *Cosmarium, Staurastrum*, *Chlorella pyrenoidosa*, *Chlorella ellipsoidea*, *Coccomyxa subellipsoidea* and *Stichocccus bacillaris* are not affected by diquat at concentrations up to 10 ppm.	Melendez et al, 1993
10 to 30 ppm stimulates growth of unicellular algae *Oocystis* spp. after 14 to 21 days of exposure.	Melendez et al, 1993
1 to 10 ppm causes the numbers of green filamentous algae to increase after 21 days of exposure.	Melendez et al, 1993
0.022 to >3.24 ppm c.e may inhibit or fail to inhibit the growth of diatoms after 3 to 10 days exposure.	Phlips, 1992; Birmingham and Colman, 1983 and Walsh, 1972
The pyrrophyte *Peridinium cinctum*, and the diatom *Ochromonas danica* are the most susceptible algal species with day-7 EC50 values of 0.001 to 0.022 ppm c.e., respectively. These species are likely to be impacted adversely since the predicted 4-day time-weighted average EEC in northwest waters after spot treatment with 0.30 to 0.37 ppm c.e is 0.021 ppm c.e. *Anabaena flos-aquae* (EC50 = 0.05 to 0.138 ppm c.e.) may also be affected by whole pond treatments with 0.224 ppm c.e. since the projected 4-day time-weighted average EEC for such a treatment may be as high as 0.059 ppm c.e.	Graham et al, 1987 in Campbell et al, 1987

Table 16: Field-Effects of Diquat Products on Phytoplankton

Results	References
0.25 to 0.5 ppm eradicated filamentous green algae *Mougeotia* and *Zygnema*.	Johnson, 1962
1.0 ppm kept unicellular green algae and filamentous green algae under control for only a brief period. Algal blooms occurred from 18 through 32 days and later in the year. These blooms provide food- stuffs for frogs (*Rana temporia*), toads (*Bufo bufo*) and the claDOeran (*Simocephalus vetulus*).	Cooke, 1977
0.25 to 1.0 ppm does not effect *Chara* spp.. Without effective competition *Chara* spp. May become the dominant species within the treated water body.	Johnson, 1962; Hulbert, 1987; Tatum and Blackburn, 1965
0.5 ppm decreases phyoplankton density for only three days. At eleven days there is a rebound effect with phytoplankton densities recovering to levels that are ~40% higher than before treatment.	Tatum and Blackburn, 1965.
Treatment with 0.5 ppm in a shallow New Zealand lake was followed by blooms of *Anabaena*, *Staurastrum* and green flagellates.	Fish, 1966 in Shearer and Halter, 1980)
0.5 to 5.0 ppm kills diatoms, blue-green algae and other filamentous algae.	Shealy, 1971 in Shearer and Halter, 1980
0.5 ppm diquat plus 0.5 ppm paraquat was not effective in reducing the standing crop of phytoplankton in a Columbia boat marina. However, population growth was stimulated within 3 weeks post-treatment.	Water investigation Branch, 1977 in Shearer and Halter, 1980
2.0 ppm was ineffective in controlling *Vaucheria dichotoma.*	Robson, 1976 in Shearer et al, 1980.

Table 17: Risk Assessment for Blue-green Algae, Green Algae, Diatoms and Macrophytes with Diquat Products

Species Name	**Test Chemicals – Diquat Products 7 to 16 day EC50 (ppm c.e.) & [Risk Quotient (RQ[1] = EEC[2]/EC50)] and {Risk Level }(Peterson et al, 1994)**					
	EEC = 0.75 (ppm)[3]	**EEC = 0.224 (ppm)[4]**	**EEC = 0.059 (ppm)[5]**	**EEC = 0.021 (ppm)[6]**	**EEC = 0.008 (ppm)[7]**	**EEC = 0.00028 (ppm)[8]**
Microcystis aeruginosa (Blue-green algae)	0.053 to 0.093 [14 to 8.1] {Very High}	0.053 to 0.093 [4.2 to 2.4] {Very High}[4]	0.053 to 0.093 [1.1 to 0.63] {Very High to High}	0.053 to 0.093 [0.40 to 0.23} {Moderate)	0.053 to 0.093 (0.15 to 0.086) {Moderate to Potentially Low}	0.053 to 0.093 [0.005 to 0.003] {Potentially Low}
Anabaena flos – aquae (Blue-green algae)	0.05 to 0.138 [15 to 5.4] {Very High}	0.05 to 0.138 [4.5 to 1.6] {Very High}	0.05 to 0.138 [1.18 to 0.42] {Very High to Moderate}	0.05 to 0.138 [0.42 to 0.15] {Moderate}	0.05 to 0.138 [0.16 to 0.057] {Moderate to Potentially Low}	0.05 to 0.138 [0.006 to 0.003] {Potentially Low}
Lyngbya wollei (Blue-green algae)	0.175 [4.28] {Very High}	0.175 [1.28] {Very High}	0.175 [0.34] {Moderate}	0.175 {0.12} {Moderate}	0.175 [0.045] {Potentially Low}	0.175 [0.002] {Potentially Low}
Selenastrum capricornutum (Green algae)	0.026 to 0.48 [29 to 1.56] {Very High}	0.026 to 0.48 [8.6 to 0.46] {Very High to Moderate}	0.026 to 0.48 [2.3 to 0.12] {Very high to Moderate}	0.026 to 0.48 [0.81 to 0.044] {High to Potentially Low}	0.026 to 0.48 [0.31 to 0.016] {Moderate to Potentially Low}	0.026 to 0.48 [0.011 to 0.0006] {Potentially Low}
Chlorella vulgaris (Green algae)	0.31 to 0.41 [2.4 to1.8] {Very High}	0.31 to 0.41 [0.72 to 0.54] {High to Moderate}	0.31 to 0.41 [0.19 to 0.14] {Moderate}	0.31 to 0.41 [0.067 to0.051] {Potentially Low}	0.31 to 0.41 [0.026 to 0.020] {Potentially Low}	0.31 to 0.41 [0.0009 to 0.0007] {Potentially Low}
Chlorococcum sp. (Marine green algae)	43 [0.017] {Potentially Low}	43 [0.0052] {Potentially Low}	43 [0.0014] {Potentially Low}	43 [0.0005] {Potentially Low}	43 [0.0002] {Potentially Low}	43 [<0.0001] {Potentially Low}
Dunaliella tertiolecta (Marine green algae)	6.5 [0.12] {Moderate}	6.5 [0.034] {Potentially Low}	6.5 [0.009] {Potentially Low}	6.5 [0.003] {Potentially Low}	6.5 [0.001] {Potentially Low}	6.5 [<0.0001] {Potentially Low}

Table 17: Risk Assessment for Blue-green Algae, Green Algae, Diatoms and Macrophytes with Diquat Products (continued)

Species Name	Test Chemicals – Diquat Products 7 to 16 day EC50 (ppm c.e.) & [Risk Quotient (RQ[1] = EEC[2]/EC50)] and {Risk Level }(Peterson et al, 1994)					
	EEC = 0.75 (ppm)[3]	**EEC = 0.224 (ppm)[4]**	**EEC = 0.059 (ppm)[5]**	**EEC = 0.021 (ppm)[6]**	**EEC = 0.008 (ppm)[7]**	**EEC = 0.00028 (ppm)[8]**
Navicula pelliculosa (Diatom)	0.065 to 0.127 [12 to 5.9] {Very High}	0.065 to 0.127 [3.4 to 1.8] {Very High}	0.065 to 0.127 [0.91 to 0.46] {High to Moderate}	0.065 to 0.127 [0.32 to 0.17] {Moderate}	0.065 to 0.127 [0.12 to 0.063] {Moderate to Potentially Low}	0.065 to 0.127 [0.004 to 0.002] {Potentially Low}
Cryptomonas ovata (Diatom)	0.046 [16] {Very High)	0.046 [4.9] {Very High)	0.046 [1.28] {Very High}	0.046 [0.46] {Moderate}	0.046 [0.17] {Moderate}	0.046 [0.006] {Potentially Low}
Cryptomonas ozolini (Diatom)	0.019 to 0.075 [39 to 10] { Very High}	0.019 to 0.075 [12 to 3.0] { Very High}	0.019 to 0.075 [3.1 to 0.78] { Very High to High}	0.019 to 0.075 [1.1 to 0.28] { Very High to Moderate}	0.019 to 0.075 [0.42 to 0.10] { Moderate}	0.019 to 0.075 [0.015 to 0.004] { Potentially Low}
Ochramonas danica (Diatom)	0.022 to 0.124 [27 to 6.0] {Very High}	0.022 to 0.124 [10 to 1.8] {Very High}	0.022 to 0.124 [2.7 to 0.47] {Very High to Moderate}	0.022 to 0.124 [0.95 to 0.17] {High to Moderate}	0.022 to 0.124 [0.36 to 0.036] {Moderate to Potentially Low}	0.022 to 0.124 [0.017 to 0.002] {Potentially Low}
Phaeodactylum tricornutum (Marine diatom)	3.2 [0.23] {Moderate}	3.2 [0.070] {Potentially Low}	3.2 [0.018] {Potentially Low}	3.2 [0.0.007] {Potentially Low}	3.2 [0.003] {Potentially Low}	3.2 [<0.001] {Potentially Low}
Isochrysis galbana (Marine diatom)	3.2 [0.23] {Moderate}	3.2 [0.070] {Potentially Low}	3.2 [0.018] {Potentially Low}	3.2 [0.0.007] {Potentially Low}	3.2 [0.003] {Potentially Low}	3.2 [<0.001] {Potentially Low}
Skeletonema costatum (Marine diatom)	>2.94 [<0.26] {Moderate to Potentially Low}	>2.94 [<0.076] {Potentially Low}	>2.94 [<0.020] {Potentially Low}	>2.94 [<0.007] {Potentially Low}	>2.94 [<0.003] {Potentially Low}	>2.94 [<0.001] {Potentially Low}

Table 17: Risk Assessment for Blue-green Algae, Green Algae, Diatoms and Macrophytes with Diquat Products (continued)

Species Name	**Test Chemicals – Diquat Products 7 to 16 day EC50 (ppm c.e.) & [Risk Quotient (RQ[1] = EEC[2]/EC50)] and {Risk Level }(Peterson et al, 1994)**					
	EEC = 0.75 (ppm)[3]	**EEC = 0.224 (ppm)[4]**	**EEC = 0.059 (ppm)[5]**	**EEC = 0.021 (ppm)[6]**	**EEC = 0.008 (ppm)[7]**	**EEC = 0.00028 (ppm)[8]**
Peridinium cinctum (Pyrrophyte)	0.001 [750] {Very High}	0.001 [224] {Very High}	0.001 [59] {Very High}	0.001 [21] {Very High}	0.001 [8.0] {Very High}	0.001 [0.28] {Moderate}
Euglena gracilis (Euglenophyte)	>2.94 [<0.25] {Moderate to Potentially Low)	>2.94 [<0.076] {Potentially Low}	>2.94 [<0.020] {Potentially Low}	>2.94 [<0.007] {Potentially Low}	>2.94 [<0.003] {Potentially Low}	>2.94 [<0.00001] {Potentially Low}
Lemna minor (Common duckweed)	0.0014 to 0.020 [535 to 38] {Very High)	0.0014 to 0.020 [160 to 11] {Very High)	0.0014 to 0.020 [42 to 3.0] {Very High)	0.0014 to 0.020 [15 to 1.1] {Very High}	0.0014 to 0.020 [5.7 to 0.4] {Very High to Moderate}	0.0014 to 0.020 [0.2 to 0.014] {Moderate to Potentially Low}
Spirodela polyrhyza (Giant duckweed)	~0.005 to 0.01 [~150 to 75] {Very High}	~0.005 to 0.01 [~45 to 22] {Very High}	~0.005 to 0.01 [~12 to 5.9] {Very High}	~0.005 to 0.01 [4.2 to 2.1] {Very High}	~0.005 to 0.01 [1.6 to 0.8] {Very High to High}	~0.005 to 0.01 [0.056 to 0.028] {Potentially Low}
Eichhornia crassipes (Waterhyacinth)	0.1 [7.5] {Very High}	0.1 [2.24] {Very High}	0.1 [0.59] {High}	0.1 [0.21] {Moderate}	0.1 [0.08] {Potentially Low}	0.1 [0.0028] {Potentially Low}
Pistia stratioites (Waterlettuce)	0.11 [6.8] {Very High}	0.11 [2.0] {Very High}	0.11 [0.53] {High}	0.11 [0.19] {Moderate}	0.11 [0.072] {Potentially Low}	0.11 [0.0025] {Potentially Low}
Hydrilla verticillata (Hydrilla)	0.08 [9.4] {Very High}	0.08 [2.8] {Very High}	0.08 [0.73] {High}	0.08 [0.26] {Moderate}	0.08 [0.1] {Potentially Low}	0.08 [0.0035] {Potentially Low)

Table 17: Risk Assessment for Blue-green Algae, Green Algae, Diatoms and Macrophytes with Diquat Products (continued)

Species Name	**Test Chemicals – Diquat Products 7 to 14 day EC50 (ppm c.e.) & [Risk Quotient (RQ[1] = EEC[2]/EC50)] and {Risk Level }(Petersen et al, 1994)**					
	EEC = 0.75 (ppm)[3]	**EEC = 0.224 (ppm)[4]**	**EEC = 0.059 (ppm)[5]**	**EEC = 0.021 (ppm)[6]**	**EEC = 0.008 (ppm)[7]**	**EEC = 0.00028 (ppm)[8]**
Potamogeton natans (Floating leaf pondweed)	0.0072 [104] {Very High}	0.0072 [31] {Very High}	0.0072 [8.1] {Very High}	0.0072 [2.9] {Very High}	0.0072 [1.1] {Very High}	0.0072 [0.038] {Potentially Low}
Elodea canadensis (American waterweed)	0.023 [33] {Very High}	0.023 [9.7] {Very High}	0.023 [2.6] {Very High}	0.023 [0.91] {High}	0.023 [0.35] {Moderate}	0.023 [0.012] {Potentially Low}
Myriophyllum spicatum (Eurasian watermilfoil)	0.046 [16] {Very High}	0.046 [4.9] {Very High}	0.046 [1.3] {Very High}	0.046 [0.46] {Moderate}	0.046 [0.17] {Moderate}	0.046 [0.006] {Potentially Low}
Azolla caroliniana (Azolla)	~0.01 to 0.12 [~75 to 6.25] {Very High}	~0.01 to 0.12 [~22 to 1.9] {Very High}	~0.01 to 0.12 [~5.9 to 0.49] {Very High to Moderate}	~0.01 to 0.12 [~2.1 to 0.18] {Very High to Moderate}	~0.01 to 0.12 [~0.80 to 0.067] {High to Potentially Low}	~0.01 to 0.12 [~0.028 to 0.002] {Potentially Low}
Wolffia columbiana (Watermeal)	~0.1 [~7.5] {Very High}	~0.1 [~2.2] {Very High}	~0.1 [~0.59] {High}	~0.1 [~0.21] {Moderate}	~0.1 [~0.080] {Potentially Low}	~0.1 [~0.0028] {Potentially Low}

[1] RQ = Risk Quotient (unitless)
[2] EEC = Expected Environmental Concentration
[3] Highest EEC immediately after treatment with highest labeled use rate = 0.75 ppm c.e.
[4] Highest EEC immediately after treatment according to EPA's worst case scenario is 0.224 ppm c.e.
[5] Typical four-day time-weighted average EEC is 0.059 ppm c.e.
[6] Highest four-day time-weighted average EEC after spot treatment with diquat at 0.30 to 0.37 ppm is 0.021 ppm c.e.
[7] Highest four- day time-weighted average EEC adjacent to a spot treatment site is 0.008 ppm c.e.
[8] Highest four-day time-weighted average EEC 375 meters from the spot treatment site = 0.000228 ppm c.e.

Table 18: Acute Toxicity of Diquat Products to Fish

Species	Size/Age Class	Hardness (ppm)	1D LC50 (ppm c.e.)	2D LC50 (ppm c.e.)	3D LC50 (ppm c.e.)	4D LC50 (ppm c.e.)	Source
Anguilla rostra (American eel)	Glass eel or Black eel stage	44	NA	NA	NA	17-29.7 (22)	Hinton & Eversole, 1979
Brachdanio rario (zebra fish)	NR[1]	NR	30	24	19	13	Benijt-Claus & Pesoone, 1975
Brachdanio rario (zebra fish)	NR	170 (Hard)	>100	NA	NA	NA	Devillers et al, 1985 in Hammer, 1994
Carrasius auratus (Goldfish)	NR	50 & NR (Inter)	>19.1	NA	NA	35-85 (55)	Gilderhaus, 1967; Berry, 1984; FWS, 1986 in Brian, 1999
Cirrhina mrigala Hamilton's carp)	40-50mm	NR	NA	NA	NA	75	Singh & Yadav, 1978
Ctenopharyngodon idella (Grass carp)	105mm,23.5g	21 (Very Soft)	NA	NA	NA	28.62	El-Deen & Rogers, 1992
Ctenophryngodon idella (Grass carp)	95mm, 15.8g	270	434	153	NA	125	Tooby et al, 1980
Cyprinodon variegatus (Sheepshead minnow)	25mm, 0.25g	Saltwater	>82	>82	>57	48	Nicholson (SBI) 1987 in Hammer, 1994 & in Brian, 1999
Cyprinus carpio (Common carp)	Fry (9 weeks,0.4227g)	22 (Very Soft)	51			27	Chin & Sudderuddin, 1979 (Probably ddb)
Cyprinus carpio (Common carp)	Fingerling (52mm,4.4g)	48 (Soft)	285	143	91	67	Tapp, 1988

Table 18: Acute Toxicity of Diquat Products to Fish (continued)

Species	Size/Age Class	Hardness (ppm)	1D LC50 (ppm c.e.)	2D LC50 (ppm c.e.)	3D LC50 (ppm c.e.)	4D LC50 (ppm c.e.)	Source
Cyprinus carpio (Common carp)	1-2 year/yearling	NR	NA	NA	NA	141	Silvo, 1967 in Hammer, 1994
Cyprinus carpio (Common carp)	Mean/fingerling	NR	17	6.2	NA	3.4-35 (11)	Skea et al 1988 in Campbell et al , 2000; Tapp, 1987
Erimyzon sucetta (Lake chubsucker)	Sac-fry	NR	NA	1.3-2.5 (1.8)	NA	NA	Hiltibran, 1967
Esox lusius (Northern pike)	NR	NR	NA	NA	NA	16	Gilderhaus, 1967
Esox masquinongy (Muskellunge)	Mean/(sac-fry, & 57mm fingerlings)	NR	18-62 (33)	7.6-10 (8.7)	NA	4.6-5.8 (5.2)	Skea et al, 1987 in Hammer, 2994
Fundulus similis (Longnosed killifish)	NR	Saltwater	>1.0	>1.0	NA	NA	Butler, 1965 (c.e. based on input from Hammer, 1994)
Gambusia affinis (Mosquito fish)	NR (probably neonates)	NR	10	NA	NA	NA	Ahmed, 1967 in Campbell, 2000
Gambusia affinis (Mosquito fish)	31mm,0.32g/juveniles	15 (Very Soft)	390	NA	NA	156	Leung et al, 1983
Ictalurus melas (Black bullhead)	0.8g	NR	NA	NA	NA	4.6	EPA, 1995 & FWS, 1986 in Brian
Ictalurus melas (Black bullhead)	0.8g	45 (Soft)	NA	NA	NA	86.7	Johnson & Finley, 1980
Ictalurus punctatus (Channel catfish)	13mm sac-fry & 50mm fingerlings	NR	NA	NA	NA	>5.4->10.0 (7.3)	Jones, 1965; Lawrence et al, 1967

Table 18: Acute Toxicity of Diquat Products to Fish (continued)

Species	Size/Age Class	Hardness (ppm)	1D LC50 (ppm c.e.)	2D LC50 (ppm c.e.)	3D LC50 (ppm c.e.)	4D LC50 (ppm c.e.)	Source
Lepomis macrochirus (Bluegill sunfish)	Mean/fry & fingerlings	22-45 (Very Soft to Soft)	20-49 (31.3)	15-43 (25.4)	NA	14-39 (23.4)	Thompson et al, 1980; Surber & Pickering, 1962
Lepomis macrochirus (Bluegill sunfish)	Mean/fry & fingerlings	53-78 (Inter)	33-175 (76)	17-88 (38)	NA	12.1-13.5 (12.8)	McCann, 1967 in Hammer, 1994; Jones, 1965; McCann, 1967 in Campbell, 2000
Lepomis macrochirus (Bluegill sunfish)	(probably fry)	120 (Inter)	NA	NA	NA	9.7	Bishop & Perry, 1961
Lepomis macrochirus (Bluegill sunfish)	45mm/ fingerlings	340 (Hard)	221	113	NA	76	Surber & Pickering, 1962
Lepomis macrochirus (Bluegill sunfish)	Mean 1.1-1.3g/ subadult	NR	NA	NA	NA	76-125 (52)	EPA RED, 195; Johnson & Finley, 1980
Micropterus dolomieu (Smallmouth bass)	Mean/sac-fry & Fry	NR	0.35-2.5 (1.29)	NA	NA	1.3	Schenk, 1967 & Skea et al, 1987 in Hammer, 1995; Hiltibran, 1967
Micropterus dolomieu (Smallmouth bass)	6-8day/fry	81 (Inter)	110	28	10	3.9	Paul et al, 1999
Micropterus salmoides (Largemouth bass)	Mean 16-18mm/sac-fry	72-78 (Inter)	3.6	1.9	NA	>0.27->1.62 (>0.62)	Jones, 1965;Skea & Shifley, 19?? In Hammer, 1994
Micropterus salmoides (Largemouth bass)	9-13 day fry	46-81 (Soft to Inter)	15	11	8	4.9	Paul et al, 1999
Micropterus salmoides (Largemouth bass)	Mean 50-63mm/fry & fingerling	NR	13	5.9		4.2->10 (>6.4)	Surber & Pickering, 1962; Lawrence et al, 1965
Micropterus salmoides (Largemouth bass)	Mean 100mm/subadult	56 (Inter)	NA	548	178	60	Skea & Shiftlet, 19?? In Hammer, 1994

Table 18: Acute Toxicity of Diquat Products to Fish (continued)

Species	Size/Age Class	Hardness (ppm)	1D LC50 (ppm c.e.)	2D LC50 (ppm c.e.)	3D LC50 (ppm c.e.)	4D LC50 (ppm c.e.)	Source
Morone saxatilis (Striped bass)	1-week sac-fry	40 (Soft)	0.54	0.54	0.54	0.54	Hughes, 1969, Hughes, 1973
Morone saxatilis (Striped bass)	1-month Fingerling	40 (Soft)	19	13	8.1	5.4	Hughes, 1969, Hughes, 1973
Morone saxatilis (Striped bass)	60mm, 2.7g subadult	40 (Soft)	170	84	NA	43	Wellborn, 1969
Notoemigonus crysoleucans (Golden shiner)	74mm	64 (Inter)	NA	70	41	33	Skea & Shiflet, 19?? In Hammer, 1994
Notropis atherinoides (Emerald shiner)	Mean 55mm/ Fingerling, NR	NR	NA	NA	NA	13.9	Swabey & Schenk, 1963,
Oncorhynchus kisutch (Coho salmon)	Mean/Probably Fingerling	NR	NA	29	NA	20.5	Watkins, 1966 & Muirhead-Thompson, 1971 in Hammer, 1994
Oncorhynchus kisutch (Coho salmon)	Yearling	100 (Inter)	NA	NA	NA	30	Lorz et al, 1979
Oncorhynchus mykiss (Rainbow trout)	Mean/Probably Fingerling	NR	90	NA	16	8-11.2 (9.46)	EPA, 1995, &F WS, 1986 in Brian, 1999
Oncorhynchus mykiss (Rainbow trout)	Mean/Fingerlings	45-52 (Soft to Inter)	31-103 (60)	27-53 (33)	89	6.1-26 (15)	Tapp & Caunter, 1989; Thomson et al, 1980; Tapp & Caunter, 1989; Hill et al, 1976
Oncorhynchus mykiss (Rainbow trout)	Mean/Fingerlings	250 (Hard)	48.6	37.8	NA	NA	Alabaster, 1969

Table 18: Acute Toxicity of Diquat Products to Fish (continued)

Species	Size/Age Class	Hardness (ppm)	1D LC50 (ppm c.e.)	2D LC50 (ppm c.e.)	3D LC50 (ppm c.e.)	4D LC50 (ppm c.e.)	Source
Oncorhynchus tshawytscha (Chinook salmon)	58-96mm	NR	16	16	NA	NA	Bond et al, 1960
Perca flavecens (Yellow perch)	0.6g	NR	NA	NA	NA	4.4	EPA, 1995 &FWS, 1986 in Brian, 1999
Perca flavecens (Yellow perch)	0.6g	45 (Soft)	NA	NA	NA	30.6	Johnson & Finley, 1980
Perca flavecens (Yellow perch)	NR	NR	NA	NA	NA	60	Holtz & Winter, 1989
Pimephales promelas (Fathead minnow)	Mean 45-48 mm/ Fingerlings	22 (Very Soft)	>30-76 (>48)	12.0-14.0 (13)	NA	7.6	Surber & Pickering, 1962
Pimephales promelas (Fathead minnow)	Mean/Probably Fingerlings	NR	NA	24	NA	>10-24.0 (>15)	Lawrence et al, 1965; Hong et al, 1988
Pimephales promelas (Fathead minnow)	47mm	379 (Hard)	140	119	NA	70	Surber & Pickering, 1962
Poecilla reticulata (Guppy)	10mm	NR	180	50	NA	4.4 (7.7 days)	Van Dord et al, 1974 in Hammer, 1994
Poecilla reticulata (Guppy)	25mm	NR	NA	NA	NA	50	Lee & Furtado, 1977
Poecilla mexicana (Mexican molly)	NR	NR	16.8	NA	NA	9.0 (5.25 days)	Moore, R.J., 1978
Rasbora heteromorpha (Harlequin fish)	Mean 20 mm/ Juveniles	20 (Soft)	39-41 (40)	20-16 (18)	NA	NA	Alabaster, 1969

Table 18: Acute Toxicity of Diquat Products to Fish (continued)

Species	Size/Age Class	Hardness (ppm)	1D LC50 (ppm c.e.)	2D LC50 (ppm c.e.)	3D LC50 (ppm c.e.)	4D LC50 (ppm c.e.)	Source
Rasbora spp. (Scissor-tail)	30mm	NR	NA	NA	NA	30	Lee & Furtadao, 1977
Salmo trutta (Brown trout)	Mean/Probably Fingerlings NR	NR	33	22	NA	17.8-20 (19)	Simonin & Skea, 1977 in Campbell et al, 2000
Salmo trutta (Brown trout)	Mean/Yearling	250 (Hard)	NA	64-123 (89)	NA	NA	Woodiwiss et al, 1974
Salmonid (Trout)	NR	NR	20	NA	NA	NA	Holden, 1964 in Hammer, 2000
Salvelinis fontinalis (Eastern brook trout)	NR	NR	>11	NA	NA	NA	Eady & Reanney, 1965
Stizostedion vitreu (Walleye)	8-10 day fry	132 (Inter)	2.9	1.6	1	0.75	Paul et al, 1999
Stizostedion vitreu (Walleye)	41-43 day fry	132 (Inter)	3.1	1.9	1.6	1.5	Paul et al, 1999
Stizostedion vitreu (Walleye)	84-46 day fry	132 (Inter)	7.8	4.9	4.9	4.9	Paul et al, 1999

[1] NR = Parameter not reported.

[2] Values in parenthesis are geometric means of all acceptable values

NA = Not available

Table 19: Acute Toxicity of Diquat Products to Invertebrates

Species	Size/Age Class	Water Hardness (ppm)	1D LC50 (ppm c.e)	2D LC50 (ppm c.e.)	4D LC50 (ppm c.e.)	Source
Alonella spp. (Water flea)	NR[1]	27 (Soft)	NA	>5.4	NA	Naqvi & Hawkins, 1989
Asellus communis (Isopod)	NR	NR (Sediment)	NA	NA	>10 (120 hrs)	Williams et, al, 1984
Callibaetis spp. (Mayfly)	NR	64 (Inter)	>100	37	16.4	Wilson & Bond, 1969; Wilson, 1967
Cardium edule (Cockle)	NR	NR	NA	>5.4	NA	Portman, 1972
Caridina nilotica (Freshwater shrimp)	Adult	12 (Very Soft)	109	NA	NA	Kevan & Pearson, 1993
Cragnon cragnon (Shrimp)	NR	NR	NA	>5.4	NA	Portman, 1972
Crassostrea virginica (Eastern oyster)	Spat	Saltwater	NA	NA	55	Dionne, 1987
Crassostrea virginica (Eastern oyster)	Shell Deposition	Saltwater	NA	NA	141	Dionne, 1987
Cypria spp. (Ostracod)	NR	27 (Very Soft)	NA	7.452	NA	Naqvi & Hawkins, 1989
Daphnia magna (Water flea)	NR	124 (Intermediate)	NA	1.62	NA	Bishop & Perry, 1961
Daphnia magna (Water flea)	Mean/1st Instar	NR	1.3-3.8 (2.2)	0.32-1.17 (0.79)	NA	Crosby & Tucker, 1966; EPA RED, 1995; Benijt-Clause Persoone

Table 19: Acute Toxicity of Diquat Products to Invertebrates (continued)

Species	Size/Age Class	Water Hardness (ppm)	1D LC50 (ppm c.e)	2D LC50 (ppm c.e.)	4D LC50 (ppm c.e.)	Source
Daphnia magna (Water flea)	NR	40 (Soft)	2.5	1.2	NA	Wheeler, 1978 & CCC, 1978 in Brian, 1999
Daphnia magna (Water flea)	Adult	NR	3.7	1.6	NA	Benijts-Claus & Persoone, 1975
Daphnia pulex (Water flea)	1st Instar	NR	0.81	0.162	NA	Benijts & Persoone, 1975
Daphnia pulex (Water flea)	Adult	NR	0.702	0.216	NA	Benijts & Persoone, 1975
Daphnia similis (Water flea)	NR	NR	4.3	NA	NA	Tadokoro, 1986 in Hammer, 1994
Diapotomus spp. (Copepod)	NR	27 (Very Soft)	NA	~5.4	NA	Naqvi & Hawkins, 1989
Enallagma spp. (Damselfly)	Larvae	64 Inter	>100	>100	>100	Wilson & Bond, 1969, Wilson, 1967
Eucyclops spp. (Copepod)	NR	27 (Very Soft)	NA	25.164	NA	Naqvi & Hawkins, 1989
Gammarus fasciatus (Scud)	NR	NR	NA	NA	18.7	EPA, 1995 & Campbell et al, 2000
Hyalella azteca (Amphipod)	Mean 4-8mm/NR	64 & NR (Inter)	0.58-0.67 (0.62)	0.12-0.64 (0.22)	0.048	Wilson & Bond, 1969; Wilson, 1967; Nicholson & Clerman, 1974; EPA RED, 1995 Williams et al, 1984 in Campbell et al 2000

Table 19: Acute Toxicity of Diquat Products to Invertebrates (continued)

Species	Size/Age Class	Water Hardness (ppm)	1D LC50 (ppm c.e)	2D LC50 (ppm c.e.)	4D LC50 (ppm c.e.)	Source
Hyalella azteca (Amphipod)	4-8mm/NR	64 & NR (Sediment) (Inter)	NA	3.4	6.8	Wilson, 1967; Williams et al, 1984
Libellula spp. (Dragonfly)	NR	64 (Inter)	>100	>100	>100	Wilson & Bond, 1969, Wilson, 1967
Limnea stagnalis (Pond snail)	NR	NR	NA	NA	1.0 (8 days)	Carter, 1971 in Hammer, 1994
Limnephilus spp. (Caddisfly)	NR	64 (Inter)	>100	65	33	Wilson & Bond, 1969, Wilson, 1967
Mysidopsis bahia (Pocket shrimp)	Juvenile	Saltwater	>1.2	0.87	0.42	EPA RED, 1995
Nymphula responsalis (Aquatic moth)	Larvae	NR	NA	NA	86.94	Lee & Furtado, 1977
Paneus duroarum (Pink shrimp)	Juvenile	Saltwater	NA	8.48	NA	EPA, 1986 in Brian, 1999
Paneus setriferus (White shrimp)	NR	Saltwater	>1.0	>1.0	NA	Butler, 1965
Pomacea pludosa (Apple snail)	7-days to adults	45 (sediment) (Soft)	NA	NA	0.34	EPA, RED 1995, Winger et al, 1977
Streptocephalus seali (Fairy shrimp)	Mature	120 (Inter)	>1.08	>2.0	NA	Moss, 1978
Tendipedinae (Bloodworm)	Larvae	64 (Inter)	>100	>100	>100	Wilson & Bond, 1969, Wilson, 1967

[1] NR = Parameter not reported.

[2] Values in parenthesis are geometric means of all acceptable values

NA = Not available

Table 20: Chronic Toxicity of Diquat Products to Fish

Species	Size Class/ Age	End -Point	Time (Days)	NOEC (ppm c.e.)	MATC (ppm c.e.)	LOEC (ppm c.e.)	LC50 (ppm c.e.)	Reference
Lepomis macrochirus (Bluegill sunfish)[2,4]	Sac-Fry	Survival	12	>10	>10	>10	>10	Hiltibran,1967
Salvelinis fontanalis (Eastern brook trout)[2,]	Eggs to sac-fry	Abnormal sac-fry	NR[1]	2.7[8]	3.8[8]	5.4[8]	>11[8]	Eadey and Reaney,1965
Ictalurus puntatus (Channel catfish)[2,4]	Egg to fry	Weight reduction	NR[1]	>1.0	>1.0	>1.0	NR	EPA RED, 1995
Pimephales promelas (Fathead minnow)[2,5]	Egg to fry	Weight reduction	34	0.12	0.2	0.32	>0.65	Suprenant. 1987
Oncorhynchus mykiss (rainbow trout)[2,6]	48.6mm, 1.89g/ fingerlings	LC50	21	<1.4	<1.4	<1.4	2.9	Tapp & Caunter, 1989
Fundulus heteroclitus (Killifish)[3]	Egg to fry	Abnormal pigmentation and slow development	32	0.0054[8]	0.017[8]	0.054[8]	<0.0054[8]	Crawford & Guarino, 1985
Fundulus heteroclitus (Killifish)[3]	Egg to fry	Abnormal pigmentation and slow development	32	<0.0054[8]	<0.0054[8]	<0.0054[8]	<0.0054[8]	Crawford & Guarino, 1985
Rana pipiens (Leopard frog)[1,2,4]	Egg to 14 day post- hatch	Developmental abnormality & Survival	21	<54[8]	<54[8]	<54[8]	<54[8]	Bimber & Mitchell, 1977
Rana pipiens (Leopard frog)[1,2,7]	Embryos	Development	13	1.08[8]	1.7[8]	2.7[8]	>5.4[8]	Dial & Dial, 1987
Rana pipiens (Leopard frog)[1,2,7]	15-day/tadpole	Survival	13	>5.4[8]	>5.4[8]	>5.4[8]	>5.4[8]	Dial & Dial, 1987
Xenopus laevis (African clawed toad)[1,2,4]	Tailbud stage to tadpoles	Body size, Reduced pigmentation, Altered Shape, Muscular Development, Survival	10	0.54[8]	0.64[8]	0.68[8]	~0.41[8]	Anderson and Prahlad, 1976
Xenopus laevis (African clawed toad)[1,24]	Tailbud stage to tadpoles	Survival	10	<0.27[8]	<0.27[8]	<0.27[8]	~0.41[8]	Anderson and Prahlad, 1976

Table 20: Chronic Toxicity of Diquat Products to Fish (continued)

[1] NR= Not reported for this parameter.
[2] Freshwater organism.
[3] Saltwater organism. Tropical species not relevant to northwest waters.
[4] Water hardness not reported.
[5] Test conducted in soft water (<50 ppm $CaCO_3$).
[6] Test conducted in intermediately hard water (50 to 150 ppm $CaCO_3$).
[7] Test conducted in hard water (>150 ppm $CaCO_3$).
[8] Values reported here are in units of ppm c.e. The values that were originally reported appear to be reported in ppm diquat dibromide (ddb) equivalents; but it was not entirely clear what the original units were. ppm c.e. = ppm ddb x 0.54 c.e./ddb.

NOEC = Statistical No Observed Effect Concentration.
MATC = Maximum Acceptable Toxic Concentration = geometric meant of NOEC and LOEC.
LOEC = Statistical Lowest Observed Effect Concentration.

Table 21: Chronic Toxicity of Diquat Products to Invertebrates (Daphnid)

Species	Type	Water	Age	Time (days)	NOEC[1] (ppm c.e.)	MATC[2] (ppm c.e.)	LOEC[3] (ppm c.e)	Reference
Daphnia magna (Daphnid)	Flow-through	FW[4,5]	Life-cycle (Reproduction and Growth)	21	0.036	0.045	0.057	Suprenant, 1987

[1] NOEC = Statistical No Observed Effect Concentration.
[2] MATC = Maximum Acceptable Toxic Concentration = geometric meant of NOEC and LOEC.
[3] LOEC = Statistical Lowest Observed Effect Concentration.
[4] FW = Freshwater.
[5] Daphnia require hard ward for best development.

Table 22: Effects of Diquat on Survival and Growth of Bluegill sunfish in Experimental Fish Ponds in Wisconsin

	Fish Species						
Diquat Treatment (ppm)	**Fingerlings**			**Adults**			
	% Surviving	Average Length Increase (inches)	Average Weight Increase (grams)	% Surviving	Average Length Increase (inches)	Average Weight Increase (grams)	Total Weight Harvested (grams)
Control	70.0	1.44	4.69	55.3	0.41	6.35	1223
1.0 once	74.0	1.38	4.32	73.8	0.27	4.60	1435
1.0 twice[1]	79.3	1.41	4.14	61.4	0.35	5.22	1282
1.0 three times[1]	80.0	1.42	4.27	91.4	0.15	2.34	1575
3.0 once	78.2	1.52	4.98	90.7	0.25	3.30	1662
3.0 twice[1]	76.0	1.53	5.0	74.2	0.37	4.95	1518

[1] When more than one treatment occurs, time between treatments was 8-weeks

Table 23: Acute and Chronic Risk Assessment for Diquat

Test Type	Exposure Time	Species	EEC[1] (ppm c.e.)	Acute LC50[2] (ppm c.e.)	MATC[3] (ppm c.e.)	Risk Quotient	Level of Concern	RQ exceeds LOC[4]
Acute	4	*Morone saxatilis* (Striped bass sac-fry)	0.059	0.54	NA	0.11	0.1/0.5	Possible Restricted Use
Acute	3	*Morone saxatilis* (Striped bass sac-fry)	0.076	0.54	NA	0.14	0.1/0.5	Possible Restricted Use
Acute	2	*Morone saxatilis* (Striped bass sac-fry)	0.102	0.54	NA	0.19	0.1/0.5	Yes/No Restricted Use
Acute	1	*Morone saxatilis* (Striped bass sac-fry)	0.146	0.54	NA	0.27	0.1/0.5	Yes/No Restricted Use
Acute	4	*Micropterus salmodies* (Largemouth bass fry)	0.059	0.62	NA	0.095	0.1/0.5	No/No
Acute	4	*Stizostedion vitreu* (walleye 8 to 10 day fry*)*	0.059	0.75	NA	0.079	0.1/0.5	No/No
Acute	2	*Fundulus similis* (Longnosed killifish)	0.102	>1.0	NA	<0.102	0.1/0.5	Probably not/ Possible Restricted Use
Acute	4	*Micropterus dolomieu* (smallmouth bass sac-fry)	0.059	1.3	NA	0.045	0.1/0.5	No/No
Acute	1	*Micropterus dolomieu* (smallmouth bass sac-fry)	0.146	1.29	NA	0.11	0.1/0.5	Probably not/ Possible Restricted Use
Acute	2	*Stizostedion vitreu* (Walleye 41 to 43-day fry*)*	0.059	1.5	NA	0.039	0.1/0.5	No/No
Acute	4	*Erimyzon sucetta* (Chubsucker sac-fry)	0.059	1.8	NA	0.033	0.1/0.5	No/No
Acute	4	*Micropterus dolomieu* (SMB 6 to 8-day fry)	0.059	3.9	NA	0.015	0.1/0.5	No/No
Acute	4	*Perca flavescens* (Perch 0.6 g fry)	0.059	4.4	NA	0.013	0.1/0.5	No/No

Table 23: Acute and Chronic Risk Assessment for Diquat (continued)

Test Type	Exposure Time	Species	EEC[1] (ppm c.e.)	Acute LC50[2] (ppm c.e.)	MATC[3] (ppm c.e.)	Risk Quotient	Level of Concern	RQ exceeds LOC[4]
Acute	4	*Pimephales promelas* (Fathead minnow 45 to 48 mm fry)	0.059	7.6	NA	0.0077	0.1/0.5	No/No
Chronic	4 Acute 28 Chr	**Morone saxatilis** (Striped bass sac-fry)	0.0087	0.54	0.014	0.62	1.0	No
Chronic	3 Acute 28 Chr	*Morone saxatilis* (Striped bass sac-fry)	0.0087	0.54	0.014	0.62	1.0	No
Chronic	2 Acute 28 Chr	*Morone saxatilis* (Striped bass sac-fry)	0.0087	0.54	0.014	0.62	1.0	No
Chronic	1 Acute 28 Chr	*Morone saxatilis* (Striped bass sac-fry)	0.0087	0.54	0.014	0.62	1.0	No
Chronic	4 Acute 28 Chr	*Micropterus salmoides* (Largemouth bass fry)	0.0087	0.62	0.016	0.54	1.0	No
Chronic	4 Acute 28 Chr	*Stizostedion vitreu* (Walleye 8 to 10 day fry*)*	0.0087	0.75	0.020	0.44	1.0	No
Chronic	2 Acute 28 Chr	*Fundulus similis* (Longnosed killifish)	0.0087	>1.0	>0.026	<0.33	1.0	No
Chronic	4 Acute 28 Chr	*Micropterus dolomieu* (Smallmouth bass sac-fry)	0.0087	1.3	0.034	0.045	1.0	No
Chronic	1 Acute 28 Chr	*Micropterus dolomieu* (Smallmouth bass sac-fry)	0.0087	1.29	0.0034	0.0.045	1.0	No
Chronic	2 Acute 28 Chr	*Stizostedion vitreu* (Walleye 41 to 43-day fry)	0.0087	1.5	0.039	0.22	1.0	No
Chronic	4 Acute 28 Chr	*Erimyzon sucetta* (Chubsucker sac-fry)	0.0087	1.8	0.047	0.19	1.0	No
Chronic	4 Acute 28 Chr	**Micropterus dolmen** (SMB 6 to 8-day fry*)*	0.0087	3.9	0.103	0.084	1.0	No
Chronic	4 Acute 28 Chr	*Perca flavescens* (Perch 0.6 g fry)	0.0087	4.4	0.120	0.073	1.0	No
Chronic	4 Acute 28 Chr	*Pimephales promelas* (egg to fry)	0.0087	7.6	0.2	0.044	1.0	No

Table 23: Acute and Chronic Risk Assessment for Diquat (continued)

Test Type	Exposure Time	Species	EEC[1] (ppm c.e.)	Acute LC50[2] (ppm c.e.)	MATC[3] (ppm c.e.)	Risk Quotient	Level of Concern	RQ exceeds LOC[4]
Invertebrates								
Acute	4	*Hyalella azteca* 4-8mm	0.059	0.048	NA	1.2	0.1/0.5	Yes/Yes (Adverse Impact)
Acute	2	*Hyalella azteca* 4-8mm	0.102	0.22	NA	0.46	0.1/0.5	Yes/No (Restricted Use)
Acute	1	*Hyalella azteca* 4-8mm	0.146	0.62	NA	0.24	0.1/0.5	Yes/No (Restricted Use)
Acute	2	*Daphnia pulex* (1st Instar)	0.102	0.162	NA	0.62	0.1/0.5	Yes/Yes (Adverse Impact)
Acute	1	*Daphnia pulex* (Adults)	0.146	0.216	NA	0.68	0.1/0.5	Yes/Yes (Adverse Impact)
Acute	4	*Pomacea pludosa* (Apple snail 7-days)	0.059	0.34	NA	0.17	0.1/0.5	Yes/No (Restricted Use)
Acute	4	*Mysidopsis bahia* (Pocket shrimp juveniles)	0.059	0.42	NA	0.14	0.1/0.5	Probably not/ Possible Restricted Use
Acute	2	*Mysidopsis bahia* (Pocket shrimp juveniles)	0.102	0.87	NA	0.12	0.1/0.5	Probably not/ Possible Restricted Use
Acute	1	*Mysidopsis bahia* (Pocket shrimp juveniles)	0.146	>1.2	NA	<0.12	0.1/0.5	Probably not/ Possible Restricted Use
Acute	2	*Daphnia magna* (1st Instar)	0.102	0.79	NA	0.13	0.1/0.5	Probably not/ Possible Restricted Use
Acute	1	*Daphnia magna* (1st Instar)	0.146	1.2	NA	0.12	0.1/0.5	Probably not/ Possible Restricted Use
Acute	8	*Limnea stagnalis* (Pond snail)	<0.059	>1.0	NA	<0.059	0.1/0.5	No/No
Acute	2	*Paneus setrifus* (White Shrimp)	0.102	>1.0	NA	<0.059	0.1/0.5	No/No
Acute	2	*Daphnia magna* (Adults)	0.102	1.6		0.064	0.1/0.5	No/No
Chronic	4 Acute 21-28 Chr	*Hyalella azteca* 4-8mm	0.011 0.0087	0.048	0.0025	4.4 3.5	1.0	Yes (Adverse Impact)

Table 23: Acute and Chronic Risk Assessment for Diquat (continued)

Test Type	Exposure Time	Species	EEC[1] (ppm c.e.)	Acute LC50[2] (ppm c.e.)	MATC[3] (ppm c.e.)	Risk Quotient	Level of Concern	RQ exceeds LOC[4]
Chronic	2 Acute 21-28 Chr	*Hyalella azteca* 4-8mm	0.011 0.0087	0.22	0.0115	0.96 0.76	1.0	No
Chronic	1 Acute 21-28 Chr	*Hyalella azteca* 4-8mm	0.011 0.0087	0.62	0.0326	0.34 0.27	1.0	No
Chronic	2 Acute 21-28 Chr	**Daphnia pulex** (1st Instar)	0.011 0.0087	0.162	0.0085	1.3 1.0	1.0	Yes (Adverse Impact)
Chronic	1 Acute 21-28 Chr	**Daphnia pulex** (Adults)	0.011 0.0087	0.216	0.011	1.0 0.79	1.0	No
Chronic	4 Acute 21-28 Chr	**Pomacea pludosa** (Apple snail 7-days)	0.011 0.0087	0.34	0.018	0.61 0.48	1.0	No
Chronic	4 Acute 21-28 Chr	**Mysidopsis bahia** (Pocket shrimp juveniles)	0.011 0.0087	0.42	0.022	0.50 0.40	1.0	No
Chronic	2 Acute 21-28 Chr	**Mysidopsis bahia** (Pocket shrimp juveniles)	0.011 0.0087	0.87	0.046	0.24 0.19	1.0	No
Chronic	1 Acute 21-28 Chr	**Mysidopsis bahia** (Pocket shrimp juveniles)	0.011 0.0087	>1.2	>0.063	<0.17 <0.14	1.0	No
Chronic	2 Acute 21-28 Chr	**Daphnia magna** (1st Instar)	0.011 0.0087	0.79	0.042	0.26 0.21	1.0	No
Chronic	1 Acute 21-28 Chr	**Daphnia magna** (1st Instar)	0.011 0.0087	1.2	0.063	0.17 0.14	1.0	No
Chronic	8 Acute 21-28 Chr	**Limnea stagnalis** (Pond snail)	0.011 0.0087	>1.0	>0.053	<0.21 <0.16	1.0	No
Chronic	2 Acute 21-28 Chr	**Paneus setrifus** (White shrimp)	0.011 0.0087	>1.0	>0.053	<0.21 <0.16	1.0	No
Chronic	2 Acute 21-28 Chr	Daphnia magna (Adults)	0.011 0.0087	1.6	0.084	0.13	1.0	No

[1] EEC = Time-weighted Average of Expected environmental concentration.

[2] Acute LC50 = Concentration of Diquat that kills or immobilized 50% of the test animals in 96 hours.

[3] Estimated Chronic MATC = (acute LC50/(acute/chronic toxicity ratio).
[4] LOC = Level of Concern = Value (EEC/toxicity) which should not be exceeded as an indicator of the safety of a particular pesticide application to the biota.
NA = Not applicable

Table 24: Chronic Toxicity (MATC) of diquat to Amphibians (Embryos and Tadpole Early Life-Stage)

Test Species	% A.I.	Test Results (MATC in ppm)	Reference
Rana Pipiens (Leopard frog)	NR	100^1 $(54)^2$	Bimber and Mitchell, 1977
Rana Pipiens (Leopard frog)	NR	3.2^1 $(1.7)^2$	Dial & Dial , 1987
Xenopus laevis (African clawed toad)	Technical (~100% ddb)	1.18^1 $(0.64)^2$	Anderson and Prahlad, 1976

[1] ppm in diquat dibromide equivalence (ddb).
[2] ppm in cation equivalence (c.e.).

Table 25: Acute Toxicity of Diquat to Birds

Diquat Formulation	Organism	LD50	Toxicity Ranking	Reference/Date Reported
Cation	Mallard duck	71 mg/kg body weight (Acute LD50)	Moderately toxic	Fink et al, 1982a
Cation	Mallard duck	60.6 mg/kg body weight (Acute LD50)	Moderately toxic	Brian Database, 1982
Cation	Bobwhite quail	575 ppm dietary (8-day LC50)	Slightly toxic	Brian Database, 1975
Cation	Japanese quail	264 ppm dietary (8-day LC50)	Moderately toxic	Brian Database, 1975
Cation	Ring-necked pheasant	734 ppm dietary (8-day LC50)	Moderately toxic	Brian Database, 1975
Cation	Bobwhite quail	>19.6 ppm dietary (NOEL) 1 generation reproduction	N/A[1]	EPA 1995 RED
Cation	Mallard duck	5 ppm dietary (NOEL) 1 generation reproduction	N/A	EPA 1995 RED

[1] N/A = Not Applicable.

Table 26: Acute Toxicity of Diquat to Mammals

Diquat Formulation	Organism	LD50 (ppm)	Toxicity Ranking	Reference/Date Reported
Acute oral rat	Test material	810 mg/kg (males) 600 mg/kg (females)	N/A[1]	EPA 1995 RED
Acute oral rat	Diquat dibromide	120 mg/kg	N/A	Ecotoxnet Database
Acute oral mouse	Diquat dibromide	233 mg/kg	N/A	Ecotoxnet Database
Acute oral guinea pig	Diquat dibromide	187 mg/kg	N/A	Ecotoxnet Database
Acute oral dog	Diquat dibromide	187 mg/kg	N/A	Ecotoxnet Database
Acute oral rabbit	Diquat dibromide	188 mg/kg	N/A	Ecotoxnet Database
Acute oral cow	Diquat dibromide	30-56 mg/kg	N/A	Ecotoxnet Database
Acute dermal rabbit	Diquat dibromide	400-500 mg/kg	N/A	Ecotoxnet Database
NOEL (cation)	Chronic dog	NOEL 0.5 mg diquat/kg/day	N/A	Hopkins, 1990
NOEL (cation)	Rat	NOEL systemic toxicity 16 ppm (0.8 mg/kg/day). NOEL reproductive toxicity 80 ppm (4 mg/kg/day).	N/A	Hodge, 1990

[1] N/A = Not Applicable.

Table 27: Terrestrial Plant, Bird and Mammal Federally Endangered Species found in the State of Washington

	Common Name	Scientific Name
Terrestrial Plants		
	Ute Ladies'-Tresses	*Spiranthes diluvialis*
	Golden Paintbrush	*Castilleja levisecta*
	Nelson's Checker- Mallow	*Sidalcea nelsoniana*
Birds	Aleutian Canada Goose	*Branta Canadensis Leucopareia*
	American Peregrine Falcon	*Falco peregrinus anatum*
	Bald Eagle	*Haliaeetus leucocephalus*
	Brown Pelican	*Pelecanus occidentalis*
	Marbled Murrelet	*Brachyramphus marmoratus*
	Northern Spotted Owl	*Strix occidentalis caurina*
	Western Snowy Plover	*Charadrius alexandinus nivosus*
Mammals	Gray Wolf	*Canis lupis*
	Grizzly Bear	*Ursus arctos horribilis*
	Woodland Caribou	*Rangifer tarandus caribou*
	Columbian White-Tailed Deer	*ODOoileus virginianus leucurus*

For a listing of Washington State sensitive, threatened and endangered species, see Appendix 5

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LIST OF APPENDICES

Appendix 1: Acute Toxicity of Diquat Products to Fish

Species	Size/Age Class	Water Hardness (ppm $CaCO_3$)	%A.I.	1D LC50 (ppm)	2D LC50 (ppm)	3D LC50 (ppm)	4D LC50 (ppm)	Source
Anguilla rostra (American eel)	Black eel stage; 97.2mm	44 (Soft)	NR[1] (ddb)[2]	NA	NA	NA	55[3] (29.7)[4]	Hinton & Eversole, 1979
Anguilla rostra (American eel)	Glass eel stage; 55mm	44 (Soft)	NR (ddb)	NA	NA	NA	30.92 (17)	Hinton & Eversole, 1979
Brachdanio rario (Zebra fish)	NR	NR	20(c.e.)[5]	56 (30)	45 (24)	36 (19)	24 (13)	Benijt-Claus & Persoone, 1975
Brachdanio rario (Zebra fish)	NR	170 (Inter)	NR(c.e.)	>100				Devillers et al, 1985 in Hammer, 1994
Carrasius auratus (Goldfish)	NR	NR	20(c.e.)	NA	NA	NA	(35)	Gilderhaus, 1967
Carrasius auratus (Goldfish)	39-60cm	50 (Soft)	NR(c.e.)	NA	NA	NA	(85)	Berry, 1984
Carrasius auratus (Goldfish)	Fingerling	50 (Soft)	35.3(ddb)	>100 form (>19)	NA	NA	NA	FWS, 1986 in Brian, 1999
Cirrhina mrigala (Hamilton's carp)	40-50mm	NR	20(c.e.)	NA	NA	NA	(75)	Singh & Yadav, 1978
Ctenophryngodon idella (Grass carp)	10.5cm,23.5 g	21 (Very Soft)	35.3ddb)	NA	NA	NA	(53) (28.62)	El-Deen & Rogers, 1992
Ctenophryngodon idella (Grass carp)	95mm, 15.8g	270 (Hard)	13.5(ddb)	5967 (form)[6] (434)[4]	2092 form[6] (153)[4]	NA	1718 form[6] (125)[4]	Tooby et al, 1980

Appendix 1: Acute Toxicity of Diquat Products to Fish (continued)

Species	Size/Age Class	Water Hardness (ppm $CaCO_3$)	%A.I.	1D LC50 (ppm)	2D LC50 (ppm)	3D LC50 (ppm)	4D LC50 (ppm)	Source
Cyprinodon variegatus (Sheepshead minnow)	25mm, 0.25g	Saltwater	41.4(ddb)	(>82)	(>82)	(>57)	(48)	Nicholson (SBI) 1987 in Hammer, 1994 & in Brian, 1999
Cyprinus carpio (Common carp)	52mm,4.4g	48 (Soft)	26.8 (ddb)	(285)	(143)	(91)	(67)	Tapp, 1988
Cyprinus carpio (Common carp)	1-2 yr	NR	NR	NA	NA	NA	(141)	Silvo, 1967 in Hammer, 1994
Cyprinus carpio (Common carp)	NR	NR	NR	NA	NA	NA	(>9)	Tadokoro, 1986 in Campbell et al, 2000
Cyprinus carpio (Common carp)	NR	NR	20(c.e.)	NA	NA	NA	(35)	Gilderhaus, 1967
Cyprinus carpio (Common carp)	9 weeks 0.4227g	22 (Soft)	0.02(ddb)	94 (51)	NA	NA	50 (27)	Chin & Sudderuddin, 1979
Cyprinus carpio (Common carp)	6.5mm	NR	NR	(17)	(6.2)		(3.4)	Skea et al, 1987 in Campbell et al, 2000
Erimyzon sucetta (Lake chubsucker)	Sac-fry	NR	NR(c.e.)	NA	(1.3)	NA	NA	Hiltibran, 1967
Erimyzon sucetta (Lake chubsucker)	Sac-fry	NR	NR(c.e.)	NA	(2.5)	NA	NA	Hiltibran, 1967
Esox lusius (Northern pike)	NR	NR	20(c.e.)	NA	NA	NA	(16)	Gilderhaus, 1967

Appendix 1: Acute Toxicity of Diquat Products to Fish (continued)

Species	Size/Age Class	Water hardness (ppm $CaCO_3$)	%A.I.	1D LC50 (ppm)	2D LC50 (ppm)	3D LC50 (ppm)	4D LC50 (ppm)	Source
Esox masquinongy (Muskellunge)	3-day sac-fry	81 (Inter)	NR	(>161)	NA	NA	NA	Skea et al, 1987 in Hammer, 1994
Esox masquinongy (Muskellunge)	10-day sac-fry	81 (Inter)	NR	(62)	(10)	NA	(5.8)	Skea et al, 1987 in Hammer, 1994
Esox masquinongy (Muskellunge)	57mm	81 (Inter)	NR	(18)	(7.6)	NA	(4.6)	Skea et al, 1987 in Hammer, 1994
Fundulus similis (Longnosed killifish)	NR	Saltwater	NR	(>1.0)	(>1.0)	NA	NA	Butler, 1965 (c.e. based on input from Hammer, 1994)
Gambusia affinis (Mosquito fish)	NR	NR	NR	(10)	NA	NA	NA	Ahmed, 1967 in Campbell, 2000
Gambusia affinis (Mosquito fish)	31mm/0.32g	15 (Very soft)	Technical = ~100(ddb)	723 (390)	NA	NA	289 (156)	Leung et al, 1983
Ictalurus melas (Black bullhead)	0.8g	45 (Soft)	35.3(ddbmh)[7]	NA	NA	NA	170 (86.7)	Johnson & Finley, 1980, USFS, 1980 in Brian, 1999
Ictalurus melas (Black bullhead)	0.8g	NR	35.3(ddb)	NA	NA	NA	24.6 form (4.6)	EPA, 1995 & FWS, 1986 in Brian
Ic*talurus punctatus* (Channel catfish)	13mm/sac-fry	78 (Inter)	40(ddb)	NA	NA	NA	>10 (>5.4)	Jones, 1965
Ic*talurus punctatus* (Channel catfish)	50 mm	NR	40(ddb)	NA	NA	NA	(>10)	Lawrence et al, 1965

Appendix 1: Acute Toxicity of Diquat Products to Fish (continued)

Species	Size/Age Class	Water hardness (ppm $CaCO_3$)	%A.I.	1D LC50 (ppm)	2D LC50 (ppm)	3D LC50 (ppm)	4D LC50 (ppm)	Source
Lepomis macrochirus (Bluegill sunfish)	40mm, 0.85g	53 (Inter)	NR(c.e.)	(175)	(88)	(64)	NA	McCann, 1967 in Campbell, 2000
Lepomis macrochirus (Bluegill sunfish)	NR	120 (Inter)	35.3(ddb)	NA	NA	NA	18 (9.72)	Bishop & Perry, 1961
Lepomis macrochirus (Bluegill sunfish)	NR	29 (Very soft)	NR(ddb)	525 (283.5)	150 (81)	NA	NA	Hughes & Davis, 1962 in Campbell et al, 1999
Lepomis macrochirus (Bluegill sunfish)	20mm/fry	78 (Inter)	40(ddb)	NA	NA	NA	25 (13.5)	Jones, 1965
Lepomis macrochirus (Bluegill sunfish)	23mm,0.11g	45 (Soft)	19.8(c.e.)	100 form (20)	75 form (15)	NA	70 form (14)	Thompson et al, 1980
Lepomis macrochirus (Bluegill sunfish)	40mm, 0.85g	53 (Inter)	35.3(ddb)	175 form (33.4)	88 form (16.8)	NA	64 (12.1)	EPA, 1995 & McCann (1967 in Brian, 1999
Lepomis macrochirus (Bluegill sunfish)	NR	NR	20(c.e.)	NA	NA	NA	(35)	Gilderhaus, 1967
Lepomis macrochirus (Bluegill sunfish)	41mm	22 (Very Soft)	40(ddb)	91 (49)	80 (43)	NA	72 (39)	Surber & Pickering, 1962
Lepomis macrochirus (Bluegill sunfish)	45mm	340 Hard	40(ddb)	410 (221)	221 (113)	NA	140 (76)	Surber & Pickering, 1962
Lepomis macrochirus (Bluegill sunfish)	50mm	NR	40(ddb)	NA	NA	NA	(>10)	Lawrence et al, 1965
Lepomis macrochirus (Bluegill sunfish)	50mm/sac-fry	NR	NR(c.e.)	NA	NA	NA	(>10) 12-days	Hiltibran, 1967
Lepomis macrochirus (Bluegill sunfish)	1.3g	45 (Soft)	35.3(ddbmh)	NA	NA	NA	245 (125)	Johnson and Finley, 1980

Appendix 1: Acute Toxicity of Diquat Products to Fish (continued)

Species	Size/Age Class	Water Hardness (ppm $CaCO_3$)	%A.I.	1D LC50 (ppm)	2D LC50 (ppm)	3D LC50 (ppm)	4D LC50 (ppm)	Source
Lepomis macrochirus (Bluegill sunfish)	1.1g	NR	35.3(ddb)	NA	NA	NA	115 form (21.5)	EPA, 1995 & FWS, 1986 in Brian, 1999
Micropterus dolomieu (Smallmouth bass)	sac-fry	NR[1]	NR(c.e.)	(2.5)	NA	NA	NA	Hiltibran, 1967
Micropterus dolomieu (Smallmouth bass)	sac-fry	NR	NR(c.e.)	NA	NA	NA	(1.3)	Hiltibran, 1967
Micropterus dolomieu (Smallmouth bass)	11mm/fry	72 (Inter)	NR	(2.4)	NA	NA	NA	Skea et al, 1987 in Hammer, 1994
Micropterus dolomieu (Smallmouth bass)	10mm, 8 mg	16 (Very Soft)	NR	(0.35)	NA	NA	NA	Schenk, 1967 in Campbell, 2000
Micropterus dolomieu (Smallmouth bass)	6-8 day fry	81 (Inter)	24(c.e.)	(110)	(28)	(10)	(3.9)	Paul et al, 1999
Micropterus salmoides (Largemouth bass)	9-13 day fry	46-81 (Soft to Inter)	24(c.e.)	(15)	(11)	(8)	(4.9)	Paul et al, 1999
Micropterus salmoides (Largemouth bass)	16mm	72 (Inter)	NR	(3.6)	(1.9)	NA		Skea & Shiftlet, 19?? In Hammer, 1994
Micropterus salmoides (Largemouth bass)	18mm fry	78 (Inter)	40(ddb)	NA	NA	NA	>0.5 (>0.27)	Jones, 1965
Micropterus salmoides (Largemouth bass)	18mm fry	78 (Inter)	40(ddb)	NA	NA	NA	>1.0 (>0.54)	Jones, 1965
Micropterus salmoides (Largemouth bass)	18 mm fry	78 (Inter)	40(ddb)	NA	NA	NA	>3.0 (>1.62)	Jones, 1965
Micropterus salmoides (Largemouth bass)	NR	NR	NR	NA	(11)	NA		Muirhead-Thompson, 1971 in Campbell et al, 2000

Appendix 1: Acute Toxicity of Diquat Products to Fish (continued)

Species	Size/Age Class	Water Hardness (ppm $CaCO_3$)	%A.I.	1D LC50 (ppm)	2D LC50 (ppm)	3D LC50 (ppm)	4D LC50 (ppm)	Source
Micropterus salmoides (Largemouth bass)	50mm	NR	40(ddb)	NA	NA	NA	(>10)	Lawrence et al, 1965
Micropterus salmoides (Largemouth bass)	100mm	56 (Inter)	NR	NA	(548)	(178)	(60)	Skea & Shiftlet, 19?? In Hammer, 1994
Micropterus salmoides (Largemouth bass)	64mm	21 (Very Soft)	40(ddb)	24 (13)	11 (5.9)	NA	7.8 (4.2)	Surber and Pickering, 1962
Morone saxatilis (Striped bass)	1-week sac-fry	40 (Soft)	35.3(ddb)	1.0 (0.54)	1.0 (0.54)	1.0 (0.54)	1.0 (0.54)	Hughes, 1969, Hughes, 1973
Morone saxatilis (Striped bass)	1-month Fingerling	40 (Soft)	35.3(ddb)	35 (19)	25 (13)	15 (8.1)	10 (5.4)	Hughes, 1969, Hughes, 1973
Morone saxatilis (Striped bass)	60mm, 2.7g/ subadult	40 (Soft)	37(ddb) 20 (c.e.)	315 (170)	155 (84)	NA	80 (43)	Wellborn, 1969
Morone saxatilis (Striped bass)	Fingerling	NR	35.39(ddb)	NA	NA	NA	43.2	EPA RED, 1995; REF, 1969 in Brian, 2000
Notoemigonus crysoleucans (Golden shiner)	74mm	64 (Inter)	NR	NA	(70)	(41)	(33)	Skea & Shiflet, 19?? In Hammer, 1994
Notropis atherinoides (Emerald shiner)	55mm,1.2g	132 (Inter)	NR(ddb)	>180 (>97)	>180 (>97)	86.2 (46.5)	25.8 (13.9)	Swabey & Schenk, 1963 in Hammer, 1994 & Campbell, 2000
Notropis atherinoides (Emerald shiner)	NR	NR	20(c.e.)	NA	NA	NA	(25.8)	REF, 1963 in Brian, 1999
Oncorhynchus kisutch (Coho salmon)	45mm	NR	NR	NA	NA	NA	(20.5)	Watkins, 1966 in Hammer, 1994
Oncorhynchus kisutch (Coho salmon)	Yearling	100 (Inter)	40(ddb)	NA	NA	NA	(30)	Lorz et al, 1979

Appendix 1: Acute Toxicity of Diquat Products to Fish (continued)

Species	Size/Age Class	Water Hardness (ppm $CaCO_3$)	%A.I.	1D LC50 (ppm)	2D LC50 (ppm)	3D LC50 (ppm)	4D LC50 (ppm)	Source
Oncorhynchus kisutch (Coho salmon)	NR	NR	NR(c.e.)	NA	(29)	NA	NA	Muirhead –Thompson, 1971
Oncorhynchus mykiss (Rainbow trout)	50mm	NR	40(ddb)	NA	NA	NA	(>5)	Lawrence et al, 1965
Oncorhynchus mykiss (Rainbow trout)	NR	NR	20(c.e.)	NA	NA	NA	(11.2)	Gilderhaus, 1967
Oncorhynchus mykiss (Rainbow trout)	35mm,0.56g	45 (Soft)	19.8(c.e.)	160 form[6] $(31)^3$	130 form (25)	NA	75 form (15)	Thompson et al (ABC), 1980
Oncorhynchus mykiss (Rainbow trout)	30mm, 0.44g	51 (Inter)	NR	(103)	(53)	NA	(26)	Hill et al, 1976 in Hammer, 1994
Oncorhynchus mykiss (Rainbow trout)	NR	NR	NR	(90)	NA	(16)	(8)	Carter, 1971 in Campbell et al, 2000
Oncorhynchus mykiss (Rainbow trout)	38.7mm, 0.89g	52 (Inter)	26.8 (c.e.)	(69)	(27)	(23)	(21)	Tapp & Caunter, 1988
Oncorhynchus mykiss (Rainbow trout)	48.6mm,1.89g	52 (Inter)	26.8 (c.e.)	NA	NA	NA	(6.1)	Tapp & Caunter, 1989
Oncorhynchus mykiss (Rainbow trout)	NR	250 (Hard)	40(ddb)	225 form (48.6)	175 form (37.8)	NA	NA	Alabaster, 1969
Oncorhynchus mykiss (Rainbow trout)	NR	250 (Hard)	100 (ddb)	90 (48.6)	70 form (37.8)	NA	NA	Alabaster, 1969
Oncorhynchus mykiss (Rainbow trout)	Fingerling	NR	35.3(ddb)	NA	NA	NA	>100 (>18.7)	EPA, 1995, &F WS, 1986 in Brian, 1999
Oncorhynchus tshawytscha (Chinook salmon)	58-96mm	NR	NR(ddb)	30 (16)	29 (16)	NA	NA	Bond et al, 1960

Appendix 1: Acute Toxicity of Diquat Products to Fish (continued)

Species	Size/Age Class	Water hardness (ppm $CaCO_3$)	%A.I.	1D LC50 (ppm)	2D LC50 (ppm)	3D LC50 (ppm)	4D LC50 (ppm)	Source
Perca flavecens (Yellow perch)	0.6g	45 (Soft)	35.3 (ddbmh)[7]	NA	NA	NA	60 (30.6)	Johnson & Finley, 1980
Perca flavecens (Yellow perch)	NR	NR	NR(ddb)	NA	NA	NA	114 (60)	Holtz & Winter, 1989
Perca flavecens (Yellow perch)	0.6g	NR	35.3(ddb)	NA	NA	NA	23.5 form (4.4)	EPA, 1995 &FWS, 1986 in Brian, 1999
Pimephales promelas (Fathead minnow)	NR	NR	NR	NA	(24)	NA	(24)	Hong et al, 1988 in Campbell, 2000
Pimephales promelas (Fathead minnow)	47mm	379 (Hard)	40(ddb)	260 (140)	220 (119)	NA	130 (70)	Surber & Pickering, 1962
Pimephales promelas (Fathead minnow)	48mm	22 (Very Soft)	40(ddb)	140 (76)	26 (14)	NA	14 (7.6)	Surber & Pickering, 1962
Pimephales promelas (Flathead minnow)	45mm	22 (Very Soft)	40(ddb)	>56 (>30)	23 (12)	NA	14 (7.6)	Surber & Pickering, 1962
Pimephales promelas (Flathead minnow)	50mm	NR	40(ddb)	NA	NA	NA	>10	Lawrence et al, 1965
Pimephales promelas (Flathead minnow)	Juveniles	NR	35(ddb)	(76.5)	NA	NA	NA	de Peyster & Long, 1993
Poecilla reticulata (Guppy)	25mm	NR	20 (c.e.)	NA	NA	NA	50	Lee & Furtado, 1977 in Hammer, 1994
Poecilla reticulata (Guppy)	10mm	NR	NR	(180)	(50)	NA	(4.4)[7.7 days]	Van Dord et al, 1974 in Hammer, 1994
Poecilla mexicana (Mexican molly)	NR	NR	NR(c.e.)	16.8	NA	NA	(9.0) [5.25 days]	Moore, R.J., 1978
Rasbora heteromorpha (Harlequin fish)	20mm	20 (Very Soft)	40(ddb)	190 form (41)	70 form (16)	NA	NA	Alabaster, 1969

Appendix 1: Acute Toxicity of Diquat Products to Fish (continued)

Species	Size/Age Class	Water hardness (ppm $CaCO_3$)	%A.I.	1D LC50 (ppm)	2D LC50 (ppm)	3D LC50 (ppm)	4D LC50 (ppm)	Source
Rasbora heteromorpha (Harlequin fish)	20mm	20 (Very Soft)	40(ddb)	180 form (39)	93 form (16)	NA	NA	Alabaster, 1969
Rasbora trillineata (Scissor-tail rasbora)	30mm	NR	20(c.e.)	NA	NA	NA	(30)	Lee & Furtado
Salmo trutta (Brown trout)	40.3mm	NR	NR	(33)	(22)	NA	(20)	Simonin & Skea, 1977 in Campbell et al, 2000
Salmo trutta (Brown trout)	NR	NR	NR	NA	NA	NA	(17.8)	Simonin & Skea, 1977 in Campbell et al, 2000
Salmo trutta (Brown trout)	yearling	~250 (Hard)	40(ddb)	NA	300 form (64.8)	NA	NA	Woodiwiss et al, 1974
Salmo trutta (Brown trout)	yearling	~250 (Hard)	40(ddb)	NA	570 form (23.12)	NA	NA	Woodiwiss et al, 1974
Salmo trutta (Brown trout)	Fingerling	NR	35.3(ddb)	NA	NA	NA	(17.77)	EPA RED, 1995 & CCC, 1977 in Brian, 1999
Salvelinis fontinalis (Eastern brook trout)	NR	NR	NR(ddb)	>20 (>11)	NA	NA	NA	Eady & Reaney, 1965
Salmonid Trout	NR	NR	NR	(33)	(22)	NA	(20)	Holden, 1964 in Hammer, 2000
Stizostedion vitreu (Walleye)	NR	NR	20(c.e.)	NA	NA	NA	(2.1)	Gilderhaus, 1967

Appendix 1: Acute Toxicity of Diquat Products to Fish (continued)

Species	Size/Age Class	Water hardness (ppm $CaCO_3$)	%A.I.	1D LC50 (ppm)	2D LC50 (ppm)	3D LC50 (ppm)	4D LC50 (ppm)	Source
Stizostedion vitreu (Walleye)	8-10 day fry	132 (Inter)	24(c.e.)	(2.9)	(1.6)	(1.0)	(0.75)	Paul et al, 1999
Stizostedion vitreu (Walleye)	41-43 day fry	132 (Inter)	24(c.e.)	(3.1)	(1.9)	(1.6)	(1.5)	Paul et al, 1999
Stizostedion vitreu (Walleye)	84-46 day fry	132 (Inter)	24(c.e.)	(7.8)	(4.9)	(4.9)	(4.9)	Paul et al, 1999

[1] NR = Not reported.
[2] ddb = diquat dibromide equivalence
[3] Values not in parenthesis expresses as ppm ddb or ppm ddbmh.
[4] Values in parenthesis are expressed as ppm c.e.
ppm c.e. = (ppm ddb) x 0.54 = (ppm ddbmh) x 0.51 = ppm form) x %AI/100 x 0.54
[5] c.e. = Cation equivalence
[6] form = formulation
[7] ddbmh = diquat dibromide monohydrate.
NA = Not available

Appendix 2: Acute Toxicity of Diquat Products to Invertebrates

Species	Size/Age Class	Water hardness (ppm $CaCO_3$)	%A.I.	1D LC50 (ppm c.e.)	2D LC50 (ppm c.e.)	3D LC50 (ppm c.e.)	4D LC50 (ppm c.e.)	Source
Alonella spp (Water flea)	NR[1]	27 (Soft)	8.53(ddb)[2]	NA	>10[3] (>5.4)[4]	NA	NA	Naqvi & Hawkins, 1989
Asellus communis (Isopod)	NR	NR +sediment	NR	NA	NA	NA	(>10) (120hrs)	Williams et, al, 1984
Callibaetis spp. (Mayfly)	NR	64 (Inter)	20 (c.e.)[5]	>100	37	NA	(16.4)	Wilson & Bond, 1969, Wilson, 1967
Cardium edule (Cockle)	NR	NR	NR(ddb)	NA	<10 (<5.4)	NA	NA	Portman, 1972
Caridina nilotica (Freshwater shrimp)	Adult	12 (Very Soft)	20(ddbmh)[6]	213.4 in 2 hrs (108.8)	NA	NA	NA	Kevan & Pearson, 1993
Cragnon cragnon (Shrimp)	NR	NR	NR(ddb)	NA	>10 (>5.4)	NA	NA	Portman, 1972
Crassostrea virginica (Eastern oyster)	Shell Deposition	Saltwater	NR	NA	NA	NA	(>1.0)	Butler , 1965 & EPA, 1986 in Brian, 1999
Crassostrea virginica (Eastern oyster)	Spat	Saltwater	41.4 (ddbmh)	NA	NA	NA	260 form[7] (55)	Dionne (SBI), 1987
Crassostrea virginica (Eastern oyster)	Shell Deposition	Saltwater	NR	NA	NA	NA	670 form (141)	Dionne (SBI), 1987
Cypria spp. (Ostracod)	NR	27 (Very Soft)	8.53(ddb)	NA	13.8 (7.452)	NA	NA	Naqvi & Hawkins, 1989
Daphnia maga (Water flea)	NR	24 (Very Soft)	NR	NA	3 (1.62)	NA	NA	Bishop & Perry, 1961

Appendix 2: Acute Toxicity of Diquat Products to Invertebrates (continued)

Species	Size/Age Class	Water hardness (ppm $CaCO_3$)	%A.I.	1D LC50 (ppm c.e)	2D LC50 (ppm c.e.)	3D LC50 (ppm c.e.)	4D LC50 (ppm c.e.)	Source
Daphnia magna (Water flea)	NR	40 (Soft)	91.6 (ddb)	(2.5)	(1.2)	NA	NA	Wheeler, 1978 & CCC, 1978 in Brian, 1999
Daphnia magna (Water flea)	1st Instar	NR	NR (ddb)	7.1 (3.834)	NA	NA	NA	Crosby & Tucker, 1966 (ddb)
Daphnia magna (Water flea)	Adult	NR	20 (ddb)	6.9 (3.7)	3.0 (1.6)	NA	NA	Benijts-Claus & Persoone, 1975
Daphnia magna (Water flea)	1st Instar	NR	46.6 (ddbmh)	NA	1.17	NA	NA	EPA, 1995 & CCC, 1978 in Brian, 1999
Daphnia magna (Water flea)	1st Instar	NR	46.6 (ddbmh)	NA	1.03	NA	NA	EPA, 1995
Daphnia magna (Water flea)	1st Instar	NR	35.2 (ddb)	NA	0.77	NA	NA	EPA, 1995 & CCC, 1978 in Brian, 1999
Daphnia magna (Water flea)	1st Instar	NR	NR	2.4 (1.296)	0.60 (0.324)	NA	NA	Benijts-Claus & Persoone, 1975
Daphnia pulex (Water flea)	Immature	NR	NR				>1.0 (8 days)	Gilderhaus, 1967
Daphnia pulex (Water flea)	Adults	NR	NR				<1.0 (8 days)	Gilderhaus, 1967
Daphnia pulex (Water flea)	Adult	NR	20 (ddb)	1.3 (0.702)	0.4 (0.216)		NA	Benijts-Claus & Persoone, 1975
Daphnia pulex (Water flea)	1st Instar	NR	20 (ddb)	1.5 (0.81)	0.3 (0.162)		NA	Benijts-Claus & Persoone, 1975 (ddb)

Appendix 2: Acute Toxicity of Diquat Products to Invertebrates (continued)

Species	Size/Age Class	Water hardness (ppm $CaCO_3$)	%A.I.	1D LC50 (ppm c.e)	2D LC50 (ppm c.e.)	3D LC50 (ppm c.e.)	4D LC50 (ppm c.e.)	Source
Daphnia similis (Water flea)	NR	NR	NR	4.3	NA	NA	NA	Tadokoro, 1986 in Hammer, 1994
Diapotomus spp. (Diapotomus)	NR	27 (Soft)	8.53 (ddb	NA	~10 (~5.4)	NA	NA	Naqvi & Hawkins, 1989
Diaptomus spp. & *Eucyclops* spp. (Copepod)	NR	NR	NR (c.e.)	74	19	NA	NA	Naqvi et al, 1980
Enallagma spp. (Dragonfly)	Larvae	64 (Inter)	20 (c.e.)	>100	>100		>100	Wilson & Bond, 1969, Wilson, 1967
Eucyclops spp. (Copepod)	NR	27 (Very Soft)	8.53 (ddb)	NA	46.6 (25.2)	NA	NA	Naqvi & Hawkins, 1989
Gammarus fasciatus (Scud)	Mature	45 (Soft)	37.3(ddbmh)	NA	NA	NA	>100 (>51)	Johnson & Finley, 1980
Gammarus fasciatus (Scud)	NR	72 (Inter)	Commercial EC (ddb)	NA	NA	NA	>100 (>54)	Sanders, 1970
Gammarus fasciatus (Scud)	NR	NR	35.3(ddb)	NA	NA	NA	18.7	EPA, 1995 & Campbell et al, 2000
Hyalella azteca (Amphipod)	4-8mm	64 Inter	20 (c.e.)	(0.58)	(0.12)	NA	(0.048)	Wilson & Bond, 1969, Wilson, 1967
Hyalella azteca (Amphipod)	4-8mm	64 +sediment (Inter)	NR	(11.5)	(10.5)	NA	(6.8)	Wilson & Bond, 1969, Wilson, 1967

Appendix 2: Acute Toxicity of Diquat Products to Invertebrates (continued)

Species	Size/Age Class	Water hardness (ppm $CaCO_3$)	%A.I.	1D LC50 (ppm c.e)	2D LC50 (ppm c.e.)	3D LC50 (ppm c.e.)	4D LC50 (ppm c.e.)	Source
Hyalella azteca (Amphipod)	NR	NR	Unknown	NA	(0.14)	NA	NA	EPA, 1995, REF, 1974 in Brian, 1999
Hyalella azteca (Amphipod)	NR	NR	NR	NA	(0.64)	NA	NA	Williams, et al, 1984 in Campbell et al 2000
Hyalella azteca (Amphipod)	NR	NR +sediment	NR	NA	(3.4)	NA	NA	Williams, et al, 1984
Hyalella azteca (Amphipod)	NR	NR	Commercial Product	1.26 (0.67)	NA	NA	NA	Nicholson & Clerman, 1974
Libellula spp. (Dragonfly)	Larvae	64 (Inter)	20 (c.e.)	(>100)	(>100)	NA	(>100)	Wilson & Bond, 1969, Wilson, 1967
Limnea stagnalis (Pond snail)	NR	NR	NR	NA	NA	NA	>1.0(8 days)	Carter, 1971
Limnephilus spp. (Caddisfly)	NR	64 (Inter)	20 (c.e.)	(>100)	(65)	NA	(33)	Wilson & Bond, 1969, Wilson, 1967
Mysidopsis bahia (Pocket shrimp)	Juvenile	Saltwater	41.1(ddbmh)	>5.7 form (>1.2)	4.1 form (0.87)	2.7 form (0.46)	2.0 form (0.42)	Hoberg (SBI), 1987
Nymphula responsalis (Aquatic moth)	Larvae	NR	20 (c.e.)	NA	NA	NA	86.94	Lee & Furtado, 1977
Paneus duroarum (Pink shrimp)	Juvenile	Saltwater	40.0 (ddb)	NA	40 form (8.48)	NA	NA	EPA, 1986 in Brian, 1999
Paneus setriferus (White shrimp)	NR	Saltwater	NR(c.e.)	(>1.0)	(>1.0)	NA	NA	Butler, 1965
Pomacea pludosa (Apple snail)	7-days to adult	45 +sediment (Soft)	35.3 (ddb)	NA	NA	NA	0.34	EPA, 1995, (FWS), 1986 & Winger et al, 1984 in Brian, 1999
Streptocephalus seali (Fairy shrimp)	Mature	120 (Inter)	35.3 (ddb)	>2.0 (>1.08)	>2.0 (>1.08)	>2.0 (>1.08)	NA	Moss, 1978
Tendipedinae (Bloodworms)	Larvae	64 (Inter)	20 (c.e.)	(>100)	(>100)		(>100)	Wilson & Bond, 1969, Wilson, 1967

[1] NR = Not reported
[2] ddb = diquat dibromide equivalence
[3] Values not in parenthesis expresses as ppm ddb or ppm ddbmh.
[4] Values in parenthesis expresses as ppm c.e.
ppm c.e. = (ppm ddb) x 0.54 = (ppm ddbmh) x 0.51 = (ppm form) x %AI/100 x 0.54.
[5] c.e. = cation equivalence.
[6] ddbmh = diquat dibromide monohydrate.
[7] form = formulation equivalence.
NA = Not available.

Appendix 3: Chronic Toxicity of Diquat Products to Fish and Amphibians

%A.I. or Form	Species	Affect	Water Hardness (ppm CaCO$_3$)	Size Class/ Age	Time (Days)	NOEC^{1} (ppm)	MATC^{2} (ppm)	LOEC^{3} (ppm)	LC50 (ppm)	Reference
NR^{1}	*Lepomis macrochirus* (Bluegill sunfish)	Survival	NR^{4}	Sac-fry	12	(>10)^{5}	(>10)^{5}	(>10)^{5}	(>10)^{5}	Hiltibran, 1967
NR	*Salvelinus fontanalis* (Eastern brook trout)	Abnormal sac-fry	NR	Eggs to sac-fry	NR	5^{6} (2.7)^{5}	7.1^{6} (3.8)^{5}	10^{6} (5.4)^{5}	>20^{6} (>11)^{5}	Eadey & Reaney, 1965
NR	*Ictalurus punctatus* (Channel catfish)	Weight reduction	NR	Eggs to free swimming fry	NR	(>1.0)	(>1.0)	(>1.0)	NR	EPA RED, 1995
41.4 (ddb)	*Pimaphales promelas* (Fathead minnow)	Weight reduction	34 (Soft)	Eggs to free swimming fry	34	(0.12)	(0.2)	(0.32)	(>0.65)	Suprenant, 1987
26.2 (ddb)	*Oncorhynchus mykiss* (Rainbow trout)	LC50	52 (Inter)	48.6mm, 1.89g	21	(1.4)	(<1.4)	(<1.4)	(2.9)	Tapp & Caunter, 1989
NR	*Fundulus heteroclitis* (Killifish)	Abnormal pigmentation and Slow development	NR	Eggs to free-swimming fry	40	0.01 (0.0054)	0.031 (0.017)	0.1 (0.054)	NR	Crawford & Guarino. 1985
NR	*Fundulus heteroclitis* (Killifish)	Survival	NR	Eggs to free -swimming fry	40	<0.01 (<0.0054)	<0.01 (<0.0054)	<0.01 (<0.0054)	<0.01 (<0.0054)	Crawford & Guarino, 1985
NR	*Rana pipiens* (Leopard frog)	Abnormal Development and Survival	NR	Eggs to 14 days post hatch	14-days post-hatch	<100 (<54)	<100 (<54)	<100 (<54)	<100 (<54)	Bimber & Mitchell, 1977
NR	*Rana pipiens* (Leopard frog)	Abnormal development	374 (Hard)	Embryos	13	2.0 (1.08)	3.2 (1.7)	5.0 (2.7)	NR	Dial & Dial, 1987
NR	*Rana pipiens* (Leopard frog)	Survival	374 (Hard)	15-day tadpole	13	>10 (>5.4)	>10 (>5.4)	>10 (>5.4)	>10 (>5.4)	Dial & Dial, 1987

Appendix 3: Chronic Toxicity of Diquat Products to Fish and Amphibians (continued)

%A.I. or Form	Species	Affect	Water Hardness (ppm $CaCO_3$)	Size Class/ Age	Time (Days)	NOEC[1] (ppm)	MATC[2] (ppm)	LOEC[3] (ppm)	LC50 (ppm)	Reference
Tech (ddb)[7]	*Xenopus laevis* (African clawed toad)	Body size reduction, Reduced pigmentation, Altered body shape, Muscular development	NR	Tailbud stage to tadpoles	10	1.0 (0.54)	1.18 (0.64)	1.25 (0.68)	~0.75 (~0.405)	Anderson & Prahlad, 1976
Tech (ddb)	*Xenopus laevis* (African clawed toad)	Survival	NR	Tailbud stage to tadpoles	10	<0.5 (<0.27)	<0.5 (<0.27)	<0.5 (<0.27)	~0.75 (~0.405)	Anderson & Prahlad, 1976

[1] NOEC = Statistical No Observed Effect Concentration. Endpoints of interest are, immobility, reduction in live-born young and reduction in growth.

[2] MATC = Maximum allowable toxic concentration. This concentration is the geometric mean of the NOEC and LOEC.

[3] LOEC = Statistical Lowest observed Effect Concentration. Endpoints of interest are: immobility, reduction in live-born young and reduction in growth.

[4] NR = Not reported.

[5] Values in parenthesis are expressed as ppm cation equivalence.

[6] Values not in parenthesis are expressed as ppm diquat dibromide equivalence. It is not clear in these papers whether the expression of active ingredient is in diquat dibromide equivalence or cation equivalence.

[7] ddb = diquat dibromide equivalence.

LC50 = Concentration that effects 50% of the test animals. In these studies, the endpoint for the LC50 effect is mortality.

Appendix 4: Chronic Toxicity of Diquat Products to Invertebrates (Daphnid)

% A.I.	Species	Water Hardness (ppm $CaCO_3$	Age	Time (days)	NOEC[1] (ppm c.e.)	MATC[2] (ppm c.e.)	LOEC[3] (ppm c.e.)	EC50 (ppm c.e.)	Reference
41.4 (ddbmh)[4]	*Daphnia magna* (Daphnid)	NR[5]	Life-cycle Reproduction & Growth	21	0.036	0.045	0.057	0.057-0.11	Suprenant, 1987

[1] NOEC = Statistical No Observed Effect Concentration. Endpoints of interest are, immobility, reduction in live-born young and reduction in growth.

[2] MATC = Maximum allowable toxic concentration. This concentration is the geometric mean of the NOEC and LOEC.

[3] LOEC = Statistical Lowest observed Effect Concentration. Endpoints of interest are: immobility, reduction in live-born young and reduction in growth.

[4] ddbmh = diquat dibromide monohydrate equivalence.

[5] NR = Not reported but daphnids grow best in hard water.

EC50 = Concentration that effects 50% of the test animals. In these studies, the endpoint for the EC50 effect is immobility or mortality.

Appendix 5: Sensitive, Threatened and Endangered Aquatic Species found in Washington State

Status	Region	Affected Counties within Region	Common Name	Species
Endangered	Upper Columbia River	All counties	Spring-run Chinook	*Oncorhynchus tshawytscha*
Endangered	Upper Columbia River	All counties	Steelhead trout	*Oncorhynchus mykiss*
Endangered	Snake River	All counties	Sockeye salmon	*Oncorhynchus nerka*
Threatened	Puget Sound	All Counties	Chinook salmon	*Oncorhynchus tshawytscha*
Threatened	Puget Sound	Clallam, Jefferson, Mason, Kitsap	Hood Canal Summer Chum salmon	*Oncorhynchus kisutch*
Threatened	Puget Sound	All , excluding Kitsap, San Juan, Island	Bull trout	*Oncorhynchus mykiss*
Threatened	Washington Coastal	Clallam	Lake Ozette Sockeye salmon	*Oncorhynchus nerka*
Threatened	Washington Coastal	All, excluding Pacific	Bull trout	*Salvelinus confluentus*
Threatened	Lower Columbia River	All counties	Spring-run Chinook salmon	*Oncorhynchus tshawytscha*
Threatened	Lower Columbia River	All counties	Chum salmon	*Oncorhynchus keta*
Threatened	Lower Columbia River	All counties	Steelhead trout	*Oncorhynchus mykiss*
Threatened	Lower Columbia River	All counties	Bull trout	*Salvelinus confluentus*
Threatened	Upper Columbia River	All counties	Bull trout	*Salvelinus confluentus*

Appendix 5: Sensitive, Threatened and Endangered Aquatic Species found in Washington State (continued)

Status	Region	Affected Counties within Region	Common Name	Species
Threatened	Middle Columbia River	All counties	Bull trout	*Salvelinus confluentus*
Threatened	Middle Columbia River	All counties	Steelhead trout	*Oncorhynchus mykiss*
Threatened	Middle Columbia River	All counties	Bull trout	*Salvelinus confluentus*
Threatened	Snake River	All counties	Spring/Summer-run Chinook salmon	*Oncorhynchus tshawytscha*
Threatened	Snake River	All counties	Fall-run Chinook salmon	*Oncorhynchus tshawytscha*
Threatened	Snake River	All counties	Steelhead trout	*Oncorhynchus mykiss*
Threatened	Snake River	All counties	Bull trout	*Salvelinus confluentus*
Threatened	Northeast Washington	All counties	Bull trout	*Salvelinus confluentus*
Proposed Threatened	Washington Coastal	Grays Harbor, Pacific, Lewis	Coastal cutthroat trout	*Oncorhynchus clarki*
Proposed as Threatened	Lower Columbia River	All counties	Coastal cutthroat trout	*Oncorhynchus clarki*
Potentially Threatened	Upper Columbia River	Chelan, Okanogan	Westslope cutthroat trout	*Oncorhynchus clarki*
Potentially Threatened	Middle Columbia River	Kittitas, Yakima, Klickitat	Westslope cutthroat trout	*Oncorhynchus clarki*

Appendix 5: Sensitive, Threatened and Endangered Species found in Washington State (continued)

Status	Region	Affected Counties within Region	Common Name	Species
Potentially Threatened	Northeast Washington	All counties	Westslope cutthroat trout	*Oncorhynchus clarki*
Candidate	Puget Sound	All counties	Coho salmon	*Oncorhynchus kisutch*
Candidate	Washington Coastal	Grays Harbor, Pacific, Lewis, Thurston	Southwest Washington Coho salmon	*Oncorhynchus kisutch*
Candidate	Lower Columbia River	All counties	Coho salmon	*Oncorhynchus kisutch*
Candidate	1[1]	All counties	Pacific Cod	*Gadua macrocephalus*
Candidate	1	All counties	Walleye Pollock	*Theragra chalcogramma* sp.
Candidate	1	All counties	Pacific Hake	*Merluccius productus*
Candidate	1	All counties	Brown Rockfish	*Sebastes* sp.
Candidate	1	All counties	Copper Rockfish	*Sebastes* sp.
Candidate	1	All counties	Quillback Rockfish	*Sebastes* sp.
Concern	1	All counties	Cherry point herring	Family Clupedidae
Concern	1	All counties	Discovery Bay Herring	Family Clupedidae
Concern	State Wide	All counties	River Lamprey	
Concern	State Wide	All counties	Van Dyke's Salamander	*Plethodon vandykei*
Concern	State Wide	All counties	Columbia torrent Salamander	*Rhyacotriton olympicus*
Concern	State Wide	All counties	Columbia spotted frog	*Rana lutieventris*
Concern	State Wide	All counties	Great Columbia River Spire Snail	
Concern	State Wide	All counties	Newcomb's Littorine Snail	
Concern	State Wide	All counties	California floater	*Adonata*

				californiensis
Concern	State Wide	All counties	Northern Abalone	*Haliotis sp.*
Concern	State Wide	All counties	Olympia Oyster	
State Endangered	State Wide	All Counties	Oregon spotted frog	*Rana pretiosa*

Appendix 5: Sensitive, Threatened and Endangered Aquatic Species found in Washington State (continued)

Status	Region	Affected Counties within Region	Common Name	Species
State Sensitive	State Wide	All counties	Pygmy whitefish	*Prosopium coolteri*
State Sensitive	State Wide	All counties	Margined sculpin	*Cottus marginatus*
State Sensitive	State Wide	All counties	Larch Mountain Salamander	*Plethodon larselli*
State Candidate	State Wide	All counties	Olympic mudminnow	
State Candidate	State Wide	All counties	Mountain sucker	*Catostomus platyrhynchus*
State Candidate	State Wide	All counties	Lake chub	*Coesius plumbeus*
State Candidate	State Wide	All counties	Leopard dace	*Rhinichthys falcatus*
State Candidate	State Wide	All counties	Umatilla dace	*Rhinichthys umatilla*
State Candidate	State Wide	All counties	Eulachon (Columbia River Smelt)	*Thaleichthys pacificus*
State Candidate	1	All counties	Black rockfish	*Sebastes* sp.
State Candidate	1	All counties	Tiger Rockfish	*Sebastes* sp.
State Candidate	1	All counties	Boccacio rockfish	*Sebastes paucispinis*
State Candidate	1	All counties	Canary rockfish	*Sebastes* sp.
State Candidate	1	All counties	Yellowtail rockfish	*Sebastes* sp.
State Candidate	1	All counties	Greenstriped rockfish	*Sebastes* sp.
State Candidate	1	All counties	Widow rockfish	*Sebastes* sp.
State Candidate	1	All counties	China rockfish	*Sebastes* sp.
State Candidate	State Wide	All counties	Dunn's Salamander	*Plethodon dunni*
State Candidate	State Wide	All counties	Cascade torrent salamander	*Rhyacotriton cascadae*
State Candidate Under Review	State Wide	All counties	Northern Leopard Frog	*Rana pipiens*
State Candidate	State Wide	All counties	Giant Columbia River Limpet	unknown

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[1] Within Puget Sound, the San Juan Islands, and the Strait of Juan de Fuca east of the Seiku River.

DIQUAT

Volume 4, Section 5

HUMAN HEALTH EFFECTS

48 Pages

TABLE OF CONTENTS

5.0 HUMAN HEALTH EFFECTS

5.1 OBJECTIVE

The Washington State Department of Ecology (WDOE) contracted with Compliance Services International (CSI) to prepare a document concerning potential human health impacts from aquatic applications of the herbicide diquat. This herbicide is currently being used to control noxious aquatic plants in the State of Washington.

The human health section is designed to provide the most recent health information to the WDOE concerning the toxicology of diquat and potential health risks to the public associated with diquat aquatic weed control. It is also the intent of this section to assist the agencies in making decisions regarding continued diquat uses and establishing various swimming alerts and waiting periods for water use following herbicide application to bodies of water.

The objectives of this section are to: 1) provide a review of the available diquat toxicology information, 2) determine the degree and types of potential exposures that may be encountered during various time periods following diquat aquatic application, and 3) present a series of risk assessments of the different types of exposures to aquatic applied diquat and determine any need or recommendations for mitigation of exposure to ensure public health.

5.2 APPROACH

5.2.1 Information Compilation

Information concerning diquat toxicology and health effects were obtained from computerized searches of the scientific and medical literature, EPA office of Pesticide Programs, WDOE and the herbicide registrant Zeneca Agrochemicals.

5.2.2 Risk Assessment Procedure

5.2.2.1 Diquat Toxicology Information and Assessment

Section 1 of this document discusses the registration and regulation of pesticides. Part of registering any pesticide with the US Environmental Protection Agency (US EPA) involves the potential registrant conducting a series of toxicology studies outlined in the Federal Insecticide, Fungicide and Rodenticide Act (FIFRA). One of EPA's functions is to ensure that all pesticides be registered in accordance with FIFRA guidelines. The series of toxicology studies include acute (one exposure), subchronic (multiple exposures, e.g. weeks or months) and chronic exposure to the chemical over the entire lifetime of the animal. These types of studies have been conducted on technical diquat.

In addition, once the technical chemical or active ingredient has been registered for specific uses with the EPA, various product formulations of the active ingredient are prepared, tested for efficacy and acute toxicology. The results of the acute toxicology studies, (see Table 1: oral, dermal, inhalation, skin and eye irritation and skin sensitization studies), are used to evaluate the health hazards that may be associated with overexposure to the applicator, bystanders and others that may contact treated areas following application of the pesticide. Once the hazards have been determined by the results of the tests, the specific product label warnings are then determined by

FIFRA guidelines and serve to alert the consumer as to the associated product health hazards and precautions to take to prevent overexposure.

The diquat product labels bear the “WARNING” signal word and precautionary statements telling the user that, depending upon the degree and duration of overexposure to the concentrated product, the chemical “may be fatal if absorbed through the skin, causes substantial, but temporary eye injury and can be harmful if swallowed or inhaled.”

The medical and toxicology literature reports a number of incidents involving human oral and dermal overexposure to diquat. The human clinical toxicology of diquat is discussed in Section 5.17. Since there are no diquat epidemiology investigations or worker exposure studies (Wojeck, 1983), the potential human exposure to diquat-treated water has been derived from the chemical use-rates and results of the diquat aquatic analytical studies. Risk assessments were calculated based on various types and routes of diquat aquatic exposure and findings from animal laboratory toxicology studies.

As described in Section 1 of this document, animal toxicology studies are typically designed to include low-mid-and high-dose test groups. The objective of having at least three dose groups is to determine the health effects observed and measured by the various degrees of exposure. The high dose group receives an amount of test material designed to overwhelm the body defense mechanisms; e.g., in suicide attempts, while the low dose is intended to not cause any observable or quantitated adverse health effects. The low dose or dose level that does not demonstrate toxicological effects is termed the no observable adverse effect (NOAEL or NOEL). The NOAEL is used in risk assessment calculations and sometimes is adjusted with an uncertainty factor(s) (UF) to compensate for extrapolation of calculated “safe” dose levels to humans from various animal species and toxicology endpoints, e.g. systemic toxicity, reproductive and fetal developmental effects, etc.

The diquat toxicology investigations are discussed in the following sections and the NOAELs and toxicology endpoints listed in Tables 2 and 4.

5.2.2.2 Exposure Assessment

The exposure assessment involves determination of populations that may be exposed to diquat and estimating degrees of exposure and doses likely to result from various uses of the herbicide products following aquatic weed control. Exposure parameters evaluated in this section are listed in Tables 3 and 5 and include governmental health advisories and exposure parameters.

Exposure assessment for herbicidal use assumed the maximum diquat use-rate of 2 pounds cation/acre of surface water treated for aquatic weed control. The immediate diquat cation concentration at the highest use-rate would be approximately 0.37 ppm (Reward® label, 2000). Based on the assumed use-rates, exposure calculations were conducted for swimmers and bystanders or non-swimmers as to their daily exposure to diquat. Swimmers were expected to spend anywhere from 0.5 to 3 hours swimming in diquat treated water. Their routes of exposure include dermal contact and incidental ingestion of treated water, dietary drinking of potable or treated surface water, and ingestion of fish taken from treated water. All subjects are divided into three groups consisting of 6 (22 kilogram weight) and 10 (35 kg) year olds and adults (70 kg).

The maximum exposure conditions included swimmers either drinking potable or treated surface water daily containing the maximum contaminant level (MCL) of 20 ug diquat/liter and use-rate of 0.37 ug diquat/liter. Also, the exposure from eating fish taken from diquat treated water was calculated (see Table 11). Since diquat is not intended to be used on food crops and does not bioconcentrate in living organisms, other potential dietary sources of diquat exposure were not conducted.

Tables 12 and 13 present the combined sources of diquat exposure. The main factor in the exposure to diquat following aquatic application for weed control concerned the incidental ingestion of water by the swimmer and ingestion of daily drinking water from diquat treated-surface water. Exposure and risk assessments were conducted and reported in Section 5.18, below.

5.2.2.3 Risk Characterization

The potential risk of non-carcinogenic effects is usually evaluated by comparing an environmental dose to a reference or "safe" dose. In the risk assessments for various diquat aquatic exposure doses the margin of safety (MOS) and reference dose (RfD) approaches were used.

The MOS is used to evaluate acute exposures. In this approach the lowest NOAEL or NOEL from the animal toxicology studies for specific endpoints, e.g. systemic toxicity, reproduction or developmental, were compared to the calculated human diquat doses. This method allows an evaluation and determination of a "safe" dose specific to each human route of exposure. Thus, the MOS is a ratio of the lowest NOAEL dose to the calculated exposure dose and is meant to be an indicator of potential risk. The standard MOS is 100, meaning that MOSs greater than 100 represent degrees of negligible risk, while values below 100 signal an increased risk of the toxic endpoint effects (Shipp, 1986). MOS findings are included in calculations in Tables 6-13.

The reference dose or RfD represents a lifetime "safe" dose for protection against threshold (non-carcinogenic) health effects. The RfD is considered a daily exposure level for a lifetime, where no adverse health effects are likely to occur for even sensitive human populations. Under the RfD approach UFs may be applied to the lowest NOAEL dose reported in the animal toxicology studies. A UF of 10 is generally used to estimate a "safe" human exposure level from experimental studies when there is no indication of carcinogenicity and valid human studies are available. A more conservative UF of 100 is supplied when there are few or no valid human studies available but there are valid long-term animal studies. In the case of diquat, since no human exposure studies have been conducted, the 100 UF was applied.

In the calculations in Tables 6-13 the percentage of the daily RfD is presented for the various types of exposure doses. Percentages below 100% of the RfD are considered negligible health risks while those greater than 100% serve as indicators that a potential health risk may exist at the specific exposure dose. Tables 12 and 13 include combined types of exposures and total daily diquat doses with their accompanying MOS and RfD risk assessments

5.3 CHEMICAL FORMULATIONS

The diquat product used for aquatic weed control is REWARD® Landscape and Aquatic Herbicide. The formulation contains 3.73 lbs of diquat dibromide salt (2.0 lbs of diquat cation) per gallon. Dose levels and water concentrations are defined in terms of diquat cation or ion. Diquat concentrations discussed and presented in this document are in terms of diquat ion. The diquat maximum use-rate for aquatic herbicide control is 2 pounds cation/acre maximum. The instantaneous maximum concentration is considered to be 0.37 mg cation/liter (ppm cation) and depending upon the water environmental conditions, decreases to approximately 0.l ppm after 24-hours and 0.01 ppm by 4 days (Reward, 2000).

5.4 EXPOSURE ASSESSMENT

Exposure assessments were conducted on three populations that included children and adults. The children were ages six and ten weighing 22 and 35 kg, respectively and 70 kg for adults. The population groups were evaluated according to their time spent swimming in diquat treated water containing 0.37 ppm and in some cases 0.l ppm diquat. Routes and types of exposure included incidental ingestion and skin contact with treated water. In addition, the population groups were also evaluated as to their exposure received from drinking potable water or treated surface water and daily consumption of fish taken from treated water. The exposure assessments appear in Tables 6-13.

5.5 EXPOSED POPULATION

The exposed population in this section refers to the general public and does not include people who may be occupationally exposed during mixing, loading or applying diquat to bodies of water. The exposed populations used in the exposure assessment are described in subsection 5.4 above.

5.6 POTENTIAL ROUTES OF EXPOSURE

The potential routes of exposure to the exposed population groups include primarily ingestion of diquat-treated water either during swimming or treated water as a drinking water source. Other potential routes of exposure included dermal contact of treated water and eating fish taken from the treated water.

The calculated doses received by the exposed population groups are discussed in subsection 5.18 Exposure and Risk Assessments and presented in Tables 6-13.

5.7 TOXICITY ASSESSMENT

The animal toxicology information concerning diquat is discussed in the following subsections and consists of a review of the acute, subchronic and chronic testing. An overview of the toxicology information indicates that the chemical is not considered to be a carcinogen or to cause adverse reproductive effects or birth defects (teratology). Diquat is considered to be a moderate systemically toxic chemical based on the findings in the acute and subchronic toxicology studies. However, the main adverse health effect appears to be associated with overexposure to concentrated diquat that can result in systemic renal toxicity and severe eye irritation. Results of the undiluted diquat product acute eye irritation studies place the chemical in FIFRA Toxicity Category II as causing substantial but reversible eye injury. Similarly, dermal overexposure to the concentrate may result in severe irritation and lesions resulting in the chemical entering the blood directly through damaged skin. The FIFRA acute dermal toxicity of the concentrated diquat is Toxicity Category II, based on the results of the rabbit acute dermal study. However, label-directed use of diquat for aquatic weed control and dilution and degradation of the chemical following application significantly reduces the potential for overexposure.

Although the finding of cataract formation in the diquat subchronic and chronic animal toxicology studies appears consistent in the high dose groups, this adverse health effect is not expected following exposure from aquatic or agricultural use of the chemical. When diquat is used according to label directions, exposure to doses sufficient to produce cataracts in laboratory animals is physically not possible to achieve because of the chemical's low use-rate and rapid dissipation in the environment. Further, no substantiated cases of human cataract formation have been associated with either the use of diquat or exposure to treated areas.

5.8 PHARMACOKINETICS

5.8.1 Absorption, Distribution and Metabolism

A review of the pharmacokinetics of diquat indicates that the chemical is poorly absorbed through oral and dermal routes of exposure. This can be explained, in part, by the chemical being a highly charged cation, very water soluble and highly lipid insoluble. Diquat also binds tenaciously to various substances, e.g. food, soil, suspended particulate and vegetation. A bound diquat molecule is inactive and biologically unavailable. In addition, available unbound diquat has difficulty in contributing to the dose a person may absorb because of its poor absorption across biological membranes (Coats, 1964; Grzenda, 1966, Beaseley, 1966, Yeo, 1967; Frank and Comes, 1969; Hiltibran, 1972; Saito, 1986). Early investigations demonstrated that administration of oral doses of diquat to rats revealed that approximately 95% was excreted in the feces and 5% absorbed into the system and excreted as the parent molecule in the urine within two days (Daniel and Gage, 1966).

Findings from subsequent intravenous and subcutaneous diquat dosing investigations provided evidence that none of the diquat was systemically metabolized before being excreted unchanged in the urine (Daniel and Gage, 1966; Bus, 1975). The intravenous and subcutaneous diquat dosing studies with 14C-labeled chemical, also demonstrated approximately 2% of the dose was found in the feces, indicating that biliary excretion also occurs in diquat systemic elimination (Daniel and Gage, 1966; Mills, 1976; Ameno, 1994).

Apparently, bacterial metabolism of diquat occurs to a minor degree in the gut. The metabolites are primarily excreted in the feces while a small amount becomes absorbed and eliminated in the urine (Daniel and Gage, 1966).

Results of rat dietary studies indicated that diquat was not accumulated in any of the body tissues. The animals were fed 250 ppm diquat daily for eight weeks and then regular diet for one week followed by sacrifice and analyses of tissues. No diquat was detected in any of the tissues (Litchfield, 1973). Rose conducted a tissue analytical investigation and found that diquat did not accumulate in the lungs and brain (Rose, 1976)

Diquat is also poorly absorbed through skin. There are several reasons for diquat's low degree of dermal penetration including low lipid solubility, highly ionized molecule and strong binding to melanin (Daniel and Gage, 1966; Larsson, 1977). Results of diquat human skin penetration studies further support the chemical and physical property reasons for low dermal absorption. The first human diquat dermal study found that only 0.3 % of the applied dose penetrated over a 24-hour exposure period (Feldmann and Maibach, 1974). Results of another similar diquat human skin penetration investigation revealed that 1.4% of the dermal dose absorbed through the dermal barrier. Therefore, it was concluded by the investigators that approximately 1-2% of the diquat contacting the skin (in non-irritating concentrations) penetrates into the system over 24-hours (Wester and Maibach, 1985).

Once into the system diquat is distributed throughout the body in the aqueous phase. It's slow transport across biological membranes leads to sequestration in some tissues towards eventual excretion by the kidney (Powell, 1983). As previously mentioned diquat is minimally metabolized. The metabolites monopyridone and dipyridone have been identified in both laboratory animals and humans. The metabolites are primarily formed in the liver by the microsomal cytochrome P450 enzymes. The metabolites are no more toxic than the parent compound and are excreted by the kidneys (Fuke, 1993, 1996; Gallagher, 1995).

Diquat has also demonstrated oxidation/reduction cycling involving the formation of the superoxide free radical that may lead to lipid peroxidation and cytotoxicity. Although not conclusively defined, superoxide formation may be responsible for the specific long term toxicity findings, e.g. cataract formation observed in diquat chronic animal toxicology studies (Sandy, 1987; Rikans, 1993; Awad, 1994).

5.9 SYNERGISM WITH OTHER PESTICIDES

A review of the scientific and medical literature indicates that there have been no investigations conducted to determine the potential of either diquat or its products to interact synergistically with other chemicals.

5.10 ACUTE TOXICITY

Results of diquat acute toxicology studies demonstrate that the chemical is considered to be moderately toxic and has significant eye irritation potential, see Table 1. The findings from the rat acute oral toxicity study classed the chemical in the EPA Federal Insecticide, Fungicide and Rodenticide Act (FIFRA) oral Toxicity Category III [Category I is the most toxic or irritating; Category IV is the lease toxic or irritating], while findings from the rabbit acute dermal toxicity test and the eye irritation study placed diquat in FIFRA Toxicity Category II. The results of the rabbit dermal toxicity and eye irritation investigations are designed to mimic extensive exposure to the concentrated chemical. The rabbit acute dermal toxicity study involves a 24-hour exposure under an occluded wrap followed by removal of the wrap and rinsing the skin. The animals are observed for clinical signs and deaths during the following 14-day observation period. The rabbit eye irritation investigation consists of putting a small amount of chemical in one eye of the animal and noting gross ocular changes over a 21-day observation period. Table 1 lists the acute toxicology studies, findings and the FIFRA Toxicity Categories. Based on the findings from the acute toxicology studies and the FIFRA categorization, the diquat product label warning signal word and precautionary statements are determined.

As stated in Section 1 of the WDOE Aquatic Herbicide EIS, one of the objectives of conducting acute toxicology studies is to determine the “LD50” of a chemical. The LD50 is defined as the “lethal dose” that kills or calculated to kill half or 50% of the animal test population. The LD50 is a universal guide in toxicology that commonly serves to categorize the degree of acute toxicity of a particular chemical or product. LD50s are determined for both the oral and dermal routes of exposure. LC50s (when discussing human health) are lethal defined as concentrations of airborne chemical vapor, aerosol or dust or dose of respirable chemicals that kills half or 50% of the animal test population. The LD50 and LC50 dose determines placement of a pesticide in FIFRA Toxicity Categories I –IV.

It is important to understand that the results of the acute toxicology studies reflect the health effects following overexposure to the concentrate or undiluted chemical or product. In most cases the acute toxicology studies represent the worst case exposure conditions. Once the diquat product is applied to a body of water according to its label directions for aquatic weed control, the massive dilution factor of the water the chemical’s propensity for binding to particulate, vegetation and sediment, significantly reduces the availability of diquat and significantly reducing the chances for oral, dermal, and eye overexposure.

Human overexposure to concentrated or undiluted diquat during application can result from accidental spilling or splashing of the chemical on the skin or in the eyes and mouth. Regardless of the route of overexposure, the first aid procedures outlined on the diquat product label should be followed immediately and where indicated, the exposed person should be examined by a physician.

Also, depending upon the spray equipment settings and wind factors, the applicator may be overexposed to the diquat spray mist during application of the product over the surface of the water. Overexposure to the concentrated diquat spray mist can result in irritation of the upper respiratory tract and possible systemic toxicity. However, spray application of diquat by boat involves equipment that forms large spray droplets that can be easily directed to the targeted aquatic treatment area. It is counter productive to apply smaller size spray droplets that would minimize the amount of herbicide contacting the designated treatment area and may result in formation of spray drift and the potential for diquat overexposure to the applicator or area bystanders and non-target vegetation.

5.10.1 Oral Toxicity

The findings from the diquat acute oral toxicology investigations indicate that the concentrated formulation is irritating to the stomach and intestinal tract, and depending upon the dose, can result in systemic poisoning and death. Administration of systemically toxic doses of diquat to laboratory animals produces signs of toxicity within 6-12 hours, while lesser effective doses produce signs within one-two days. Signs include decrease in body weight, reduction in water and food intake, piloerection and increased excretion of water and greenish-yellow mucoid stools. The greenish stool color is due to the bacterial reduction of diquat in the intestine. Due to the acute depletion of water, hemoconcentration may also be noted (Verbetskii and Pushka, 1968; Clark and Hurst, 1970; Crabtree, 1977; Cobb and Grimshaw, 1979).

The next phase of diquat oral toxicity is the development of a paralytic illeus. It has been noted in both animal studies and human suicide cases that fluid accumulates in the intestines and peristalsis decreases or ceases to occur. Diquat appears to have an effect on the water distribution in the body suggesting a direct action on the stomach and intestinal mucosa. Depending upon the dose, the test animals may demonstrate lethargy, collapse, coma and death (Crabtree, 1977; Cobb and Grimshaw, 1979; Schmidt, 2000).

Clinically, diquat affects the kidneys and liver as evidenced by elevation of the serum BUN, creatinine, SGOT and SGPT. Histologically, observations of the test animals revealed distention of the gastrointestinal tract, swollen caecum, ulceration and severe irritation, necrosis and exfoliation of the stomach and intestinal tract mucosal epithelium. The intestinal submucosal epithelium demonstrated infiltration of lymphocytes and polymorphonuclear and mononuclear cells. Death in the test animals was attributed to destruction of the intestinal epithelium and kidney damage (Crabtree, 1977; Cobb and Grimshaw, 1979).

Kidney damage at high dose levels of diquat have been observed in acute and subchronic animal toxicology studies and human suicide poisonings. In animal investigations and human poisoning cases it has been noted that high dose group animals demonstrated severe histological and biochemical changes that decrease renal urinary function (Lock and Ishmael, 1979; Hantson, 2000). The histopathological renal changes included renal papillary infarct, glomeruli fibrin deposits, degeneration and necrosis of the proximal and distal convoluted tubule epithelium and epithelial cell exfoliation and pycnotic nuclei (Verbetskii and Pushka, 1968; Cobb and Grimshaw, 1979; Lock and Ishmael, 1979; Hantson, 2000).

Although the liver may also be affected in diquat poisoning, only minor histopathological changes have been reported in animal toxicology studies and human suicide case reports. However, the Fischer rats appear to be more sensitive to liver toxicity effects of hepatic necrosis and oxidative damage of diquat than Sprague-Dawley rats (Smith, 1985, 1987; Tsokos-Kuhn, 1988; Spalding, 1989, Rikans, 1993; Awad, 1994; Vulimiri, 1995).

5.10.2 Dermal Toxicity

Systemic toxicity from dermal exposure to diquat has been demonstrated in laboratory toxicology studies. The signs of systemic toxicity discussed in Section 5.10.1 Oral Toxicity, also apply to findings in the animal studies. However, signs of systemic toxicity following diquat dermal overexposure in the animal dermal studies do not involve the gastrointestinal tract but primarily affect the renal function.

As listed in Table 1, diquat is not considered to be a significant skin irritant when tested in a single dose application to rabbits (Clark and Hurst, 1970; Levy, 1979). However, in acute dermal toxicology studies the chemical is applied to the skin and then covered with an impervious occlusive wrap for 24-hours. Under such test conditions the skin becomes hydrated, irritated and the dermal barrier breaks down allowing increased diquat permeability into the blood. Consequently, sufficient diquat can be absorbed through the damaged skin under occluded test conditions to result in systemic toxicity (Clark and Hurst, 1970; Levy, 1979; EPA, 1995). As mentioned previously, animal toxicology test conditions are extreme compared to the exposures encountered during label use conditions of the chemical. Although occupational cases of diquat dermal overexposure have resulted in minor skin irritation, no substantiated cases of human systemic poisoning have been reported. It has only been through ingestion of diquat that human poisoning has occurred.

Results of the rabbit acute dermal toxicity study primarily demonstrated that a 24-hour skin exposure to the concentrated test material produced severe irritation and injury allowing for toxic and lethal doses of diquat to readily pass into the blood resulting in signs of systemic toxicity. Signs of systemic involvement included decreased activity and muscle tone, diarrhea, decreased body weights and death. Findings from a gross pathological examination of the dosed animals revealed distended and discolored intestines, pale and/or mottled kidneys, lungs and liver. Table 1 lists the rabbit diquat acute dermal LD50 for male and female animals as 262 and 315 mg/kg, respectively (Clark and Hurst, 1970;Bullock, 1980).

5.10.3 Inhalation Toxicity

Inhalation overexposure to diquat spray mist during label directed use for aquatic herbicide weed control is not expected to be a significant health risk. The size of spray droplets for aquatic application must be large so that they fall directly on the intended target area. This is also to prevent human exposure and drift to non-target vegetation or other areas.

Results of the rat acute inhalation toxicity studies involved a 4-hour exposure to respirable diquat aerosol particles. The animals displayed signs of respiratory tract irritation during the exposure and the 14-day recovery periods, e.g. labored breathing, decreased respiratory rates, and increased eye and nasal secretions (Bradfield, 1980; Bruce, 1985). The investigation by Bruce (1985), revealed that signs of respiratory tract irritation and deaths occurred primarily in the high-and mid-dose groups. No gross pathological findings were found at necropsy associated with exposure to the test material. The combined-sexes diquat acute inhalation LC50 for the investigation was 0.97 mg/L, classing the chemical in EPA FIFRA Toxicity Category range of III for acute inhalation toxicity (Bradfield, 1980; Bruce, 1985).

Based on the results of the diquat rat acute inhalation studies, the type of aquatic weed control spray application and the large size of the spray droplets, it is unlikely that applicator workers or bystanders will be overexposed to diquat during aquatic herbicidal spraying.

In the animal inhalation studies the particle sizes were nearly all respirable (less than or equal to 10 microns in diameter), while typical agricultural spray equipment delivers particles in the size range of 200 microns. In the case of diquat spray application to bodies of water the particle sizes would be much greater than 200 microns for reasons described above, thus minimizing formation of respirable particles. Due to the design of the mammalian respiratory tract, particles less than 10 microns are required to reach the air sacs or alveoli of the lung. Larger particles of 200 microns and greater are deposited in the nasal passages and throat of the upper respiratory tract.

As with the dermal route of exposure, there are no substantiated reports of human diquat poisoning following acute or repeated inhalation exposure to the chemical spray mist or particles.

5.10.4 Skin Irritation

A review of the rabbit toxicology dermal irritation studies concerning diquat indicate that the formulations were only slightly irritating (Bainova, 1969; Clark and Hurst, 1970; Levy, 1979; Robbins, 1987). The results of the investigations class diquat in FIFRA Toxicity Category IV for skin irritation. The skin irritation investigations involve application of the concentrated test material to the skin for a four-hour exposure and then the skin is washed and evaluated and scored over a three-day observation period.

5.10.5 Eye Irritation

Based on the results of rabbit eye irritation studies, concentrated (undiluted) diquat is considered to be a severe eye irritant and is classed in EPA FIFRA Toxicity Category II. The findings from the diquat eye irritation study demonstrated moderate to severe conjunctival irritation that persisted beyond eight days. No irreversible conjunctival or corneal damage was observed at the end of the 21-day observation period (Bullock, 1976).

Significant ocular irritation is not expected from eye contact with diquat treated water because of the rapid dissipation of the chemical by means of dilution and binding of the chemical to particulate, vegetation and sediments.

5.10.6 Skin Sensitization

Results of the diquat guinea pig skin sensitization studies indicated that the chemical was not a skin sensitizer (Bainova, 1969; Thompson, 1985). Based on the results of the guinea pig skin sensitization investigations, the low product use rates, water dilution factor and binding and degradation of the chemical in the aquatic environment, allergic skin reactions are not expected from persons contacting diquat treated bodies of water.

5.11 SUBCHRONIC TOXICITY

Subchronic or repeated daily or weekly chemical exposures for short time periods typically occur during the application of a chemical or through dietary intake of a treated food crop or water. Most human chemical exposures are either acute (one time exposure) or subchronic (exposure to a chemical for a few days or weeks). The potential for subchronic exposure to diquat when used for aquatic weed control is unlikely since the chemical dissipates quickly over three-four days (REWARD, 2000). Significant exposure to persons in contact with recently diquat-treated water would primarily involve incidental ingestion of the chemical while swimming or drinking treated water as a daily drinking water source.

Inhalation exposures to diquat in aquatic herbicidal use situations basically applies to field applicators where possible generation of a spray mist may occur. However, aquatic application of diquat in compliance with label directions is not expected to result in adverse health effects following contact with treated water. Further, factors mitigating against any adverse health effects from applied diquat are the significant water dilution, poor dermal and gut absorption, rapid excretion of absorbed diquat and short half-life in water all of which support the conclusion that overexposure to the chemical is unlikely.

Subchronic toxicology studies are designed to determine the target organ(s) associated with overexposure to a chemical for a few weeks or months. Such studies usually consist of four groups of animals, a control (non-exposed group) and low, mid and high dose test groups. The parameters of subchronic investigations are designed to define the toxic effects of the chemical, identify the specific target organ(s) affected, note signs of toxicity, changes in body weight and food consumption, blood chemistry and urine analyses, hematology and gross and histopathological examination.

Based on the results, the target organ(s) associated with overexposure to the test compound can be identified and a no observable adverse effect level (NOAEL) dose can be determined for the chemical. The findings from the investigation can also be used for determining the degree of toxicity of the chemical, risk assessments, establishment of acceptable exposure levels, dietary and drinking water standards, label precautionary statements and other sources of health information.

5.11.1 Oral

Results of a mouse diquat ten-day gavage study demonstrated that animals in the high dose group of 10 mg/kg/dy displayed signs of decreased food consumption, weight loss, hunched posture, dyspnea, ptosis, piloerection and deaths. Sporadic weight loss and piloerection were observed in the 5 mg/kg/dy dose group animals. The NOAEL was 2.5 mg/kg/dy (Palmer, 1977).

Rats fed diets containing diquat at dose levels of 6.1 and 37.7 mg/kg/dy/ 4 wks demonstrated only increased liver weight in the high dose males. The male and female NOAELs were 6.1 and 37.7 mg/kg/dy, respectively (Colley, 1981).

5.11.2 Dermal

Bainova (1969) conducted a 20-day rat subchronic diquat dermal study with dose levels of 0, 5, 10, 20, 60 and 120 mg/kg/dy. Systemic effects that were noted at the four high dose group animals included slight skin irritation, dilatation of the gastrointestinal tract, histopathological changes in the kidney, gastrointestinal tract liver, and lung and deaths. The NOAEL was 5.0 mg/kg/dy. It appears that the animals may have been exposed orally by findings of gastrointestinal pathology.

A rat subchronic 21-day dermal investigation applying diquat doses of 0, 5, 20, 40 and 60 mg/kg/dy produced dose-related dermal irritation and skin lesions in all test groups. Mortalities occurred in the three highest dose groups in addition to systemic effects of decreased food consumption, hypothermia, hypoactivity, dyspnea, cyanosis, pale extremities and emaciation. Histopathological changes were observed in the kidneys, liver and lungs. The systemic NOAEL was 5.0 mg/kg/dy (Auletta, 1987).

A diquat rabbit subchronic dermal study involved exposure to the animals for 20-days to dose levels of 0, 20, 40, 80 and 160 mg/kg/dy. Systemic effects were observed in the 3 highest dose groups and included weight loss, unsteadiness, muscular weakness and inability to stand. Skin irritation and lesions were observed in animals in all treated dose groups. Histopathological changes associated with diquat were necrosis of the kidney distal convoluted tubules. The study systemic NOAEL was 20 mg/kg/dy (Clark and Hurst, 1970).

5.11.3 Inhalation

A rat 15-day diquat respirable aerosol inhalation study produced clinical signs of respiratory tract irritation and hematological and histological changes in the lungs of animals exposed to 2.0

mg/m3/6 hr/dy. The mid dose rats, 1.6 mg/m3/6 hr/dy, demonstrated minimal effects while the study diquat NOAEL was 0.5 mg/m3/6 hr/dy (Gage, 1968).

Rats exposed to a diquat respirable aerosol concentrations of 0, 0.32 and 1.9 mg/M3/6 hr/dy/4.5 months, demonstrated dose-related effects of biochemical and lung histopathological changes. A NOAEL was not established in the study (Bainova, 1972).

A similar rat diquat respirable aerosol exposure study exposed animals to dose levels of 0, 0.4, 0.7 and 1.9 mg/M3/4 hr/dy/4 months. The investigator reported that signs of respiratory tract irritation and signs of toxicity were dose-related and observed in all exposed groups (Makovskii, 1972).

A more recent diquat rat subchronic inhalation study involved animals exposed to respirable aerosols in concentrations of 0.1, 0.49, 1.1 and 3.8 mg/M3/6 hrs/dy/5 dys/wk/3 wks. Treatment-related effects, observed in the three highest test groups, included decreased reticulocytes and platelets, increased lung weights, lung/body weight and lung/brain weight ratios, mottling and reddening of the lungs and lung lesions (multi-focal chronic interstitial pneumonia and alveolar macrophages). A recovery period of 21-days for all animals demonstrated that the diquat related effects remitted except mottling and reddening of the lungs. The study diquat NOAEL was 0.1 mg/M3/6 hrs/dy/5 dys/wk/3 wks (Bruce, 1985).

5.11.3 Neurotoxicity

Results of both acute and subchronic rat neurotoxicity studies demonstrated that diquat was not neurotoxic when evaluated by functional observation battery (FOB) and motor activity testing and histopathological examination.

An acute rat neurotoxicity investigation was conducted to determine the delay neuropathy potential of diquat. Rats were administered diquat doses of 0, 25, 75 and 150 mg/kg and then assessed by means of FOB and motor activity testing at 6 hours, 8 and 14 days. At the end of the 14-day observation period the animals were sacrificed and a histopathological examination of the nerve tissue conducted. Animals in the 150 mg/kg group demonstrated signs of toxicity that included piloerection, diarrhea, urinary incontinence, nose and mouth staining and abnormal gait and posture. One animal in the high dose groups was sacrificed *in extremis* on day six. The males were less affected by the doses of diquat than the females. Diquat was not considered to be a nerve toxin based on the negative histopathological findings and the clinical observations. The NOAEL for systemic toxicity and neurotoxicity was 75 and greater than 150 mg/kg, respectively (Horner, 1992a).

The results of a rat subchronic dietary neurotoxicity study revealed that animals fed diquat daily for 14 weeks did not develop any clinical signs or histopathological changes consistent with nerve injury or damage. Male animals received diquat dietary doses of 0, 1.6, 8.0, and 32.4 mg/kg/dy, while the females were fed dose levels of 0, 1.9, 9.5 and 38.5 mg/kg/dy. Potential neurotoxicity effects were evaluated by FOB and motor activity testing and neurohistopathology. Signs of toxicity were present in the high dose group animals and included decreased body weights and cataracts. Diquat was not considered neurotoxic based on the negative findings of the FOB and motor activity testing and histopathological examination. The NOAEL for systemic effects was 8.0 and 9.5 mg/kg/dy for the males and females, respectively. The NOAEL for neurotoxicity was greater than 32.4 and 38.5 mg/kg/dy for male and female rats, respectively (Horner, 1992b).

5.11.4 Immunotoxicity

A review of the animal toxicology studies and scientific and medical literature does not indicate that any specific diquat immunotoxicity investigations have been conducted. However, based on the negative findings from the subchronic, chronic, multigenerational reproduction and mutagenicity toxicology studies, it seems unlikely that diquat has significant immunotoxic potential.

5.11.5 Estrogen Disruption

There is no evidence or results from any toxicology investigation that demonstrates either exposure or overexposure to diquat results in any findings or changes associated with adverse endocrine function or mimicking effects. The results of the diquat teratology and reproduction studies did not provide any evidence that the chemical demonstrated teratogenic potential or reproductive changes. Similarly, results of the subchronic and chronic investigations did not reveal any findings of behavioral, clinical or histopathological changes considered to be associated with adverse endocrine effects.

5.12 CHRONIC TOXICITY

The chronic or lifetime exposure effects from diquat have been evaluated in the mouse, rat and dog. The findings from the investigations are consistent in that long-term exposure at high-and mid-diquat dose levels primarily resulted in the development of cataracts. Diquat did not demonstrate evidence of carcinogenicity in the studies.
A rat lifetime dietary study involved feeding diquat daily in doses of 0, 5, 15, 75 and 375 ppm or EPA lifetime estimated doses of 0, 0.19, 0.58, 2.9l and 14.88 mg/kg/dy for males and 0, 0.24, 0.72, 3.64 and 19.44 mg/kg/dy for females. Cataracts were observed in a few high dose animals by 13 weeks and in nearly the entire group by 1-year. At the end of the two-year study, cataracts were identified in the three highest dose groups. Because of poor survival in the high dose groups, thus limiting the statistical power of the study, it was questionable whether the incidence of 2/42 cataracts found in the 15 ppm (0.58 and 0.72 mg/kg/dy) group was significant. In addition to cataracts and lens opacities, the animals also demonstrated decreased renal clearance (Colley, 1985).

Discrepancies exist between the California Department of Pesticide Registration (CDPR) and the EPA as to the systemic NOAEL of the Colley, 1985, diquat rat feeding study. CDPR is of the opinion that the NOAEL for the study is 5 ppm (combined dose 0.22 mg/kg/dy) based on the absence of cataracts in the low dose group (DPR, 1994). The EPA did not find a statistically significant increase in the 15 ppm (combined dose 0.65 mg/kg/dy) group using the paired t-test and claimed the study NOAEL to be 0.65 mg/kg/dy (USEPA, 1995). However, the EPA's calculation of the diquat reference dose (RfD) uses the 0.22 mg/kg/dy as the NOAEL (USEPA IRIS Substance File, 1995).

Hodge (1991) conducted a mouse diquat lifetime feeding study using doses of 0, 30, 100 and 300 ppm. Results of the 104 week investigation indicated that animals receiving the mid and high doses, demonstrated slightly increased (but insignificant) kidney weights associated with kidney tubule dilation in males, while the females had increased kidney tubule hyaline droplets. Eye discharge was also noted in the two high dose groups. No evidence of carcinogenicity was observed in the histopathology. The NOAEL for systemic toxicity was 30 ppm (calculated lifetime dose of 3.56 and 4.78 mg/kg/dy for male and female mice, respectively (Hodge, 1991).

Another mouse 80-week diquat feeding study revealed that animals dosed with 150 ppm of the chemical developed reduced growth rates and liver vacuolation. No carcinogenic findings were noted. The NOAEL was 30 ppm or 4.5 mg/kg/dy (Ben-Dyke, 1975)

A 52-week diquat dog feeding study demonstrated the development of cataracts in animals fed 12.5 mg/kg/dy and in one female in the mid dose of 2.5 mg/kg/dy. Other adverse health effects observed were inflammatory lesions of the intestinal epithelium and increased kidney weights in the high dose and decreased adrenal and epididymal weights in the male mid dose dogs. The systemic NOAEL was 0.5 mg/kg/dy (Hopkins, 1990).

Hurst (1966) conducted a 2two-year dog study where diquat was administered at doses of 0, 1.7, 5.0 and 15 mg/kg/dy. At the end of the study one dog from each group was sacrificed and the investigation continued for another two years. Results of the study found that cataracts developed in the high dose animals in less than a year and in the mid dose by 15-17 months. No other effects were noted. The systemic NOAEL was 1.7 mg/kg/dy (Hurst, 1966).

5.13 DEVELOPMENTAL AND REPRODUCTIVE TOXICITY

A review of the diquat reproduction and teratology toxicology investigations, did not reveal any evidence that diquat has been associated with any significant findings of reproduction dysfunction or teratological effects.

5.13.1 Teratology Studies

Results of a diquat rat developmental investigation demonstrated that signs of maternal toxicity were displayed in the mid and high dose groups animals administered 12 and 40 mg/kg/dy on gestation days 7 through 16, repectively. Maternal signs of toxicity were displayed by reduced food consumption and decreased weight gain. The maternal toxicity NOAEL was 4.0 mg/kg/dy. The offspring exhibited decreased weight gain, delayed skeletal ossification and hemorrhagic kidneys in the high dose group. The developmental NOAEL was 12 mg/kg/dy (Wickramaratne, 1989).

A teratology study involving mice dosed with diquat at doses of 0, 1, 2, and 4 mg/kg/dy during gestation days 6 through 15 demonstrated signs of maternal toxicity at the two high dose levels. Dams exhibited signs of piloerection, dyspnea, respiratory noise, hunched posture, decreased body weight gain and deaths. The maternal NOAEL was 1 mg/kg/dy. Developmental effects were noted at the two high dose levels displayed as decreased fetal body weight, skeletal anomalies, exencephally, premature opening of the eyes, and umbilical hernia. The diquat developmental NOAEL for the study was 1 mg/kg/dy (Palmer, 1978; DPR, 1994; EPA, 1995).

A rabbit diquat developmental study involved administering doses of 0, 1, 3, and 10 mg/kg/dy through gestation days 7-19. Animals were sacrificed on day 30. In the high dose animals there was significant maternal mortality and signs of toxicity demonstrated by decreased body weight gain, stomach ulceration and histopathological changes in the liver and intestines. The mid dose dams demonstrated similar effects to a lesser degree. No maternal toxicity was noted in the low dose rabbits that was considered the NOAEL. Developmental effects were noted in all dose levels, however CDPR and USEPA have different interpretations of the study endpoints. The CDPR interpretation following their review of the data indicates that developmental effects occurred at all dose levels. USEPA concluded from their review that there were a significant increased incidence of developmental changes that occurred only in the high dose group. Based on the EPA conclusion, the maternal toxicity and developmental NOAELs for the study were l and 3 mg/kg/dy, respectively (Hodge, 1989; DPR, 1994; USEPA, 1995).

5.13.2 Reproduction Studies

A rat reproduction study involved dietary dosing of male and female animals with diquat at concentrations of 0, 16, 80 and 400/240 ppm (0.8, 4 and 20/12 mg/kg/dy) for 12 weeks (Fo generation) prior to mating and was continued through the weaning of the offspring. Due to the adverse systemic toxicity to the F1 high dose animals the dose was decreased to 240 mg/kg/dy. Adverse parental effects were noted in the high dose group animals of the next generation as evidenced by red/brown urine, piloerection, oral ulceration, decreased body weight gains and ocular changes, e.g. opacity, partial or total lenticular cataracts. Adverse reproductive toxicity effects were observed only in the high dose group animals in both generations displayed by decreased numbers of live F1 pups/litter and decreased body weight gains. The diquat systemic and reproductive toxicity NOAELs were 80 ppm (4 mg/kg/dy) (Hodge, 1990).

Griffiths (1966) and Fletcher (1972), conducted diquat 3-generation rat studies where the animals were dosed at concentrations of 0, 125 and 500 ppm (0, 6.25 and 25 mg/kg/dy). Both investigators noted decreased body weight gain and cataract formation in the high dose animals in the parental generations. The systemic and reproductive toxicity NOAELs for both studies was determined to be 125 ppm or 6.25 mg/kg/dy.

5.14 MUTAGENIC EFFECTS

Diquat has demonstrated both positive and negative findings in mutagenesis testing. The chemical was negative in the Ames assay both with and without metabolic activation (Andersen, 1972; Benigni, 1979; Levin, 1982). However, diquat caused forward mutations, increased thymidine kinase in the mouse lymphoma cell plate assay and decreased the mitotic index and increased chromosome aberrations in human lymphocyte tests (Bignami and Crebelli, 1979; DPR, 1994; USEPA, 1995; Richardson, 1986)

Diquat was negative in a mouse micronucleus test where the animals were dosed with 62.5 and 100 mg/kg of the chemical. Examination of the bone marrow provided evidence that a cytotoxic effect to the marrow cells occurred at both dose levels (McGregor, 1974).

An *in vitro* human blood lymphocyte chromosome aberration study was conducted using varying concentrations of diquat. A dose-related effect was demonstrated. Diquat was not clastogenic at concentrations of 12.9 and 25.8 ug/ml with metabolic activation and also without activation at the low dose. However, dose levels of 64.5 and 129 ug/ml demonstrated increased chromosomal damage (Wildgoose, 1986).

An *in vivo* dominant lethal study in mice receiving diquat doses of 0, 0.1, l.0 and 10 mg/kg/dy/5dys was negative (Sheldon, 1986). Findings from the Sheldon study confirmed negative results observed in previous diquat dominant lethal investigations (Pasi, 1974; Pasi and Embree, 1975; Anderson, 1976; Selypes, 1980).

A rat *in vivo* unscheduled DNA synthesis study was conducted involving animals dosed with 0, 225, 450 and 900 mg/kg diquat. The rats were sacrificed and liver hepatocytes examined for any signs of unscheduled DNA synthesis. Although there was evidence of diquat associated hepatocyte cytotoxicity, examination of remaining normal liver cells showed no increase in unscheduled DNA synthesis (Trueman, 1986).

It appears that diquat has genotoxic potential at dose levels associated with cellular toxicity. In addition, due to diquat being highly ionized and having poor lipid solubility, transport across cell membranes at low dose levels may reduce the potential to cause cellular genetic injury or damage.

5.15 CARCINOGENICITY REVIEW

Based on the toxicology database, findings from the three diquat chronic feeding studies and several mutagenicity investigations, there is no definitive evidence that diquat is carcinogenic. The findings from the chronic toxicology investigations discussed in Section 5.12 indicate that the long-term toxic endpoint from diquat overexposure was primarily the development of cataracts. There was a statistically non-significant finding of osteosarcomas in the rat lifetime feeding study.

The carcinogenic potential of diquat was evaluated by the Health Effects Division Reference Dose (RfD)/Peer Review Committee. The committee concluded that diquat was not a carcinogen and classified the chemical in Group E (evidence of noncarcinogenicity for humans (EPA, 1994).

5.16 EPIDEMIOLOGY REVIEW

A review of the scientific and medical literature provided no citations that any epidemiological investigations concerning diquat have been conducted.

5.17 HUMAN CASE REPORTS AND STUDIES

There are no substantiated reported cases of human systemic diquat poisoning from either skin or inhalation exposure to the chemical. Systemic diquat poisoning and deaths have occurred when either toxic or lethal amounts of the chemical were ingested. These incidents have involved mostly suicidal and some accidental diquat ingestion cases. Signs and symptoms associated with skin and inhalation overexposure to diquat concentrate or spray mist are primarily displayed by manifestation of tissue irritation (Schonborn, 1971; Crabtree, 1977; Vanholder, 1981;Van Den Heede, 1982; Pond, 1983; Hantson, 2000; Schmidt, 2000).

Depending upon the degree and duration of overexposure, dermal contact with diquat concentrate may result in severe skin irritation, while prolonged contact with diluted spray may cause slight irritation. The absence of systemic poisoning associated with diquat dermal exposure either in occupational or contact through aquatic herbicidal use, is due to the poor skin penetration of the chemical and low product label use-rates (Clark and Hurst, 1970; Feldman and Maibach, 1974; MacGregor, 1979; Wojeck, 1983; Scott, 1985, 1988). Fingernails or toenails exposed to diquat concentrate will crack and shed, followed by normal regrowth. Nail contact with dilute diquat spray may lead to the formation of white spots (Samman and Johnson, 1969; Clark and Hurst, 1970; Baran, 1974).

When diquat is used for aquatic weed control, persons exposed to the treated water would not be expected to develop dermal irritation or finger or toe nail effects because of the huge dilution factor and the low use rate of the chemical.

Agricultural spray equipment produces large droplets, in the range of 200 microns, that when inhaled are deposited in the upper respiratory tract, e.g. the nasal passages, throat and upper trachea. Therefore, depending upon the concentration of the diquat spray mix and degree and duration of overexposure, signs and symptoms of upper respiratory tract irritation may be evidenced by nose bleeding, excessive nasal discharge, sneezing, coughing, sore throat, headache and nausea. The symptom of nausea may result from swallowing a sufficient amount of saliva containing diquat to cause minor irritation of the stomach mucosa. Once a person wears protective breathing equipment or is removed from the exposure situation, the signs and symptoms of diquat inhalation overexposure remit. Spray particle sizes necessary to reach the lung alveoli or air spaces must be in the respirable range of <10 microns (Swan, 1969; Wojeck, 1983). Diquat surface water spray applications for aquatic weed control use even larger droplets than used in terrestrial agricultural situations in order to ensure direct application to the target vegetation and prevent drift to nontarget areas. In summary, there have been no substantiated cases of human diquat inhalation overexposure resulting in either lung damage or systemic poisoning (Gage, 1968; Swan, 1969; Clark and Hurst, 1970; Bruce, 1985, 1987).

In addition, diquat has a very low vapor pressure of <1.0 x 10-5 mmHg @20oC, therefore no systemic toxicity, skin or eye irritation or respiratory tract irritation effects are expected from exposure to diquat vapor (Pack, 1987).

It should also be noted that overexposure to diquat spray mist may also cause mild irritation to the eyes. The results of the animal acute eye irritation studies and human case reports indicate that diquat concentrate can cause severe eye irritation. However, in the case of eye contact with water treated with diquat for aquatic weed control, minimal eye irritation is expected because the low product use-rate and huge dilution factor would reduce the amount of chemical contacting the eye. For the reasons discussed above, there is no label water re-entry restriction for swimming in diquat treated water (Cant and Lewis, 1968a,b; Swan, 1968; Rittenhouse, 1976; Nirei, 1993).

5.17.1 Human Neurological Case Reports

There are no reports in the scientific and medical literature listing any findings that either animal or human overexposure to diquat has resulted in adverse affects to the central or peripheral nervous systems. However, there are reports in some human poisoning cases involving ingestion of large amounts (1-2 ounces) of diquat concentrate, where a few patients displayed signs of central nervous system effects, e.g. agitated and combative behavior, stupor, coma. Patients surviving the poisoning that had demonstrated signs of neurological involvement recovered completely. The reasons for the clinical neurological effects are not understood, do not occur in all cases of human diquat poisoning and cannot be duplicated in animal toxicology studies. Based on the weight of the evidence from the diquat toxicology investigations, occupational use and clinical findings, the chemical is not considered to be a neurotoxin. No neurological effects have been reported from incidental ingesting of spray dilution or swallowing of diquat treated water while swimming or in situations where the treated water was used as a drinking water source (Swan, 1969; Schonborn, 1971; Crabtree, 1977; Vanholder, 1981; Van Den Heede, 1982; Pond, 1983; Wojeck, 1983; Hantson, 2000; Schmidt, 2000).

5.17.2 Human Reproduction Case Reports

A review of the diquat animal toxicology studies failed to demonstrate any findings of adverse developmental and reproductive effects (Griffiths, 1966; Fletcher, 1972; Palmer, 1978; Hodge, 1989 Wickramaratne, 1989). Further, no reports were found in the scientific and medical literature associating exposure to diquat with any human teratology or reproductive dysfunction.

5.17.3 Chronic Toxicity

A review of the medical and scientific literature did not provide any findings or reports that chronic diquat exposure occurs when the product is used according to label directions. Subchronic exposure over several days following application of diquat would not be expected to result in systemic toxic effects primarily due to the difficulty involved in absorbing a sufficient dose from contacting treated vegetation, soil and water where the chemical is either significantly bound or diluted. Secondly, diquat is not highly toxic and poorly absorbed through the skin. Thirdly, the kidneys rapidly eliminate any diquat that enters the system.

5.18 EXPOSURE AND RISK ASSESSMENTS

The exposure and risk assessments associated with diquat use as an aquatic herbicide are presented in the following Tables, concerning persons engaged in swimming, drinking both potable and treated surface water and eating fish from water where the chemical has been applied. The different types of daily exposures and risk assessments were calculated for both individual and combined scenarios. Based on the estimated diquat exposures, the risk assessments were determined by the margins of safety (MOS) and the % of the reference dose (RfD). The diquat calculations were conducted using the label maximum use-rate of 2 pound cation/ surface acre. The label states that the instantaneous diquat ion concentration in the treated water following application is 0.37 ppm or 0.37 mg cation/Liter or 0.37 ug/ml. The diquat label claims that the concentration of the chemical decreases to approximately 0.1 ppm by 24-hours and then to 0.01 in 4 days (REWARD, 2000). The calculations presented in the following Tables concerning diquat estimated exposures and risk assessments are based on the initial 0.37 ppm diquat water concentration, time spent swimming ranging from 0.5 to 3 hours and body weights of 22, 35 and 70 kgs for 6 and 10 year old children and adults, respectively.

Table 2 lists the quantitative toxicology parameters and diquat product use-rates that were utilized in conducting the exposure and risk assessments. EPA has determined that the diquat RfD is 0.0020 mg/kg/dy, based on the NOAEL of 0.22 mg/kg/dy finding for minimal lens opacities and cataracts in the rat 2-year chronic feeding study, (Table 4, Colley, 1985). The acute dermal toxicology NOAEL of 100 mg/kg was not used in the calculations since even using the immediate water concentration of 0.37 ppm diquat, demonstrated that dermal exposure accounted for only 1-2% of the total diquat exposure, (Table 8, Lees, 1996). The diquat dermal parameters of an approximate skin absorption rate of 0.3%/24 hours, permeability coefficient and flux rates were used to calculate herbicide dermal exposures from contacting herbicide treated water, see Tables 6-8 (Corrigan and Scott, 1989; USEPA, 1993; Lunchick, 1994).

The diquat products have label use-rate ranges of 1 to 4 pounds of cation/surface acre (0.08 – 0.37 ppm or mg/L or ug/ml) depending upon the intended target vegetation to be eliminated. Therefore, based on the use-rates, the exposure estimates and risk assessments were conducted for persons exposed to water containing 0.37 ppm diquat for the first 24 hours followed by a water concentration of the chemical of 0.1 ppm on day 2. The amount of available diquat continues to decrease so that by day 4 the water would contain <0.01 ppm of the chemical (Reward®, Landscape and Aquatic Herbicide –Label).

Since diquat is applied by subsurface injection or sprayed directly over the surface of water for aquatic weed control, based on the above discussion, it is highly unlikely that the spray or vapor will drift to expose either applicators or bystanders. For these reasons applicator or bystander diquat exposure or risk assessments were not conducted.

Exposure estimates and risk assessments to diquat resulting from aquatic weed control were determined for cases involving ingestion and dermal contact with treated water while swimming, drinking potable water containing the maximum contaminant level (MCL) of 0.02 mg/L/day, daily drinking of treated surface water and ingestion of fish from treated water. These various calculations are presented in the following Tables.

Table 3 lists the EPA health advisories for diquat in drinking water (EPA, 1995). The exposure and risk assessment for ingestion of potable water utilized the maximum contaminant level (MCL) of 0.02 mg/L for diquat daily exposure for 10, 35 and 70 kg persons. Since the chemical is not registered or intended for food crop use and the amounts of diquat found and calculated from eating fish was negligible, other potential dietary sources of exposure were not included in the calculations.

One of the purposes of conducting a review and health risk assessment for use of diquat as an aquatic herbicide is to determine whether swimming or contacting water that has been treated according to product label directions, should be a health concern. The situation is discussed in this section and the exposure dose and risk assessment calculations presented in Tables 5-7.

However, before addressing the exposure to diquat treated water, it is important to understand the toxicity classification of undiluted diquat according to the results of the acute toxicology studies summarized on Table 1. As indicated in Table 1, the results of the diquat product acute oral and dermal animal toxicology studies demonstrate that the chemical is not considered highly toxic and classed for both routes of exposure in FIFRA toxicity categories III and II, respectively. A review of Table 1 reveals that one of the most significant findings was that the concentrated diquat formulation is classed as a FIFRA category II eye irritant. The results of the eye irritation studies indicated severe irritation and substantial but temporary eye injury. Section 5.10 Acute Toxicity of this document, discusses how the studies are designed to maximize the skin and eye irritation potential.

One of the reasons for conducting the acute toxicology studies is to provide results for determining the label signal word warning and precautionary and first-aid statements based on FIFRA toxicity criteria. Thus, the findings from the diquat acute toxicology studies dictate the "WARNING" label signal word and the precautionary statements:

> *"WARNING: May be fatal if absorbed through skin. Harmful if swallowed or inhaled. Causes substantial, but temporary eye injury. Causes skin irritation. Contact with irritated skin, or a cut, or repeated contact with intact skin may result in poisoning*."

Although the diquat products in their undiluted form are severe eye irritants, once the products have been applied to water according to label directions, they become diluted by the huge water volume, absorb into vegetation, begin degradation and bind to suspended particulate and the sediment. The decrease in the amount of diquat in the treated water reduces the amount of chemical available for exposure, thus decreasing the potential for systemic toxicity and eye irritation. The following discussion reviews the diquat label dilution use-rate water concentration of 0.37 ppm and the calculated exposure doses and associated health risk assessments to swimmers and others receiving various types of exposure to the aquatically applied herbicide.

The diquat products are applied at prescribed label use-rates in terms of gallons of product/acre of water in order to obtain the specific herbicidal concentration to eradicate the targeted aquatic weed. As mentioned previously, the use-rates vary depending upon the type of vegetation and the size of the area to be treated.

The exposure and risk assessment parameters regarding persons swimming in diquat treated water are presented on Tables 5-8. Calculation of diquat exposures utilized the swimmer's weight (kg), the skin surface area available for exposure (cm2), the amount of time (hours) spent in the treated water containing 0.037 and 0.1 ppm diquat, amount of water swallowed while swimming over specific time periods and the human skin permeability coefficient.

Tables 6-8 list the estimated diquat oral and dermal exposures and risk assessments for swimmers. The exposure conditions are defined in Table 5. Based on the calculations, it appears that the greatest diquat exposure occurs from incidental ingestion of water while swimming. It is estimated that a swimmer swallows approximately 50 ml (nearly 2 ounces) of water/hour (Lunchick, 1994). As listed in Table 8, approximately 97-99% of diquat exposure while swimming occurs through incidental ingestion of water.

A review of Table 6, indicates that the worst case exposure situation involves the six year old group that spend three hours swimming in water containing 0.37 ppm diquat. The diquat risk assessments, using the margin of safety (MOS) of 220 ug/kg/dy and the % of the reference dose (RfD) of 2.0 ug/kg/dy, for the 6 year old group calculates a MOS of 88 and exceeds the daily RfD by 125%.

Based on label statements, the water concentration of diquat becomes significantly reduced to 0.1 ppm 24-hours after herbicidal application. Results of risk assessment calculations based on the 24-hour diquat water content, indicated no significant chemical exposures in any of the age groups swimming for three hours/day.

A review of Tables 7 and 8 reveals that the swimmers' dermal diquat exposures are insignificant compared to the oral exposures as supported by the large MOSs and low RfDs.

A review of Table 8 indicates that the dermal route comprises only 1-3% of the total swimmer exposure to diquat.

Diquat exposure through ingestion and dermal contact with sediment is considered insignificant because of the chemical's high water solubility, rapid adsorption by aquatic plants and tenacious binding to particulate matter and sediments. The persistence of diquat in water is minimal due to these properties. Therefore, exposure to diquat from either ingestion or contact with sediments has not been calculated as part of the exposure and health risk assessment evaluation in this document. As a further note, because of diquat's high binding capacity to soil, clinically, slurries of bentonite clay or activated charcoal are administered to patients who have swallowed the chemical in order to prevent systemic poisoning. Once diquat is bound to clay particles in the gut it is biologically unavailable and cannot be absorbed into the system (Coats, 1966; Malquori, 1966; Hiltibran, 1972; Simsiman, 1976; Corwin, 1985; Fugie, 1988; Dyson, 2000; Schmidt, 2000; Hantson, 2000).

Diquat has a vapor pressure of <4.0 x 10-9 mmHg @ 25°C, therefore any vapor emitted from treated water is considered to be an insignificant route of exposure (Pack, 1987). No exposure or risk assessments regarding swimmer or bystander exposure to diquat vapor from treated water has been included in this document. Since occupational exposure to diquat during mixing, loading and applying the chemical is also beyond the scope of this document, this route of exposure is not discussed. As mentioned previously, inhalation of diquat spray is not considered to result in significant inhalation exposure since aquatic spray equipment applies large droplets designed to target specific vegetation and not drift out of the treatment area.

Table 8 is a compilation of the various types of exposure and total estimated diquat doses that a person may receive when swimming in treated water containing either 0.37 or 0.1 ppm of the chemical. The incidental ingestion of water while swimming is the most significant route of exposure. In all cases, approximately 97 - 99% of the total diquat exposure that occurs while swimming is through the oral route. The same situation discussed above concerning exposure and risk assessments for oral exposure to diquat while swimming also applies in the case of the combined chemical exposure where the dose (ug/kg/day) and the MOS and RfD for the six-year-old group swimming for three-hours in treated water containing 0.37 ppm diquat are similarly affected. The MOS and RfD for the same group does not exceed the risk assessment parameters when the diquat level decreases to 0.1 ppm 24-hours after application. Although the risk assessment for the six-year-old group swimming for three-hours immediately following diquat herbicidal application, exceeds the MOS and RfD, the exposure is not greater than the animal systemic toxicity NOAEL of 2,000 ug/kg/day.

At this point it is important to evaluate any adverse health effects that could potentially be associated with exposure to diquat while swimming in treated water. The largest acute exposure (single-day exposure) would be expected to occur on the day of the diquat product application. Once applied, the concentration of the chemical will decrease to approximately 0.1 ppm within 24-hours, thus reducing the daily dose. (See diquat half-life discussion in Table 5.)

Extensive eye contact with water containing 0.37 ppm diquat may result in the swimmer experiencing some possible chemical-associated temporary minor eye irritation or conjunctivitis. Even eye contact with water not containing diquat will result in some minor eye irritation. However, as diquat decreases in the water, minor potential eye irritation is not considered to be a significant problem.

In summary, it appears that persons swimming in water treated with the highest use-rate of diquat of 0.37 ppm are not expected to experience significant adverse health effects. Based on results of toxicology studies, product use-rates and diquat binding properties, systemic toxicity seems unlikely unless a 10-year-old child drank a massive amount of water, e.g. ~3.5 gallons, containing 0.37 ppm diquat during a 3-hour swimming period. A 22 kg child ingesting 3.5 gallons of treated water would equal the NOAEL dose of 220 mg/kg/dy. Also, some minor eye irritation may be associated with prolonged contact with the 0.37 ppm treated water; however any redness would remit within a day. The wearing of swim goggles or other eye protection may be useful in avoiding potential minor eye irritation.

Drinking water sources are significant factors in determining the overall exposure to diquat particularly during the first day following application. Table 9 demonstrates that potable drinking water containing the established diquat MCL of 20 ug/L does not pose a significant exposure or health risk. The calculations assume daily ingestion of 1-2 liters of water containing 20 ug of diquat for 6 and 10-year-old children. The MCL represents the diquat concentration in drinking water that is not expected to cause any adverse noncarcinogenic health effects. A 10-year-old child would need to drink approximately 65 gallons/day of water containing 20 ug/L to reach the MCL.

Conversely, Table 10 demonstrates that ingestion of drinking water from treated diquat surface water significantly increases the estimated daily exposures and adversely affects the calculated risk assessments. Based on the calculations, ingestion of diquat-treated water as a daily drinking water source during the 24 to 36 hours post application can result in dose levels that exceed the MOSs and RfDs. Therefore, a three-day waiting period as indicated by calculations in Table 10 may be advisable.

Consumption of fish taken from diquat treated bodies of water appears to be a minor potential dietary source of exposure to the chemical. Hamer (1987), found that diquat did not bioconcentrate in bluegill sunfish. However, a report by Carter, 1971, notes the detection of 25 ug/kg in the edible portion of fish. Using Carter's analytical finding in calculating the exposure doses in Table 11, it does not appear that ingestion of fish from treated water poses any significant health problem or risk.

The total daily-calculated diquat exposures are presented in Tables 12 and 13. The difference in the two tables concern potable vs. the diquat-treated surface water as sources of daily drinking water. Apparently, there are locations that receive residential drinking water directly from treated water sources, e.g. ponds, lakes and rivers. Since ingestion of water accounts for the greatest potential exposure to diquat, both sets of exposure estimates and risk assessments are presented. The total daily exposures listed are the summation of diquat daily dose levels received from swimming (Table 8), type of drinking water source (Tables 9 and 10) and ingestion of fish (Table 11).

The findings in Table 12 concerning a summation of the daily estimated diquat exposures and risk assessments that include potable water and essentially mimics the calculations presented in Tables 6 and 8. Again, the significant exposures are to the six and ten-year-old children swimming in 0.37 ppm diquat treated water for two and three hours, respectively. However, approximately 24 hours following herbicide application, the six year old child swimming for three hours in water containing 0.1 ppm diquat does not exceed the total MOS and RfD risk assessment parameters.

Examination of Table 13 reveals a different situation whereby the MOSs and RfDs are exceeded in all age/weight groups. The daily dose levels resulting from ingestion of diquat treated surface water at both the 0.37 and 0.1 ppm concentrations significantly increases the daily dose in all subjects and swimming exposure times. Although the calculations represent significantly elevated diquat exposures, the doses still remain below the lowest animal chronic toxicology study NOAEL dose of 220 ug/kg/dy. Nevertheless, as stated above, it is important to follow the label directions for domestic water use of diquat treated water and wait three-days following application of the chemical before resumption of using the treated water for daily drinking purposes.

5.19 CONCLUSION

Based on a review of the diquat chemical and physical properties, use-rates, rapid removal from the aquatic environment by adsorption to particulate, vegetation and sediments, toxicology studies, biotransformation, exposure estimates and risk assessments, it appears that the label directed use of the herbicide for aquatic weed control purposes is not expected to result in any significant adverse health effects. The exposure evaluation of persons swimming in water containing 0.37 ppm (highest label use-rate) and 0.1 ppm (24-hour dissipation level) diquat does not indicate that significant adverse systemic health effects would occur based on the exposure and risk assessment parameters. The only health-based screening level exceeded involved the six-year-old group swimming for three hours in water containing 0.37 ppm diquat. Even though the health-based screening level was exceeded in this case, the potential diquat dose calculates to approximately 88 times less than the systemic MOS based on the animal chronic toxicology NOAEL of 220 ug/kg/day.

> *Risk assessments were significantly exceeded in situations where the source of drinking water involved diquat treated water during the 24 to 36 hours following aquatic application. Although the calculated diquat doses were elevated they remained 13 - 76 times below the systemic animal chronic toxicology NOAEL. Again, it is important to follow the diquat product label that recommends waiting three days before resuming use of treated water as a drinking water source.*
>
> *Based on the label use directions and the results of the diquat toxicology studies, the aggregate or combined daily exposure to the chemical from aquatic herbicidal weed control does not pose a significant adverse health concern.*

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LIST OF TABLES

Table 1: Diquat Acute Toxicology

Study Type	**Results (mg/kg)**	**Toxicity Category**	**Reference**
Acute Oral Rats	LD50 M= 810 F= 600	 III III	Rittenhouse, 1979
Acute Dermal Rabbits	LD50 M= 262 F= 315	 II II	Bullock, 1980
Acute Inhalation Rats	LC50 **Combined = 0.97 mg/L** M= 0.80 F = 1.90	 III III III	 Bradfield, 1980 Bruce, 1985
Dermal Irritation Rabbits	Slight	IV	Clark, 1970 Levy, 1979
Eye Irritation Rabbits	Slight to Severe	II	Bullock, 1976
Skin Sensitization Guinea Pigs	None		Bainova, 1969 Thompson, 1985

Table 2: Diquat Toxicology Quantitative Parameters

Regulatory Guideline	Standard/Dose (mg/kg/dy)	Reference
Toxicology		
Acute Rabbit Dermal NOEL	100 mg/kg	(Lees, 1996)
Subchronic NOEL	5.0 mg/kg/dy/21 dys	(Bainova, 1969; Auletta, 1987)
Chronic NOEL - Dog	0.5 mg/kg/dy	(Hopkins, 1990)
Chronic RfD	0.005 mg/kg/dy or 5.0 ug/kg/dy	(EPA, 1995)
Reproduction NOEL - Rat	4.0 mg/kg/dy	(Hodge, 1990)
Cancer Classification		
EPA	Group E (evidence of noncarcinogenicity in humans)	
IARC	Not Evaluated	
Dermal		
Absorption = ~0.3%/ 24-hours - Human (Feldman and Maibach, 1974)		
Permeability Coefficient = 5.8 x 10-5 cm/hr (Corrigan and Scott, 1989)		
Flux Rate (0.37 ppm use-rate) = 2.14 x 10-5 mg/cm2/hr		
Inhalation		
ACGIH TLV[a] = 0.5 mg/M3		
Diquat Label Use-Rates		
Reward® Landscape and Aquatic Herbicide		
Contains 2 pounds diquat cation/gallon as 3.73 pounds salt/gallon Range = 1 – 4 pounds diquat cation/ surface acre = 0.08 to 0.37 parts per million (ppm) or (mg/L) or (ug/ml) as immediate water concentrations of diquat cation		

[a] ACGIH – American Conference of Governmental Industrial Hygienists – an organization devoted to the administrative and technical aspects of occupational and environmental health. The organization is a professional society, not a government agency. The organization establishes TLVs. TLV – Threshold Limit Value is an airborne concentration of a specific chemical expressed as a time-weighted average concentration for a conventional 8 hr/dy/40 hr/wk, where it is believed that nearly all workers may be repeatedly exposed, day after day, without adverse effects (ACGIH, 1997).

Table 3: Diquat Health Advisories

Drinking Water	
MCLG[a]	0.02 mg/L
MCL[b]	0.02 mg/L or 20 ug/L
DWEL[c]	0.08 mg/L (RfD x 70 kg/ 2 L water/day)
1 day HA[d]	0.20 mg/L (10 kg child)
10 day HA[e]	0.04 mg/L (10 kg child)
Longer term HA[f]	0.04 mg/L (10 kg child)
Longer term HA[f]	0.10 mg/L (70 kg adult)
Tolerance Potable Water	0.01 mg/L
Dietary ADI	0.005 mg/kg/dy (Chronic NOAEL / 100 uncertainty factor) (EPA, 1995)
Tolerance	
Meat products 0.05 ppm	Milk 0.02 ppm
Fish 0.06 “	Shellfish 20 “
Potatoes 0.1 “	Fruiting and leafy vegetables 0.05 ppm
Cucurbits, fruits, grain crops, nuts and root, seed and pod vegetables 0.02 ppm	
Avacado, cottonseed, grasses, hops and sugarcane 0.1 ppm	

[a] Maximum Contamination Level Goal – A non-enforceable concentration of a drinking water contamination that is protective of adverse human health effects and allows an adequate MOS.

[b] Maximum Contamination Level – Maximum permissible level of contamination in water, which is delivered to any user of public water system.

[c] Drinking Water Equivalent Level – A lifetime exposure concentration protective of adverse, noncancer health effects that assumes all of the exposure to a contaminant that is from a drinking water source.

[d] One day health advisory – concentration of a chemical in drinking water that is not expected to cause any adverse non carcinogenic effects for up to 5 consecutive days of exposure, with a MOS.

[e] Ten day health advisory – same as one day HA for up to 14 consecutive days of exposure, with MOS

[f] Longer Term health advisory – same as one day HA for up to 7 years (10% of lifetime of exposure) consecutive exposure, with MOS.

[g] Lifetime health advisory – same as one day health advisory for a lifetime of exposure, with MOS.

Reference: USEPA, 1995

Table 4: Diquat Risk Assessment Noncarcinogenic Parameters

(Doses represent Diquat Ion)

Effect	**Study**	**NOEL**	**LEL**	**Reference**
Systemic Toxicity	Chronic: 1-Yr Dog Dietary Doses: 0, 0.5, 2.5, or 12.5 mg/kg/dy	0.5 mg/kg/dy or 500 ug/kg/dy	2.5[a]	Hopkins, 1990
Reproduction Toxicology	Rat 3-Generation Dietary Doses: 0, 0.8, 4 or 20/12 mg/kg/dy	4.0 mg/kg/dy or 4000 ug/kg/dy	20/12[b]	Hodge, 1990
Teratology	Rabbits dosed on days 7-16 of gestation Doses: 0, 1, 3 or 10 mg/kg/dy	Developmental effects 3.0 mg/kg/dy Maternal toxicity 1.0 mg/kg/dy	10[c] 3[d]	Hodge, 1989

[a] Increased unilateral cataracts, decreased male adrenal and epididymidies weights.

[b] Decreased numbers of live F1 pups/litter and decreased body wt gains and signs of systemic toxicity, e.g. red/brown urine, piloerection, oral ulceration, decreased body wt gains and ocular changes.

[c] Delayed ossification and various malformations possibly associated with interference of cell migration.

[d] Signs of maternal toxicity included decreases in body weight, stomach ulceration, histopathological changes in the liver and intestines and deaths.

Table 5: Swimming: Diquat Aquatic Exposure and Risk Assessment Parameters

Subjects

Age	**Weight (kg)**	**Pounds**	**Body Surface Area (cm^2)**
6 years	22	48	8,800
10 years	35	77	12,000
Adult	70	154	18,000

Water Exposure Time (Hours)

0.5
1.0
2.0
3.0

Amount Of Water Ingested During Swimming

25 ml/0.5 hours
50 ml/ l hour
100 ml/ 2 hours
150 ml/ 3 hours

Diquat Constants

Maximum use-rate = 0.37 ppm or 0.37 mg cation/L of water (Reward®, 2000)
Typical use rate = 0.08 – 0.37 ppm
Permeability Coefficient (skin) = 5.8 x 10-5 cm/hr
Octanol/Water Partition Coefficient = 2.5 x 10-5 (Worthing and Hance, 1991)
Vapor pressure = <4.0 x 10-9 mmHg @ 25^{o}C (Pack, 1987)
Water Solubility = 700 gm/L @ 20^{o}C (Worthing and Hance, 1991)

Aquatic Environmental Fate

A review of the field study data indicates that following diquat aquatic herbicidal application, the chemical is rapidly removed from water in natural systems through uptake into aquatic vegetation and binding to particulate and sediment. Bound diquat is biologically unavailable. Application of the maximum label diquat use-rate results in an initial nominal water concentration of 0.37 mg/L and depending upon the aquatic environment unbound cation decreases to approximately 0.1 mg/L within 24-hours and further reduced to <0.0l mg/L by 4 days (Calderbank, 1968; Hamer, 1994).

Table 6: Swimming: Diquat Oral Exposure and Risk Assessment[a]

Age (yrs)	Wt (kg)	Exposure Time (hrs)	Water Ingested (mls)	Diquat Total Exposure[a] (ugs)	Daily Oral Dose[b] (ug/ml/kg)	Margin of Safety		%RfD (ug/kg/dy)
						Systemic 220 ug/kg/dy	Repro 4,000ug/kg/dy	
0.37 ppm use-rate								
6	22	0.5	25	9.3	0.4	550	10,000	8
		1.0	50	18.5	0.8	275	5,000	16
		2.0	100	37	1.7	129	2,353	34
		3.0	150	55.5	2.5	88[c]	1,600	50
10	35	0.5	25	9.5	0.3	733	1,112	6
		1.0	50	18.5	0.5	440	8,000	10
		2.0	100	37	1.1	200	3,636	22
		3.0	150	55.5	1.6	138	2,500	32
Adults	70	0.5	25	9.3	0.1	2,200	40,000	2
		1.0	50	18.5	0.3	733	13,333	6
		2.0	100	37	0.5	440	8,000	10
		3.0	150	55.5	0.8	275	3,000	16
0.1 ppm 24-hours post-application								
6	22	0.5	25	2.5	0.11	2,000	36,363	2.2
		1.0	50	5.0	0.23	957	17,391	4.6
		2.0	100	10	0.45	489	8,889	9
10	35	3.0	150	15	0.68	324	5.882	13.6
		0.5	25	2.5	0.07	3,143	57,143	1.4
		1.0	50	5.0	0.14	1,571	28,571	2.8
		2.0	100	10	0.29	759	13,793	5.8
		3.0	150	15	0.43	512	9,302	8.6
Adult	70	0.5	25	2.5	0.04	5,500	100,000	1
		1.0	50	5.0	0.07	3,143	57,143	1.4
		2.0	100	10	0.14	1,571	28,571	2.8
		3.0	150	15	0.21	104	19,048	4

Table 6: Swimming: Diquat Oral Exposure and Risk Assessment[a](continued)

[a] Oral Exposure (OE) = Exposure Time (hrs) x 50 ml (water ingested/hr) x Diquat water con. (ppm) = Diquat exposure (mg/day)

[b] Oral Dose (OD) = OE / Body Weight (kg) = Diquat Dose (mg/kg/dy or converted as above to ug/kg/dy)

[c] Ingestion of ~3.5 gallons of water containing 0.37 ppm diquat during 3 hours swimming to equal systemic NOAEL of 220 ug/kg/dy

Table 7: Swimming: Diquat Dermal Exposure and Risk Assessment[a]

Total Dermal Exposure (TDE mg/day) = Exposure time (hrs) x SA x Flux Rate

Total Dermal Dose (TDD mg/kg/dy) = TDE / BW

ET = Exposure Time (0.5, 1.0, 2.0 or 3.0 hours swimming/day)

SA = Total Body Surface Area (cm2)
Flux Rate = permeability coefficient x Diquat water concentration
(5.8 x 10-5) x 0.37 ppm or ug/ml = 2.14 x 10-5 mg/cm2/hr
TDD = Total Daily Diquat Exposure / Body Weight (expressed below in ug/kg/day)
BW = Body Weight (kg)

Age (yrs)	**Wt (kg)**	**Exposure Time (hrs)**	**Body Surface Area (cm2)**	**Flux Rate**	**Dose/Day Dermal (ug/kg/day)**	**Margin of Safety**	
						Systemic 220 ug/kg/day	**Repro Tox 4000 ug/kg/day**
0.37 ppm use-rate							
6	22	0.5	8,800	2.14 x 10-5	0.0043	51,163	930,233
		1.0			0.0086	25,581	465,116
		2. 0			0.017	12,941	235,294
		3.0			0.026	8,462	153,864
10	35	0.5	12,000	2.14 x 10-	0.0037	59,459	1,081,081
		1.0			0.0073	30,137	547,945
		2.0			0.015	14,667	266,667
		3.0			0.022	10,000	90,909
Adult	70	0.5	18,000	2.14 x 10-5	0.0028	78,571	714,286
		1.0			0.0055	40,000	363,636
		2.0			0.011	20,000	181,818
		3.0			0.017	12,941	117,647

[a] Due to the low amounts of the diquat dermal doses calculated for the 0.37 ppm use-rate, no calculations were conducted to determine doses at the 0.1 ppm dissipation 24-hour level.

Table 8: Swimming: Diquat Total Exposure and Assessment

TOTAL DIQUAT EXPOSURE= Total Oral + Total Dermal (ug/kg/dy)

Age	**Wt (kg)**	**Exp. Time (hr)**	**Oral (ug/kg/dy)**	**% tot exp. oral**	**Dermal (ug/kg/dy)**	**Total Dose (ug/kg/dy)**	**Margin of Safety**		**% of RfD**
							220 ug/kg/dy	**4000 ug/kg/dy**	
0.37 ppm use-rate									
6	22	0.5	0.4	99	0.0043	0.4043	544	9,894	8
		1.0	0.8	99	0.0086	0.8086	272	4,947	16
		2.0	1.7	99	0.017	1.717	128	2,330	34
		3.0	2.5	99	0.026	2.526	87[a]	1,584	50
10	35	0.5	0.3	99	0.0037	0.3037	724	13,171	6
		1.0	0.5	99	0.0073	0.5073	434	7,885	10
		2.0	1.1	99	0.015	1.115	197	3,587	22
		3.0	1.6	99	0.022	1.622	136	2,466	32
Adult	70	0.5	0.1	97	0.0028	0.1028	2,140	38,911	2
		1.0	0.3	98	0.0055	0.3055	720	13,093	6
		2.0	0.5	98	0.011	0.511	431	7,828	10
		3.0	0.8	98	0.017	0.817	269	4,000	16
0.1 ppm 24-hour post-application									
6	22	0.5	0.11	99	0.0012	0.1112	1,978	35,971	2
		1.0	0.23	99	0.0023	0.2323	947	17,219	5
		2.0	0.45	99	0.0046	0.4546	484	8,800	9
		3.0	0.68	99	0.0076	0.6876	320	5,817	14

[a] A 22 kg child needs to drink approximately 3.4 gallons of water containing 370 ug/L diquat during 3 hours swimming to equal the systemic NOAEL of 220 ug/kg/day.

Table 9: Diquat Exposure and Risk Assessment Drinking Potable Water

PARAMETERS:

Maximum Contaminant Level (MCL) = 20 ug Diquat/liter water

Drinking Water Intake/Day

6 year old = 1000 ml or 1.0 liters
10 “ “ = 1000 ml or 1.0 liters
Adult = 2000 ml or 2.0 liters

Age	Wt	Diquat Exposure (ug)	Water Dose (ug/kg/dy)	Margin of Safety		% RfD
				Systemic 220 ug/kg/dy	**Repro Tox 4000 ug/kg/dy**	
6	22	20	0.9	244[a]	4,444	18
10	35	20	0.6	367	6,666	12
Adult	70	40	0.6	367	6,666	12

[a] A 22 kg child needs to daily drink approximately 64 gallons of water containing 20 ug/L diquat to equal the systemic NOAEL of 220 ug/kg/dy.

Table 10: Diquat Exposure and Risk Assessment Drinking Treated Surface Water

ORAL EXPOSURE (OE) = IR x WC

ORAL DOSE (OD) = OE / BW

OE = Oral Exposure (ug/day)

IR = Ingestion Rate (6 and 10 year olds 1 liter/day; adult 2 liters/day)
WC = Water Concentration (0.37, 0.1 and 0.01 ppm or ug/ml)
OD = Oral Dose (ug/kg/dy)
BW = Body Weight (kg)

Age yrs	**Wt kg**	**IR L/de**	**OE ug/dy**	**OD (ug/kg/dy)**	**Margin of Safety**		**% RfD**
					Systemic 220 ug/kg/dy	**Repro Tox** 4,000 ug/kg/dy	
0.37 ppm Use-Rate = 370 ug/L							
6	22	1	370	16.8	13[a]	238	336
10	35	1	370	10.6	21	377	212
Adult	70	2	740	10.6	21	377	212
0.1 ppm 24 Hours Post-Application							
6	22	1	100	4.5	49	889	90
10	35	1	100	2.9	76	2,759	58
Adult	70	2	200	2.9	76	2,759	58
0.01 ppm 4 Days Post-Application							
6	22	1	10	0.45	488	8,889	9
10	35	1	10	0.29	759	13,793	6
Adult	70	2	20	0.29	759	13,793	6

[a] A 22 kg child needs to drink approximately 3.4 gallons of diquat treated water each day containing 370 ug/L or 0.37 ug/ml to equal the systemic NOAEL of 220 ug/kg/dy.

Table 11: Diquat Exposure and Risk Assessment Ingestion of Fish

Diquat does not bioconcentrate in edible tissue of fish.
Typical analytical level detected in fish from recently treated water has been reported to contain

0.025 mg/kg or 25 ug/kg (Carter, 1971)

Human fish consumption (USEPA, 1989):

70 kg person fish intake/meal = 0.4 kg
35 “ “ “ “ = 0.2 kg
10 “ “ “ “ = 0.06 kg

Age	Wt	Fish Meal Wt (kg)	Diquat Exposure (ug)	Dose ug/kg/d	Margin of Safety		% RfD
					Systemic 220 ug/kg/dy	Repro Tox 4,000 ug/kg/dy	
6	22	0.06	1.5	0.07	3,143	57,143	1.4
10	35	0.2	5.0	0.14	1,571	28,571	3
Adult	70	0.4	10.0	0.14	1,571	28,571	3

Table 12: Diquat: Total Calculated Daily Exposure and Risk Assessment Drinking Potable Water

Age	Wt	Exposure Time (hrs)	Swim[a] Dose (ug/kg/dy)	Water[b] Dose (ug/kg/dy)	Fish Dose (ug/kg/dy)	Total Dose (ug/kg/dy)	Margin of Safety		% of RfD
							Systemic 220 ug/kg/dy	Repro Tox 4,000 ug/kg/dy	
MCL = 20 ug/L									
6	22	0.5	0.4043	0.9	0.07	1.37	161	2,919	28
		1.0	0.8086	“	“	1.78	124	2,247	36
		2.0	1.717	“	“	2.69	81	1,487	54
		3.0	2.526	“	“	3.50	63	1,143	70
10	35	0.5	0.3037	0.6	0.14	1.04	212	3,846	21
		1.0	0.5073	“	“	1.25	176	3,200	25
		2.0	1.115	“	“	1.86	118	2,151	37
		3.0	1.622	“	“	2.36	93	1,695	47
Adult	70	0.5	0.1028	0.6	0.14	0.84	262	4,762	17
		1.0	0.3055	“	“	1.05	210	3,510	21
		2.0	0.511	“	“	1.25	176	3,200	25
		3.0	0.817	“	“	1.56	141	2,564	31
0.1 ppm 24 Hour Post-Application[c]									
6	22	0.5	0.1112	0.9		1.1	200	3,636	22
		1.0	0.2323	“		1.2	183	5,333	24
		2.0	0.4546	“		1.4	157	2,857	28
		3.0	0.6876	“		1.7	129	2,353	34

[a] Swim dose received from water containing 370 ug/L diquat

[b] Represents diquat exposure from potable drinking water (MCL = 20 ug/L)

[c] Swim dose received from water containing 100 ug/L diquat

Table 13: Diquat: Total Calculated Daily Exposure and Risk Assessment Drinking Treated Surface Water

Age	Wt	Exposure Time (hrs)	Swim[a] Dose (ug/kg/dy)	Water[b] Dose (ug/kg/dy)	Fish Dose (ug/kg/dy)	Total Dose (ug/kg/dy)	Margin of Safety		% of RfD
							Systemic 220 ug/kg/dy	Repro Tox 4000 ug/kg/dy	
0.37 ppm Use-Rate									
6	22	0.5	0.4043	16.8	0.07	17	13	235	340
		1.0	0.8086			18	12	222	360
		2.0	1.717			19	12	211	380
		3.0	2.526			19	12	211	380
10	35	0.5	0.3037	10.6	0.14	11	20	364	220
		1.0	0.5073			11	20	364	220
		2.0	1.115			12	18	333	240
		3.0	1.622			12	18	333	240
Adult	70	0.5	0.1028	10.6	0.14	11	20	364	220
		1.0	0.3055			11	20	364	220
		2.0	0.511			11	20	364	220
		3.0	0.871			12	18	333	240
0.1 ppm 24 Hours Post-Application[c]									
6	22	0.5	0.1112	4.5	0.07	4.7	47	851	94
		1.0	0.2323	4.5		4.8	46	833	96
		2.0	0.4546	4.5		5	44	800	100
		3.0	0.6876	4.5		5.3	42	755	106

[a] Swim dose received from water containing 370 ug/L diquat

[b] Represents diquat exposure from drinking treated surface water containing 370 ug/L diquat

[c] Swim dose received from water containing 100 ug/L diquat