State of Washington
DEPARTMENT OF FISH AND WILDLIFE
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Main Office Location: Natural Resources Building, 1111 Washington Street SE, Olympia, WA

January 21, 2020

Mr. Kevin Bright
Permit Coordinator
Cooke Aquaculture Pacific, LLC
Post Office Box 669
Anacortes, WA 98221

Re: Approval of Cooke Aquaculture Pacific's Application for a Marine Finfish Aquaculture Permit to Raise All-female Triploid Rainbow Trout (Steelhead; *Oncorhynchus mykiss*)

Mr. Kevin Bright:

On January 18, 2019 Washington Department of Fish and Wildlife (WDFW) received from Cooke Aquaculture Pacific LLC (Cooke) a Marine Finfish Aquaculture Permit (Permit) application to raise all-female triploid rainbow trout (steelhead trout; *Oncorhynchus mykiss*) in existing marine net-pen facilities in Puget Sound. Also included in the January 18, 2019 correspondence were a Finfish Aquaculture Permit- Plan of Operation for all-female triploid Rainbow Trout, and updated Fish Escape Prevention, Response, and Reporting Plan and Regulated Finfish Pathogen Reporting Plan. On March 19, 2019 I informed you that your permit application would require State Environmental Policy Act (SEPA) action and that you are to submit, at a minimum, the SEPA Environmental Checklist (WAC 197-11-960). WDFW received from you on July 25, 2019 your completed SEPA Environmental Checklist and supporting documents. On October 1, 2019 WDFW responded to your SEPA Environmental Checklist by issuing at Mitigated Determination of Nonsignificance (MDNS) (SEPA 19-056), which was available for public review until November 22, 2019.

WDFW received a total of 3,581 SEPA comments on your proposed activity, although 2,669 comments were duplicates of existing comments. We have reviewed all comments and are preparing a response to those comments.

Based on the material you have submitted to WDFW, our detailed understanding of the scientific literature relevant to the Permit application (see Attachment 2), consultation with experts within and outside WDFW, tribal consultation with the Swinomish Indian Tribal Community and with consideration of the public comments, **I am approving your application to raise all-female triploid rainbow trout (steelhead trout) in existing marine net-pen facilities in Puget Sound**

where you hold valid aquatic land leases from the Washington Department of Natural Resources. This is a five-year permit and will expire on January 21, 2025.

There are two attachments to this letter. Attachment 1 includes the Permit Conditions. These conditions are the mitigating provisions that are included in our final SEPA determination. Failure to comply with these conditions may be grounds for termination and revocation of the Permit. Upon written notice of a violation, Cooke shall undertake those steps necessary to remedy the violation within the time period identified. Failure to timely and substantially remedy the violation may result in a final termination/revocation notice. You may contest a decision to terminate this Permit by filing a Notice of Appeal with WDFW addressed to the Director in accordance with the provisions of the Administrative Procedures Act (Chapter 34.05 Revised Code of Washington) and as specified in WAC 220-125-050.

Attachment 2 is a lengthy and detailed document, which will be made available to the public, that provides WDFW's justification for the MDNS decision and for the approval of your five-year Marine Aquaculture Permit, and is a response, in part, to pubic comment.

This letter, including the Permit Conditions in Attachment 1, will serve as your Marine Aquaculture Permit.

If you have questions, please contact the Dr. Kenneth Warheit, Supervisor of the WDFW Fish Health and Molecular Genetics Section, at 360-902-2595 (desk), 360-999-7889 (mobile), or kenneth.warheit@dfw.wa.gov.

Sincerely,

Kelly Susewind
Director

Attachments

cc: Amy Windrope
Kelly Cunningham
Eric Kinne
Kenneth Warheit

ATTACHMENT 1 - New

MARINE FINFISH AQUACULTURE PERMIT
Mitigating Provisions

COOKE AQUACULTURE PACIFIC

Implementation of these provisions is a requirement of the Permit

Operations, including future finfish transport permits:

1. This Permit is for the marine cultivation of all-female triploid steelhead trout (*Oncorhynchus mykiss*) from embryos originating from Troutlodge, Bonney Lake, Washington.
2. Transgenic fish, as defined in WAC 220-370-100, are not permitted
3. In accordance with Washington State Law (2018 c 179 § 3; RCW 77.175.050) this permit is valid for existing marine net-pen facilities with valid leases of state-owned aquatic lands (Fort Ward, Orchard Rocks, Clam Bay, and Hope Island facilities). This permit will become valid for existing facilities without leases of state-owned aquatic lands (Cypress 1, Cypress 2, and Port Angeles) if these leases are restored, or new leases issued.
4. In accordance with WAC 220-370-100, this permit is valid for a maximum of five years, starting from the date of this correspondence and ending January 21, 2025 or on the date of termination of leases of state-owned aquatic lands, whichever is sooner (RCW 77.175.050).
5. Cooke must receive from the Washington Department of Ecology NPDES authorization to raise all-female triploid steelhead trout in their net-pen facilities in Puget Sound before Cooke can stock facilities with steelhead trout. All requirements and provisions stipulated by Ecology on NPDES permits must be followed.
6. All activities described in Cooke Aquaculture's Plan of Operation – All-female Triploid Rainbow Trout; Fish Escape Prevention, Response, and Report Plan; and Regulated Finfish Pathogen Report Plan must be followed as written, unless otherwise specified below. All plans must be updated annually and in consultation with WDFW Fish Health and Hatchery programs, with final drafts submitted to WDFW for approval no later than November 30 of the calendar year. The Fish Escape Prevention, Response, and Report Plan must be drafted in consultation with DNR, Ecology, WDFW, and effected treaty tribes.
7. All fish transported into net-pens must contain one or more visual marks, other than the shape of each fish, that *unambiguously* identifies each fish as commercial aquaculture fish, as opposed to hatchery- or natural-origin free ranging fish of Washington State. WDFW considers that commercial aquaculture steelhead marked with adipose fin clip only presents a risk of confusion with the state's hatchery-origin steelhead. Before July 2020 Cooke must implement an alternate method, approved by WDFW, to visually identify their fish.
8. For each lot of fish to be transported into marine net-pen facilities, Cooke must provide to WDFW a sample of tissue from 150 fish appropriate for genetic analyses, if the lot is derived from a single brood line. If the lot is composed of more than one brood line, Cooke must provide to WDFW samples of tissue from 150 fish from each brood line. The fish tissue can be from live or lethal sampling. WDFW will genotype samples using their baseline assay of SNP markers and will

use the information only to determine if steelhead samples from hatchery- or natural-spawning fish are commercial aquaculture fish or F1 offspring of commercial aquaculture fish.

9. Prior to stocking net pens, Cooke must provide WDFW, DNR, and Ecology the approximate dates for stocking. Within one month after stocking is completed Cooke must provide to WDFW, DNR, and Ecology a report documenting the facility stocked, dates in which stocking occurred, the total number of fish stocked per day, and any complications that may have occurred during stocking. Cooke must report immediately if fish escaped during stocking. If requested by WDFW, DNR, or Ecology, Cooke must allow appropriately trained personnel from these agencies to monitor the stocking activities.
10. Prior to harvest, Cooke must provide WDFW, DNR, and Ecology the approximate dates for harvest. Within one month after harvesting is completed Cooke must provide to WDFW, DNR, and Ecology a report documenting the facility harvested, dates in which harvesting occurred, the total number of fish harvested per day, and any complications that may have occurred during harvesting. Cooke must report immediately if any live fish escaped during harvesting, or if any fish carcass, parts, or offal were discarded into the Puget Sound waters. The discard of carcasses, fish parts, or offal is also a violation of Cooke's NPDES permit. Cooke also must report the number and species of bycatch caught during harvesting. If requested by WDFW, DNR, or Ecology, Cooke must allow appropriately trained personnel from these agencies to monitor the harvesting activities.
11. The following monitoring data needs to be reported to WDFW, DNR, and Ecology as part of an expanded Monthly Feed, Biomass, and Disease Control Chemical Use Report, or as separate monthly report(s): (1) the feed conversion rates at each facility, (2) the estimated number of live individuals at each facility, and (3) the number of dead fish collected or observed (the greater of these two numbers) at each facility during the period since the prior reporting month.
12. For each of their facilities, Cooke must continue the net hygiene monitoring protocol developed cooperatively by Cooke and DNR (see Section 4.3.1 above).
13. WDFW Finfish Transport Permits are required when moving fish from freshwater facilities to marine net pens, or between aquatic farm sites.

Escape Prevention, Response, and Reporting:

1. In accordance with Washington State Law (2018 c 179 § 12; RCW 77.175.060) for each net-pen facility, Cooke must hire, at their own expense, a marine engineering firm approved by WDFW to conduct inspections. Inspections must occur approximately every two years, when net pens are fallow, and must include topside and mooring assessments related to escapement potential, structural integrity, permit compliance, and operations. Analyses of the mooring and cage systems of each net-pen facility must use environmental condition data that are consistent with the Norwegian aquaculture standard NS 9415 (see Section 4.3.1 above).
2. Cooke must report to WDFW Fish Health Supervisor, Lead Veterinarian, or Aquaculture Coordinator within 24 hours of discovery of any fish that has been observed to have escaped from any net-pen facility or during transfer into or out of a net-pen facility, regardless of numbers of fish involved (i.e., the minimum reporting number is one).
3. It is conceivable that an attempt to recover fish after an escape event might negatively affect native Pacific salmonids more than no attempt to recover fish. Cooke is required to work with WDFW, Ecology, DNR, effected treaty tribes, and NOAA to include a no-recovery option in the

2021 Fish Escape Prevention, Response, and Reporting Plan, to be finalized December 2020. This option should include when, where, and under what conditions a recovery effort should not be attempted. A no-recovery option would be triggered by the state, in consultation with co-managers and federal agencies for the purpose of protecting native Pacific salmonids.

4. Both the Washington Department of Health and WDFW need to be notified if escaped fish were on medicated feed at the time of their escape or are within the required withdrawal period for the medicated feed used.
5. Before January 1, 2021, Cooke must have engineered mooring and anchoring plans and site-specific engineered drawings stamped by a structural engineer, for each net-pen facility.

Triploidy error rate

1. Cooke is to work with Troutlodge and WDFW to develop or implement an alternative method or employ a different sampling and statistical design to estimate the triploidy error rate. This method will be implemented on each lot of fish to be transported into marine net-pen facilities and provide the state with an estimated number of diploid-fertile fish in that lot. This alternative method or design must be implemented no later than July 2020, unless stated otherwise by WDFW. In the absence of the alternative method Cooke will be required to sample 600 fish from each lot to determine triploidy error rate (see Section 4.3.5 above)

Finfish Pathogen Reporting and Biosecurity:

1. Cooke must ensure that all state and federal Veterinary-Client-Patient-Relationship (VCPR), Veterinarian of Record (VOR), and Veterinary Feed Directive (VFD) rules and laws are followed (e.g., WAC 246-933-200, 21 CFR 514, 21 CFR 558).
2. In accordance with WAC 220-370-080 and 220-370-130 authorized WDFW employees shall have access to freshwater hatchery facilities and marine net-pen facilities to conduct inspections, to collect samples for disease surveillance, and to inspect net-pen infrastructure.
3. Net-pen facilities must remain fallow for 42 days after the last fish are harvested and the last containment net is removed for cleaning and repair. This number can be increased per determination of WDFW veterinarian due to disease prevalence just prior to or at the time of harvest.
4. Net-pen facilities must be managed as single-generation stocking.
5. Broodstock (parents) of embryos or fish going to Cooke Aquaculture freshwater rearing facilities will be sampled and tested at a certified lab for Washington Regulated Pathogens (see Table 1 below) at the 2% APPL annually within three months of transfer from Troutlodge to Cooke's freshwater facility.
6. Lots of pre-marine smolts prior to transfer from Cooke's freshwater faculties to marine net-pens will be sampled and tested at a certified testing lab for Washington State Regulated and Reportable pathogens (see #2 above) at the 2% APPL.
7. Cooke's freshwater and marine facilities are subject to inspections by WDFW to ensure proper biosecurity, fish health, and pathogen sampling. Sampling levels can be modified by WDFW in response to pathogen findings.
8. Under no conditions should fish carcasses be removed from the net-pens and returned into waters of Puget Sound. The discard of carcasses is also a violation of Cooke's NPDES permit.

9. All disease outbreaks, unexplained mortality, regulated, reportable, or exotic pathogen findings must be reported to the WDFW Fish Health Supervisor, Lead Veterinarian, or Aquaculture Coordinator within 24 hours.
10. A fish health evaluation report written by a certified fish health inspector must be submitted to WDFW each year, no later than January 31, summarizing fish health inspections, laboratory tests, and the presence of pathogens, for the previous calendar year, at each net-pen facility (one report that includes all net-pen facilities).

Table 1. Regulated and Reportable pathogens described in WAC 220-370 and in The Salmonid Disease Control Policy of the Fisheries Co-Managers of Washington State.

a. Regulated Pathogens:
    i. Infectious hematopoietic necrosis virus (IHNV)
    ii. Infectious pancreatic necrosis virus (IPNV)
    iii. Infectious salmon anemia virus (ISAV)
    iv. *Oncorhynchus masou* virus (OMV)
    v. Viral hemorrhagic septicemia virus (VHSV)
    vi. *Myxobolus cerebralis* (whirling disease only known in fresh water)

b. Reportable Pathogen:
    i. All viral replicating agents other than those listed as Regulated pathogens that are found on cell culture using procedures outlined in the AFS-USFWS Specific Procedures for Aquatic Animal Health Inspections or OIE Aquatic Code.
    ii. Strains of pathogenic bacteria resistant to antimicrobial agents approved for use in fish or used through an extra-label prescription or INAD permit.
    iii. *Piscirickettsia salmonis*
    iv. *Nucleospora salmonis*
    v. North Atlantic variants of PRV 1, and all variants of PRV 3

ATTACHMENT 2 - New

Justification for the Mitigated Determination of Non-Significance (MDNS) for Washington Department of Fish and Wildlife SEPA 19-056 and for the Approval of Cooke Aquaculture Pacific's Marine Aquaculture Permit Application

Washington Department of Fish and Wildlife
(contact: Dr. Kenneth I. Warheit; kenneth.warheit@dfw.wa.gov)

January 21, 2020

INTRODUCTION

In January 2019, Cooke Aquaculture Pacific (hereafter termed Cooke) submitted to the Washington Department of Fish and Wildlife (WDFW) two applications: (1) an application to renew an expiring 5-year Marine Aquaculture Permit to continue to culture Atlantic salmon (*Salmo salar*) at Cooke's marine net-pen facilities in Puget Sound; and (2) an application for a new 5-year Marine Aquaculture Permit to transition production from Atlantic salmon to all-female triploid (sterile) steelhead trout (*Oncorhynchus mykiss*) at Cooke's existing marine net-pen facilities in Puget Sound. Included with these applications were Fish Escape, Prevention, Response, and Reporting Plan; Regulated Finfish Pathogen Reporting Plan; Plan of Operation for All-female Triploid Rainbow Trout[1]; and Plan of Operation for Atlantic Salmon Rearing.

In March 2019, WDFW approved and issued to Cooke a renewal of their 5-year Marine Aquaculture Permit for Atlantic salmon, contingent on the requirement specified in EHB 2957 that farming of nonnative marine finfish in Puget Sound is valid only with a current lease of state-owned aquatic lands. At the same time, WDFW responded to Cooke's second application by informing them that before WDFW could decide on their permit application a SEPA process was required to determine the environmental effects of transitioning production from Atlantic salmon to all-female, triploid steelhead trout at their existing facilities. On July 25, 2019 Cooke submitted to WDFW a completed SEPA checklist and a set of supporting documents, including information that would add to or complement the 1990 PEIS for environmental impact of fish culture in floating net-pens located anywhere in Washington State marine waters.

On October 1, 2019 WDFW issued a Mitigated Determination of Nonsignificance (MDNS) for Cooke's proposed action described in their SEPA Checklist and supporting documents. We emphasize here and elsewhere that this determination is specific and limited to Cooke's proposed action: *to transition production from Atlantic salmon to all-female, triploid steelhead trout in existing Puget Sound net-pen facilities*. This SEPA determination is tied to WDFW's substantive decision on the 5-year Marine Aquaculture Permit application for steelhead trout. This SEPA determination anticipates and discusses Cooke's planned transfers of juvenile steelhead trout from its freshwater hatchery to marine net-pens

[1] Rainbow Trout is the standard commercial aquaculture terminology for the species *Oncorhynchus mykiss*, which also includes steelhead trout. In many of their documents, Cooke uses the commercial aquaculture terminology. Since these fish are being reared in salt water, in this document we will refer to the species as steelhead trout.

facilities as part of its regular operations. These transfers would require finfish transfer permits from WDFW. The current SEPA determination therefore is intended to double as the SEPA analysis for all anticipated transfer permits inherently connected to Cooke's operations approved under the 5-year Marine Aquaculture Permit. Each individual Finfish Transport Permit application would still require a fish health and net-pen facility evaluation.

On January 21, 2020, WDFW granted Cooke's application for a 5-year Marine Aquaculture Permit to transition production from Atlantic salmon to all-female, triploid steelhead trout at their existing Puget Sound net-pen facilities where they have valid aquatic lands leases from the Department of Natural Resources (DNR).

This document provides a description of WDFW's deliberative process associated with our SEPA determination and 5-year Marine Aquaculture Permit decision. In an upcoming document WDFW will provide a response summary to the comments we received during the 52-day Public Comment period associated with our October 1, 2019 SEPA determination. However, answers to most public comments can be found within this document.

DELIBERATIVE PROCESS

1. Regulatory Authority

1.1. 2018 law sunsetting non-native finfish marine net-pen aquaculture

EHB 2957: "AN ACT Relating to reducing escape of nonnative finfish from marine finfish aquaculture facilities."

EHB 2957 became 2018 session law June 7, 2018, after passing the Washington Legislative House on February 14, 2018 and Senate on March 2, 2018, and signed by Governor Inslee on March 22, 2018. In signing the bill, Governor Inslee issued a partial veto, deleting Section 1 of the bill from the enacted law. The Governor stated that "[s]ection 1 is unnecessary to implement the bill and [he does] not agree with all the assertions made in this section." Despite the Acts title, the law's intent is three-fold: (1) the elimination of commercial nonnative finfish marine aquaculture; (2) the elimination of escapes of finfish from commercial marine net-pens; and (3) the completion of a guidance document for the planning and permitting of commercial finfish marine net-pen aquaculture. With Governor Inslee's veto of Section 1, the new law does not characterize commercial marine net-pen aquaculture as posing unacceptable risks to native salmon or the marine environment.

The new law, with bipartisan support, and the clear and explicit backing from many tribes and environmental NGOs *unambiguously allows for the continued operation of commercial net-pen aquaculture in Puget Sound, including in areas where current operations currently exist*. The new law imposes only a few constraints related to the continued operations of commercial net-pen aquaculture: (1) Washington Department of Natural Resources (DNR) may not allow the commercial culturing of nonnative finfish as an authorized use under any new state-owned aquatic lands lease, and DNR cannot renew or extend current leases for nonnative finfish aquaculture beyond their current termination date; (2) Washington departments of Ecology (Ecology) and Fish and Wildlife (WDFW) may authorize or permit the commercial culturing of nonnative finfish, or related activities only if these activities are performed under a valid lease of state-owned aquatic lands; (3) approximately every two-years, when net-pens are fallow, each facility must be inspected by an independent marine engineering firm, approved by WDFW, and to receive fish the facility must be considered in good working order; and (4) WDFW is authorized to require the immediate removal of fish from a net-pen, or deny a transport permit if the facility is in “imminent danger of collapse or release of finfish.”

The commercial culturing of native finfish (e.g., all-female triploid steelhead trout) in marine net-pens in Puget Sound is not constrained by this new law. If the commercial aquatic farmer has an aquatic farm registration, valid DNR lease, and appropriate permits from WDFW and Ecology, based on state law, **that farmer (e.g., Cooke) can legally operate native finfish net-pen aquaculture in the marine waters of Washington State**.

1.2. Washington State Regulations

WDFW received from Cooke in January 2019 an application for a new Marine Finfish Aquaculture Permit to raise all-female triploid steelhead trout in existing net-pen facilities in Puget Sound. Along with the permit application itself, Cooke also submitted a “Fish Escape Prevention, Response and Reporting Plan,” “Regulated Finfish Pathogen Reporting Plan,” and a “Plan of Operation for All-female Triploid Rainbow Trout (*Oncorhynchus mykiss*).” In March 2019 WDFW notified Cooke that the Marine Finfish Aquaculture Permit application would require analysis under SEPA (RCW Chapter 43.21C and WAC Chapter 197-11).

1.2.1. WDFW Aquaculture Rules and Regulations

WDFW’s aquaculture rules and regulations are described in RCW Chapters 77.115 and 77.125, and WAC Chapter 220-370. WAC 220-370 was last updated June 6, 2017, roughly two months prior to the collapse of Cooke’s Cypress #2 net-pen facility. WDFW currently is updating this WAC Chapter to be reflect changes in the new law (see Section 1.1 above), as detailed in RCW 77.125.

1.2.1.1. Limitations to WDFW Authority

With respect to commercial marine net-pen finfish aquaculture, WDFW’s authority is constrained by RCWs 77.115 and 77.125, and WAC 220-370. In general, WDFW’s authority is limited to (1) assessing and controlling the transmission of disease; (2) assessing genetic and ecological risk of net-pen operations to native species and their habitat; (3) preventing, reporting, and recapturing finfish released from commercial net-pen facilities; and (4) determining if the structural integrity of net-pen facilities is sufficiently adequate to receive or continue to hold the aquacultural product (e.g., Atlantic salmon or steelhead trout). In administering a disease control program, the Director of WDFW “shall not place constraints on or take enforcement actions in respect to the aquaculture industry that are more rigorous than those placed on the department or other fish-rearing entities” (RCW 77.115.010(6)).

1.2.1.2. Marine Finfish Aquaculture Permit (WAC 220-370-100)

The Marine Finfish Aquaculture Permit is described wholly in WAC 220-370-100. This Section requires that the Marine Finfish Aquaculture Permit applications be accompanied by escape prevention and escape reporting and recapture plans. The Director of WDFW can either approve or deny a permit application, and the reasons for denying an application are explicitly stated as "significant genetic, ecological or fish health risks of the proposed fish rearing program on naturally occurring fish and wildlife, their habitat or other existing fish rearing programs" (WAC 220-370-100(1)). WDFW's aquaculture regulations do not allow the Director to deny a Marine Finfish Aquaculture Permit application based on economics, social, political, or other concerns, nor is the decision subject to a vote of the people. The Director's concerns here are limited to *significant* genetic, ecological, and fish health risks.

A provision in this rule stipulates that "transgenic" fish are prohibited from being used in marine finfish aquaculture. The rule defines transgenic as the "actual transfer of genetic material from *one species to another*" (WAC 220-370-100(1)).

1.2.1.3. Finfish Transport Permit (WAC 220-370-190)

It is unlawful for any person to import into or transport within the state of Washington finfish aquaculture products (e.g., live fish, embryos (fertilized eggs), or gametes) without a Finfish Transport Permit (FTP). An FTP application is complete when all required information is submitted, including laboratory results from disease testing. No FTP application will be approved unless the aquaculture products being transported are free of regulated pathogens (see WAC 220-370-050(20)). In addition, the Director of WDFW can condition an FTP (1) "to ensure the protection of aquaculture products and native finfish from disease when the director concludes that there is a reasonable risk of disease transmission associated with the finfish aquaculture products" (WAC 220-370-190(2)); (2) to ensure the structural integrity of the net-pen facility; (3) and to prevent the captive finfish from escaping (see above Section 1.1).

1.2.1.4. Aquaculture Disease Control (WAC 220-370 -080, -180, -190, -240)

All aquatic farms, including marine net-pens, are subject to inspection by WDFW "for the prevention and suppression of aquaculture diseases, including, but not limited to, taking samples for detection of regulated finfish pathogens and other diseases" (WAC 220-370-080). Aquatic farmers are required to report by the end of the following day the detection of regulated pathogens regardless of whether fish are showing symptoms of disease or appear healthy (WAC 220-370-190(2)). If an outbreak occurs at any aquaculture facility, the aquatic farmer is required to report the outbreak immediately to WDFW (WAC 220-370-180). WDFW has great latitude to order emergency actions if the Director determines that such actions are necessary to protect native stocks from disease that will cause severe mortality. These actions include denial of a transport permit, quarantining the aquaculture products, confiscating or ordering the destruction of the aquaculture products, or requiring that the products be removed from state waters (WACs 220-370-190 and 220-370-240).

When Cooke submitted their completed Marine Finfish Aquaculture Permit application in January 2019, they complied with the requirements of WAC 220-370-100 by submitting with their application an Escape Prevention, Response and Reporting Plan, which includes all elements required by the Escape Prevention plan (WAC 220-370-110) and Escape reporting and recapture plan (WAC 220-370-120). In their Marine Finfish Aquaculture Permit application Cooke proposed a legal activity. WDFW's regulatory authority is limited and applications can be denied only if there are significant genetic, ecological, and fish health risks, or if the net-pen infrastructure is impaired enough to risk the escapement of the aquacultural product. WDFW's regulatory oversight allows for the inspection of the infrastructure, evaluation of the facilities biosecurity, and the testing of finfish for pathogens of concerns.

1.2.2. Washington State SEPA Rules (WAC 197-11)

After receiving an environmental checklist and all supporting documents, the lead agency undertakes a deliberative process and makes a threshold determination (WAC 197-11-797). In making a threshold determination, the lead agency must: (1) review the environmental checklist and all supporting documents; (2) determine if the proposed action is "likely to have a probable significant adverse environmental impact"; (3) consider procedures that may mitigate or minimize environmental impacts (WAC 197-11-768); and (4) determine if the proposed action had been analyzed in a previous environmental document (e.g., a previously prepared EIS), which can be adopted or incorporated by reference; among other elements (see WAC 197-11-330).

A "determination of significance" (DS) is made when the lead agency concludes that the proposed action would have a *probable significant* adverse environmental impact, and a DS would then require an EIS (WAC 197-11-736). It is important to note that an EIS is required *after* a determination of significance is made. SEPA defines "significant" as "a reasonable likelihood of more than a moderate impact on environmental quality" (WAC 197-11-794). SEPA does not define reasonable, likelihood, or moderate, but WDFW considered an action to have significant adverse environmental impact if a review of the scientific literature, including any existing regulatory documents, including prior EISs, supplemented by data analysis and consultation with experts, suggest that the proposed action under consideration will produce a more than moderate adverse effect. A "determination of nonsignificance" (DNS) is the opposite of a DS; that is, the proposed action will not have a significant adverse environmental impact or that the impact is something less than moderate (WAC 197-11-340). A proposed action can lead to a mitigated DNS, if the implementation of the mitigating provisions minimizes environmental impacts that otherwise may have resulted in a DS.

SEPA anticipates an evaluation of an application prior to making a threshold determination, and if based on that evaluation, a DNS or a mitigated DNS is made, an EIS is not required. <u>Since Cooke's proposed action is limited to switching production from Atlantic salmon to all-female triploid (sterile) steelhead trout</u>, WDFW's evaluation of their

application was limited to the genetic and biological risks associated with that action, and to the structural integrity of the net-pen infrastructure, as required by EHB 2957.

1.2.3. Fish Culture in Floating Net-Pens Final Programmatic EIS (January 1990)

At the direction of the Washington State Legislature, the Washington Department of Fisheries (WDF) in 1990 prepared a non-project or programmatic EIS (PEIS), in consultation with the departments of Ecology, Natural Resources, and Agriculture. The purpose of the EIS was two-fold: to assess the adequacy of the existing regulations that affect commercial marine net-pen aquaculture; and to present a Preferred Alternative that identifies governmental actions aimed at reducing or eliminating significant adverse environmental impacts. For each of nine elements of the Natural Environment[2], and the nine elements of the Built Environment[3] the PEIS describes the affected environmental element, impacts from commercial net-pen aquaculture on the affected elements, mitigation measures, and unavoidable significant adverse impacts. The PEIS also considered the cumulative impacts of the number and geographic distribution of fish farms (commercial net-pen facilities) in Puget Sound. This PEIS is foundational to any serious evaluation of the commercial finfish net-pen aquaculture in Washington State. Not only does the PEIS lay out 18 wide-ranging environmental elements that may be impacted by net-pen aquaculture, it provides actions that would mitigate for adverse impacts, discusses unavoidable significant adverse impacts (based on 1990 technology), and includes 22 pages of references. **WDFW is required by SEPA to ascertain if previous environmental documents are relevant to or have already addressed marine net-pen aquaculture in Washington State, and therefore, we incorporated by reference the PEIS in our SEPA determination (see above 1.2.2, first paragraph, #4). The incorporation of the 1990 PEIS in our SEPA determination does not indicate that the document was the only or main source used for our determination.**

2. Tribal Consultation

The Centennial Accord between federally recognized Indian tribes in Washington State and the State of Washington, dated Aug 4, 1989, provides a framework to implement government to government relationships. WDFW recognizes the sovereignty of each federally recognized Indian tribe in Washington and strives to implement government to government coordination to improve communication around Department decisions that may impact treaty resources. As such, WDFW and the Swinomish Indian Tribal Community (SITC) held a government to government consultation on December 17, 2019 to discuss impacts to treaty resources presented by Cooke's proposal to raise all-female triploid steelhead trout. The SITC emphasized that Cooke's Hope Island net-pen was of paramount concern as it lies within the boundaries of their ancestral homelands. In addition, the SITC presented testimony that the presence of the net-pens impedes their fishing treaty rights. The SITC raised additional concerns about disease transmission, genetic introgression, and ecosystem quality, which we address below in Section 4.

[2] Bottom sediments and benthos, water quality, phytoplankton, chemicals, food fish and shellfish, importation of new fish species, genetic issues, disease, and marine mammals and birds.

[3] visual quality, navigations, commercial fishing, human health, recreation, noise, odors, upland and shoreline use, and local services

WDFW appreciates the concerns raised by the SITC regarding the presence of the net-pens and the possible impacts to their fishing rights. As summarized above, *Cooke's proposed action is limited to switching production from Atlantic salmon to all-female triploid steelhead trout*. WDFW's evaluation of Cooke's application is limited by our regulatory authority over the genetic and biological risks associated with Cooke's proposed action, and to the structural integrity of net-pen infrastructure, as required by EHB 2957. WDFW's SEPA review and determination is limited in scope to Cooke's proposed action, which does not include siting issues related to already existing net-pen infrastructure. Therefore, for this SEPA action, WDFW will not review the impact of Cooke's Hope Island net-pen on SITC's fishing treaty rights.

3. Cooke's SEPA Checklist and Supporting Documents

As part of the SEPA process, Cooke submitted the required SEPA Environmental Checklist, with the following supporting documents: (1) Troutlodge Triploid Testing Results (Attachment A); (2) Additional Information: Response to WDFW Questions (Attachment B); (3) Annotated Bibliography, Prepared by Walton Dickoff, Ph.D. and Don Weitkamp, Ph.D (Attachment C); (4) Threatened and Endangered Species: 1990 Programmatic EIS Update (Attachment D); and (5) Curriculum Vitae: Don Weitkamp, Ph.D, and Walton Dickoff, Ph.D. (Attachment E). Also included with the SEPA package was a transmittal letter from Cooke and a map of the net-pen facility locations. Cooke's SEPA submission was filed by WDFW as SEPA #19056 and the entire package can be downloaded from WDFW's SEPA website: https://wdfw.wa.gov/licenses/environmental/sepa/open-comments, while the file remains open, or https://wdfw.wa.gov/licenses/environmental/sepa/closed-final, after the SEPA process closes.

4. WDFW's Environmental Review

4.1. Summary of types of data included

WDFW first began our analysis of Cooke's proposal using the information provided by Cooke in the SEPA Checklist and supporting documents (Section 3). Initially, we considered the 1990 PEIS, and these summary publications: Nash 2001, Waples et al. 2012, Price and Morris 2013, Rust et al. 2014, Hawkins et al. 2019. However, our primary evaluation was based on over 300 publications, including publications as recent as 2020. This document cites nearly 150 documents. In addition to the literature, we consulted with experts within and outside of WDFW, used unpublished data or analyses when required, and considered public comment.

4.2. Disease, Pathogen, and Parasite Control

4.2.1. Introduction

Among the often-stated concerns associated with open net-pen aquaculture, voiced in public comment and in some scientific publications, is that marine aquaculture promotes (1) the introduction of non-native pathogens, (2) amplifies rate of infection and therefore amplifies pathogen abundance, (3) promotes the increase in virulence of existing pathogens or is the nexus for the emergence of new pathogens, and (4) promotes disease in wild finfish. All these elements are thought to add risk to the viability of listed populations.

Disease in an organism is a function of the interaction between the environment (e.g., stress resulting from too high or low temperatures, high densities, lack of food; pollution), the infectious (e.g., pathogen) or non-infectious (e.g., toxin) agent, and the organism itself (e.g., genetics, immune system) (Reno 1998). In aquaculture there is an attempt to manage all three components to control pathogens and parasites, and to prevent disease (McVicar 1997).

Disease management in marine aquaculture of salmonids begins with the source material – the origin and health status of the broodstock, of the embryos, and of juvenile fish reared in freshwater hatcheries. By preventing the introduction of pathogens, especially non-native pathogens, into the cultured environment, the health status of the populations may be maintained. WDFW's regulatory authority is designed to prevent the introduction of specific pathogens by testing fish, gametes, and embryos at their source and preventing their transport if they test positive for these specific pathogens. Best management practices while the fish are cultured in marine waters can reduce stress thereby reducing risk of infection, and disease amplification and transmission. Vaccinations prepare the individual organisms' immune system to combat pathogens, and to reduce the risks of infection, pathogen amplification and transmission, and disease.

4.2.2. Importation of non-native pathogens

There are both Federal (50 CFR 16) and Washington State (see Section 1.2 above) regulations that govern the importation of salmonid gametes, embryos, and live fish into Washington State. Federal rules apply only to international importation, while Washington State rules apply to any gametes, embryos, or live fish that are transported into or through Washington State, regardless of their origin. Both Federal and State rules require that the live fish or the broodstock that produced the gametes or embryos be free of the viruses causing viral hemorrhagic septicemia (VHS), infectious hematopoietic necrosis (IHN), infectious salmon anemia (ISA; WDFW only), and infectious pancreatic necrosis (IPN), and *Oncorhynchus masou* virus, (all five viruses collectively referred to as "regulated viruses"). Rules also apply to the disinfection of the surface of embryos. In addition, since early 2018, WDFW requires that the live fish or the broodstock that produced the gametes or embryos be tested for both piscine orthoreovirus-1 (PRV-1) and PRV-3. Transport permits will be denied if the fish or broodstock test positive for North Atlantic Ocean variants of PRV-1, any variant of PRV-3, or any of the regulated viruses listed above. Lastly, WDFW requires a second round of tests after hatching when the fry's yolk sack is absorbed. If at this time the lot of fish tested positive for regulated pathogens or North Atlantic PRV-1, WDFW would require either destruction of the lot or deny any transport permit application to move live fish out of the freshwater hatchery.

Since the WDFW PRV testing requirements went into effect in 2018, WDFW denied two separate transport permit applications from Cooke because their Atlantic salmon that originated from Iceland tested positive for North Atlantic variants of PRV-1.

In their Marine Aquaculture Permit application Cooke proposes to culture only all-female triploid steelhead trout from Troutlodge, a Washington State company based out of Bonney

Lake. The broodline Cooke will use in their operation was locally derived from Puyallup River (Puget Sound) steelhead trout around 1960. WDFW will verify genetically that each lot is from this locally derived, native population of steelhead trout. **Therefore, by switching from culturing Atlantic salmon, originating from the North Atlantic, to native, locally derived steelhead trout, Cooke will dramatically reduce the risk of importing non-native pathogens.**

4.2.3. Disease Prevention

4.2.3.1. Biosecurity

We define biosecurity as precautions taken to minimize the risk of introducing, establishing, and spreading an infectious disease in an aquatic animal population. This includes, but is not limited to, disinfection of equipment, use of foot baths, limiting personnel movement, fish health monitoring, and general cleaning practices. Biosecurity also includes management activities that are designed to reduce or eliminate stress to the cultured fish. Stress can negatively affect the immune system, which can increase the fish's vulnerability to disease.

"To promote good health in farm stocks, it is in the self-interest of fish farmers to maintain good environmental conditions in their farms and in the surrounding areas" (McVicar 1997:1095). To accomplish this, and as required by WDFW, each year Cooke provides an updated "Regulated Finfish Pathogen Reporting Plan" that is reviewed and requires approval by WDFW. Within this Plan is a biosecurity section entitled "Disease Prevention and Control Measures." This section includes descriptions of specific management activities that "are designed to reduce the risk of disease occurrence at each farming location and help prevent transmission of pathogens" (p. 2 of Plan). The biosecurity activities start at the spawning facility where embryos are disinfected prior to shipping. Fish are tested for Regulated Pathogens at 30 days post swim-up after hatching, and again prior to transport to marine net-pens. Biosecurity measures continue while the fish are reared in the net-pens, and there are routine fish health exams administered by Cooke. WDFW will inspect each facility at least once per year, but more optimally twice per year. During these inspections, fish will be sampled for the presence of Regulated Pathogens and PRV.

Cooke maintains single generation stocking of their net-pens, which is a biosecurity measure that reduces stress and breaks pathogen transmission chains (see Section 4.2.4). Net-pens are also fallowed for at least 42 days after harvest and before restocking. This will allow time for the containment and predator nets to be cleaned and repaired, and contributes to breaking pathogen transmission chains.

4.2.3.2. Vaccination

The purpose of a vaccine is to provide immunological protection against a specific pathogen to prevent the onset of disease. Vaccines work by providing an initial or primary immunization – a response to an antigen (i.e., the vaccine) that results ultimately in the production of antibodies (Newman 1993). Vaccines prime the immune response by creating B-cell lymphocytes (plasma cells) that produce the antibodies that are specific to the antigen presented by the vaccine. When an individual fish encounters the pathogen for which the vaccine was produced, the immune system is already primed to secrete antibodies specific to that pathogen. This can result in a range of responses from the amelioration of clinical signs to a rapid immunological response and the prevention of infection and disease. The efficacy of a vaccine varies depending on the type of vaccine, the immunological response, and the pathogen itself. Not all vaccines are 100% efficacious, and when they are effective, that effectiveness may not last through the life of the individual fish.

Cooke vaccinated each juvenile Atlantic salmon by injection prior to their transport to salt water, targeting Infectious Hematopoetic Necrosis virus (IHNV) and the following bacteria: *Vibrio anguillarum*, *V. ordali*, *Aeromonas salmonicida*, *Tenacibaculum maritimum*, *Piscirickettsia* sp., *Moritella viscosa*, and *Allivibrio wodanis*. Vaccines have not been developed for all pathogens, and which vaccines are administered is based on the experience and knowledge of the veterinarian of record (VOR) who is licensed in Washington and has established a veterinary-client-patient-relationship (VCPR) with Cooke and the fish. Cooke anticipates using a subset of the suite of vaccines used for Atlantic salmon for their production of all-female triploid steelhead trout, focusing initially on IHNV, *V. anguillarum*, *V. ordali*, *A. salmonicida*, *and M. viscosa*.

4.2.4. Pathogen Amplification and Transmission

Net-pen aquaculture can present a variety of disease risks to wild populations (McVicar 1997, Kurath and Winton 2011). Left unmitigated, these risks may have negative effects on these populations. Aquatic farms are monocultures where fish may be handled extensively and are crowded into unnaturally high densities in environments that are not optimal for the fish. These conditions may lead to immune suppression, placing net-pen fish at risk of infection and disease (Murray and Peeler 2005, Kurath and Winton 2011). When fish are moved from the freshwater hatchery environment to the marine net-pens, they are subjected to a new environment that contributes to stress. These fish also are exposed to "wild" pathogens. The monoculture, high densities, suppressed immune systems, and the presence of wild pathogens to which these fish are naïve are conditions that can promote the amplification and transmission of these pathogens among the cultured fish (Kurath and Winton 2011). These conditions can lead to disease outbreaks, placing the farm fish population at risk.

Any disease outbreak is detrimental both to fish farms and to the aquaculture industry. Diseased individuals require treatment and treatment is expensive. Some fish will die, further eroding the business' profit margins. For these reasons the aquaculture industry is motivated to reduce the incidence of disease. For example, in Norway, risk of salmon alphavirus (SAV) and infectious salmon anemia virus (ISAV) transmission was mitigated by coordinating among neighboring farms the stocking, harvesting, and net-pen fallowing. Vaccination and early pathogen detection programs were implemented, as were veterinary prescribed treatments (Jones et al. 2015). As discussed above, Cooke runs their Atlantic salmon net-pens as single generation operations, limiting the number of times fish are handled thereby reducing some stress that may promote infection and disease. Net-pens are fallowed, and nets are cleaned following harvest eliminating potential sources of pathogens and breaking pathogen transmission chains. Fish are vaccinated for a set of pathogens, reducing the risk of infection and disease. At the onset of certain diseases, fish are treated with antibiotics (see Section 4.2.7), and WDFW regulations and Cooke's biosecurity protocols reduce the introduction of certain pathogens into net-pen populations. These mitigating operations reduce the risk of infection and disease within Cooke's Atlantic salmon program, and these same mitigating operations will also be in place for Cooke's steelhead trout program.

Despite these mitigating operations, farm fish will become infected by wild pathogens transmitted from wild populations. If fish farms amplify these pathogens, are individuals from the wild populations at increased risk if the pathogens spill back into the wild environment? Kurath and Winton (2011:73) demonstrated that "viruses move from wild fish reservoirs to infect domestic fish in aquaculture more readily than 'domestic' viruses move across the interface to infect wild stocks." They also showed 15 examples of pathogens moving from wild populations to domestic populations, and only five examples for the reverse transmission. Taranger et al. (2015:1008) state "[f]or most pathogens, clear evidence for transmission from farmed to wild fish is limited . . . . [and that] [m]ost of the diseases that currently cause problems in fish farms are likely enzootic, originating from wild fish stocks." Taranger et al. (2015) focused on four viruses and their associated diseases that result in outbreak conditions in the Atlantic salmon industry in Norway. Included in this study was heart and skeletal muscle inflammation (HSMI) and its etiological agent piscine orthoreovirus (PRV). In each of these four cases, the viruses do occur in the wild populations, to varying degrees, and may have been transmitted from the farm fish back to the wild fish (and from farm population to farm population), but the incidence of disease was either extremely low or non-existent in the wild populations. Overall, although there may be a few documented cases of bacterial or viral transmission from fish culture to wild populations, only a small subset of those involve marine net-pens (Kurath and Winton 2011), and there is limited evidence that these transmissions result in disease in the wild populations, even if the transmission is associated with disease outbreaks in the net-pens (Wallace et al. 2017).

The net-pen environment differs from the wild environment, which affects pathogen transmission, and the incidence of infection and disease. This helps explain why the amplification of wild pathogens by farmed fish does not appear to put wild populations at increased risk of disease. Wild salmonid populations, for example, would be exposed to net-pen pathogens as they migrate from fresh- to marine-water as juveniles and when they return to freshwater as adults. These populations are not subjected to the stress-inducing net-pen environment, as they travel in densities considerably lower than what occur in net-pens (Kennedy et al. 2016). The pathogens themselves do not stay concentrated in halos around net-pens, as water movement diffuses the pathogens (Brooks 2005, Brooks and Stucchi 2006), and solar radiation and microbial activity may further reduce pathogen numbers (Garver et al. 2013). Disease is intermittent within the net-pen environment, and net-pens are not a continual source of pathogens. There is evidence that pathogens can remain in sufficient concentrations to cause infection as they are dispersed from their source net-pen, but the evidence is based only on farm to farm transmission, not farm to wild transmission, and that transmission is limited by distance and time (e.g., Gustafson et al. 2007, Salama and Murray 2011, Murray 2013, Salama and Murray 2013). Compared with farmed fish, wild fish are not immune compromised, and they travel through environments that are not favorable for the transmission of pathogens. Except perhaps in freshwater spawning aggregations, and in freshwater hatcheries, wild fish are exposed to pathogen densities that are lower than that within net-pen facilities, even in wild environments in the vicinity of farms that are experiencing a disease outbreak.

4.2.5. Pathogen Virulence and Emergence of New Pathogens

Similar to the amplification of wild pathogens within aquatic farms discussed above, stocking densities and aquacultural practices can lead to the emergence of new diseases and the increase in virulence of existing pathogens (Murray and Peeler 2005, Mennerat et al. 2010, Pulkkinen et al. 2010, Walker and Winton 2010, Kennedy et al. 2016). Based on the evolution of virulence theory, Kennedy et al. (2016) outlined factors related to aquaculture operations that may lead to the increase in virulence of existing pathogens. These factors include rearing at high densities, compression of the rearing cycle, use of broodstock with limited host genetic diversity, and accepting endemic disease in cultured populations. These factors together can contribute to unbroken pathogen transmission chains, which can lead to increase in virulence or the emergence of new pathogens (e.g., Breyta et al. 2016b). For example, high rearing densities can occur in both WDFW hatchery programs and in healthy wild populations spawning naturally. However, in both cases, the high densities are not sustained and only exist during one part of the life cycle, thereby breaking pathogen transmission chains associated with high densities. In aquatic farms, transmission chains are maintained by immediately stocking after harvest the empty net-pens with new smolts from freshwater hatcheries, resulting in continuous occupancy of the aquatic farm.

As discussed above, Cooke maintains their Atlantic salmon net-pens as single generation operations, net-pens are fallowed for at least 42 days, and nets are cleaned following harvest. This process maintains fish health and breaks pathogen transmission chains. In addition, prior to transport into net-pens, each lot of fish is tested for regulated pathogens and PRV, and fish are vaccinated. While in the net-pens, when necessary, fish are treated with antibiotics to remedy disease and reduce mortality, with a secondary benefit to prevent the transmission of endemic pathogen infections. These processes maintain fish health and break pathogen transmission chains. Cooke will be required to continue these operations when culturing all-female triploid steelhead trout. **Therefore, WDFW considered Cooke's culturing of Atlantic salmon in Puget Sound to be of low risk to promote the evolution of pathogen virulence or of new pathogens and considers there to be <u>no change in that risk</u> when culturing of all-female triploid steelhead trout.**

Mordecai et al. (2019) describe three newly discovered viruses that occur in out-migrating juvenile Chinook and sockeye salmon, and in hatchery and commercial aquaculture production of Chinook salmon in British Columbia. The authors discussed the potential link between salmonid declines and the presence of viruses, and suggested that farmed Pacific salmon "may pose some transmission risk to their wild counterparts" (Mordecai et al. 2019:2). In the paper's abstract, the authors also connect dead or dying farmed fish, the presence of these new viruses in those fish and in wild fish, and the health of wild fish populations. Unfortunately, by suggesting a connection among the occurrence of the new viruses in both farmed and wild fish, disease (e.g., death and dying) in farmed fish, and the health of wild fish populations, the authors opened the door for others to make a causal link among these elements (e.g., Our Sound, Our Salmon[4]). Mordecai et al. (2019) suggest, but do not show, that these viruses are the etiological agent for any disease. Furthermore, only one of these viruses (SPAV-1) was associated with symptoms consistent with disease, and this occurred only in the farmed Chinook salmon, not in wild Chinook or sockeye salmon. Based on the data provided by the authors in a supplemental file associated with their Figure 2[5], prevalence of each virus, summing across species and origin, is 1.63% for SPAV-1, 1.78% for SPAV-2, 2.85% for PsNV, and 0.39% for CAV. CAV occurs only in farmed Chinook salmon, while SPAV-2 effectively occurs only in wild Chinook salmon (one fish each in aquaculture and hatchery, out of a combined total of 5716 fish). As Mordecai et al. (2019) indicate, the transmission of these viruses among aquaculture, hatchery, and wild fish is not known. These viruses are not common, have not been shown to cause disease, and are not associated with outbreaks. Considering all four viruses together, prevalence in wild fish is negatively correlated with prevalence in farmed fish; that is, the higher the virus's occurrence in farm fish the lower its occurrence in wild fish, and *vice versa*. **Mordecai et al. (2019) is a sound scientific publication, but it provides no evidence for either pathogen amplification within farmed fish and disease transmission from farm fish to wild fish; or viral evolution (virulence or new species) associated with net-pen aquaculture.**

4.2.6. Summary Discussion of Three Pathogens: IHNV, PRV, and Sea Lice

4.2.6.1. IHNV

Infectious hematopoietic necrosis virus (IHNV) is part of the Rhabdoviridae family; can cause acute infection, disease, and mortality in salmonids, especially in juvenile fish; affects both cultured and wild fish in fresh- and salt-water; and is endemic throughout the Pacific Northwest from Alaska to California and east to Idaho (Morzunov et al. 1995, Anderson et al. 2000, Kurath et al. 2003). The virus (IHNV) is listed by Washington State as a Regulated Pathogen (WAC 220-370-050(20)(a)(i)), and the disease (IHN) is recognized by the World Organization of Animal Health (OIE) as a Notifiable Disease[6]. WDFW is required by policy[7] to test for IHNV in broodstock for all anadromous salmonids at their hatchery facilities. In addition, embryos or live fish transported by private or commercial entities into or through Washington must be tested for IHNV; Finfish Transport Permit applications will be denied by WDFW for any lot that tests positive for IHNV. Lots of fertilized Atlantic salmon or steelhead trout

[4] https://static1.squarespace.com/static/5898d1b3cd0f689b98657619/t/5ddc152426c4ae3e67cd892b/1574704429197/OSOS_Final_SEPA_Comments.pdf

[5] DOI: https://doi.org/10.7554/eLife.47615.015

[6] https://www.oie.int/animal-health-in-the-world/oie-listed-diseases-2020/

[7] The Salmonid Disease Control Policy of the Fisheries Co-Managers of Washington State (July 2006)

embryos transported from a spawning facility to Cooke's freshwater hatchery must be free of IHNV to receive a Finfish Transport Permit from WDFW. Likewise, any lot of Atlantic salmon or steelhead trout smolt transported into Cooke's marine net-pens must be free of IHNV to receive a Finfish Transport Permit from WDFW.

There are three genogroups of IHNV in North America (U, M, and L) based on phylogenetic analyses of the middle portion of the G-gene (mid-G) (Kurath et al. 2003). The genogroups or clades have different primary hosts and different geographic distributions. In Washington, only the U and M clades exist, with the U clade divided into two subgroups (UP and UC) (Breyta et al. 2016a).

The UP clade occurs primarily along the outer coast, Puget Sound, and the Columbia River watershed upriver of the confluence with the Snake River. The UP clade is the dominant group in Puget Sound and the upper Columbia River (i.e., found in at least 75% of detections). Its primary host is sockeye salmon, with approximately 90% of its occurrence, and there is high mortality associated with this subgroup.

The UC clade occurs throughout the Columbia and lower Snake River watersheds and is the dominant group in most of this area, except in the lower and upper Columbia River. The UC clade appears to be a generalist, occurring in both Chinook salmon and steelhead trout, and it has low pathogenicity and is associated with low mortality.

The M clade in Washington is represented by the MD subgroup and throughout its range it is sympatric with either the UP or UC clades. The MD clade is the dominant group only in the lower Columbia River and it occurred briefly on the outer coast in 2007-2013. The primary hosts for the M clade are steelhead and rainbow trout, and as with the UP clade there is high mortality associated with this subgroup (Breyta et al. 2016a).

In Puget Sound only the UP clade currently exists, its primary host is sockeye salmon, and mortality can be high. The UP clade can infect steelhead trout and Chinook salmon, but there are only a few detections (adults) in both species and the virus is not associated with disease in these species (G. Kurath, pers. comm 2019).

Kurath et al. (2016) conducted laboratory challenges with Atlantic salmon, immersing juvenile fish for 1 hour in water containing IHNV. They used different variants of the U, M and L clades, and found that in Atlantic salmon, unlike sockeye and Chinook salmon, or steelhead trout, all variants caused mortality to a varying degree (20-100% for the U clade, 30-63% for the M clade, and 41-81% for the L clade). Similar studies were conducted on sockeye salmon (Garver et al. 2006, Purcell et al. 2009, Penaranda et al. 2011b), Chinook salmon (Hernandez et al. 2016), and steelhead and rainbow trout (Garver et al. 2006, Penaranda et al. 2009, Penaranda et al. 2011b, Breyta et al. 2014), with results consistent with the field observations discussed above: U clade has high virulence in sockeye salmon but low virulence in Chinook salmon, and steelhead and rainbow trout; and M clade has high virulence in steelhead and rainbow trout, low virulence in Chinook and sockeye salmon, but can replicate in sockeye salmon.

As discussed in section 4.2.3.2, Cooke vaccinates their Atlantic salmon for IHNV, and will be vaccinating their all-female triploid steelhead trout for IHNV prior to transport to marine net-pens. Cooke uses a DNA vaccine that encodes the virus's transmembrane glycoprotein (Kurath et al. 2006; H. Mitchell, pers. comm. 2019). This vaccine was derived from IHNV WRAC (039-82) strain from rainbow trout in Southern Idaho (Corbeil et al. 1999, Corbeil et al. 2000). This strain is part of the M clade genogroup (Penaranda et al. 2011a), and the vaccine is highly efficacious in steelhead and rainbow trout (Corbeil et al. 1999, Corbeil et al. 2000, LaPatra et al. 2000, LaPatra et al. 2001, Purcell et al. 2004). The vaccine confers homologous (M clade) and cross-genogroup (U clade) protection (Penaranda et al. 2011a) for up to two years (Kurath et al. 2006). The vaccine also is efficacious when administered to Chinook and sockeye salmon, but with lower relative percent survival values than in steelhead and rainbow trout (Garver et al. 2005). The vaccine appears efficacious in Atlantic salmon where Cooke's Puget Sound net-pens have tested negative since the 2012 IHN outbreak of unvaccinated Atlantic salmon.

Based on the epidemiology of the 2012 IHN outbreak in the Rich Passage net-pens (prior to ownership by Cooke), the virus was transmitted from wild fish to farmed fish (G. Kurath, pers. comm 2019), and there is no evidence that the virus was transmitted back to the wild fish and resulted in disease in wild fish.

In summary, the UP clade is the only IHNV genogroup that occurs in Puget Sound. This genogroup has high virulence in Atlantic and sockeye salmon, but low virulence in Chinook salmon and steelhead trout. Cooke will continue to vaccinate using an M clade DNA vaccine, which is efficacious against both U and M clade IHNV. **Based on the phylogeography and relative virulence of the IHNV genogroups, the risk of viral transmission from the farmed fish back to wild fish, and vaccination status of the farm fish, Cooke's net-pen facilities in Puget Sound present low risk of transmission of IHNV to wild salmonid populations. In addition, since the UP clade is of low virulence in steelhead trout, by switching from Atlantic salmon to all-female triploid steelhead trout Cooke is lowering the already low risk to wild populations.**

4.2.6.2. PRV [8]

Piscine orthoreovirus (PRV), originally named piscine reovirus (Palacios et al. 2010) but renamed by Markussen et al. (2013) to reflect the virus's phylogenetic relationships within the Reoviridae family, is a double stranded RNA virus endemic to the North Atlantic and the North Pacific, but also occurs in Chile. There are three PRV genogroups, defined genetically: PRV-1, PRV-2, and PRV-3 (Palacios et al. 2010, Olsen et al. 2015, Takano et al. 2016). PRV-1 was initially identified in Atlantic salmon farms in Norway (Palacios et al. 2010). It is now known to occur in Atlantic salmon farms throughout the North Atlantic (Garseth et al. 2013, Kibenge et al. 2013, Marty et al. 2015, Adamek et al. 2019; for other references see Table 1 in Polinski and Garver

[8] As part of Fisheries and Oceans Canada's pathogen transfer risk assessment of PRV in British Columbia, Polinski and Garver (2019) provided an excellent summary of the virus. The summary presented below follows Polinski and Garver (2019).

2019), and in Chile (Kibenge et al. 2013, Godoy et al. 2016). Retrospective analyses show that the virus has been commonly present in both the North Atlantic and eastern North Pacific Ocean regions since the mid-1980s or earlier (Marty et al. 2015, Polinski and Garver 2019). PRV-1 also occurs in:

(1) farmed Chinook salmon in British Columbia (Di Cicco et al. 2018),
(2) farmed coho salmon in Chile (Godoy et al. 2016),
(3) wild sockeye, Chinook, coho, pink, and chum salmon, and steelhead, cutthroat, and Dolly Varden trout in the eastern North Pacific (Kibenge et al. 2013, Miller et al. 2014, Morton et al. 2017, Purcell et al. 2018), and
(4) wild Atlantic salmon and sea (brown) trout in the North Atlantic (Garseth et al. 2013, Vendramin et al. 2019).

PRV-2 is known only from coho salmon in Japan, is the etiological agent for erythrocytic inclusion body syndrome (EIBS), and is associated with anemia (Takano et al. 2016).

PRV-3[9] was first discovered in farmed rainbow trout in Norway (Olsen et al. 2015). Later it was found in

(1) farmed rainbow trout throughout the North Atlantic (Dhamotharan et al. 2018, Adamek et al. 2019, Polinski and Garver 2019),
(2) brown trout in Germany (Kuehn et al. 2018), and
(3) coho salmon in Chile (Godoy et al. 2016).

PRV-3 is associated with heart and skeletal muscle inflammation (HSMI)-like disease in rainbow trout (Hauge et al. 2017, Dhamotharan et al. 2018) and proliferative darkening syndrome (PDS) in brown trout in Germany (Kuehn et al. 2018), although Fux et al. (2019) argued that PRV-3 is not associated with PDS.

Kibenge et al. (2013), Siah et al. (2015), and others have showed that there are two different subgroups within PRV-1 (PRV-1a, PRV-1b [Group I in Siah et al. 2015]). PRV-1b is restricted to the North Atlantic and in Chile, while PRV-1a occurs in the North Atlantic, eastern North Pacific, and in Chile. Within PRV-1a, genotypes from the eastern North Pacific are a monophyletic group (bold Group II in Siah et al. 2015), derived from a single ancestral viral strain, and are distinct from PRV-1a from the North Atlantic (Warheit, unpublished data). The earliest record of PRV from the eastern North Pacific is from a wild steelhead trout taken in 1977 (Marty et al. 2015). This, plus the monophyly of the eastern North Pacific PRV-1 indicates that there is a distinct evolutionary lineage of PRV-1a that is endemic to the eastern North Pacific and occurs naturally in Pacific salmon.

The viral kinetics of PRV-1 have been studied in detail in laboratories and involves three phases of infection (Polinski et al. 2019, see also Finstad et al. 2014, Wessel et al. 2015, Garver et al. 2016a, Haatveit et al. 2016, Haatveit et al. 2017, Malik et al. 2019, Wessel et al. 2019). The first or early phase includes host entry, viral replication, and

[9] Originally described as PRV-*Oncorhynchus mykiss* or PRV-*Om* since it was described in rainbow trout

dissemination to erythrocytes (red blood cells). The phase lasts 2-3 weeks, and during this time the host's immune system does not appear to recognize the virus, nor does viral shedding into the environment occur, indicating that transmission of the virus is either weak or not occurring. During the second phase, which also lasts 2-3 weeks, viral replication in the red blood cells reaches its peak, viral inclusions bodies within the red blood cells develop, host viral recognition may occur leading to an immune response, and viral shedding and therefore viral transmission occur. Throughout the last or persistent phase, which can go on indefinitely, high viral loads can be maintained but virus replication is reduced, there is no apparent host immune response, viral shedding declines, and there is poor viral transmission.

PRV-1 is infectious during that short 2-3-week second phase when the virus can be transmitted to other fish. If farm fish become infected while in the net-pens, then PRV-1 can amplify resulting in widespread transmission within the farm and most-likely among farms in relatively close proximity, as the virus is robust and can survive in adverse environmental conditions (Aldrin et al. 2010, Lovoll et al. 2012). As such, PRV-1 is ubiquitous among Atlantic salmon farms in Norway (Lovoll et al. 2012), and is pervasive within and among Atlantic salmon farms in British Columbia (Polinski and Garver 2019). During this time the virus can be transmitted to wild populations. If farm fish become infected while in the freshwater hatchery, there is a good chance that these fish enter the marine net-pens when they are already in the third or persistent phase when viral shedding and transmission is low.

The infection dynamics differ between Norway and eastern North Pacific PRV-1 strains (Polinski et al. 2019). First, PRV-1 from the eastern North Pacific is not detected in the blood plasma. However, infection by the Norway PRV-1 resulted in high viral loads in the plasma after one week of initial infection, lasting upwards of nearly eight weeks. Second, host recognition (immune response) was 2-10 times greater in the blood and more than 100 times greater in the heart for PRV-1 from Norway than PRV-1 from the eastern North Pacific. Third, heart inflammation from Norway PRV-1 reached high severity 1-2 weeks after peak viral load, thereafter the inflammation diminished. However, heart inflammation associated with eastern North Pacific PRV-1 occurred later, about four weeks after peak viral load and was maintained at high prevalence, but low severity until the end of the experiment, seven weeks later.

Two diseases have been associated with PRV-1: Heart and skeletal muscle inflammation (HSMI), occurring in farmed Atlantic salmon in Norway, and jaundice syndrome, occurring in farmed Chinook salmon in British Columbia (Wessel et al. 2019). PRV-1b has been shown to be the etiological agent of HSMI in farmed Atlantic salmon in Norway (Wessel et al. 2017). HSMI-like disease, also associated with PRV-1, has also been described from Atlantic salmon net-pens in British Columbia (Di Cicco et al. 2017, Di Cicco et al. 2018), Chile (Godoy et al. 2016), and Scotland (Ferguson et al. 2005). Although PRV-1 is ubiquitous in Atlantic salmon net-pen farms in Norway, HSMI does not occur in all fish, and when it does, it is associated with mortality upwards of 20%, and morbidity as high as 100% (Kongtorp et al. 2004, Kongtorp et al. 2006).

The relationship between PRV-1 infection and disease is complex and may be dependent on the PRV-1 strain. Although PRV-1 occurs in wild, hatchery, and net-pen farmed fish, disease is associated only with farmed fish. In Norway, PRV-1 is widespread and HSMI is common and continues to be a significant problem for the Atlantic salmon industry (Hjeltnes et al. 2019). In laboratory experiments involving PRV-1 from Norway, experimental fish were exposed to PRV, and although HSMI did not develop, moderate to severe heart lesions consistent with HSMI did occur (Hauge et al. 2016, Wessel et al. 2017). There also appears to be an association with viral load and HSMI disease in Norway (Lovoll et al. 2012). In British Columbia, Di Cicco and colleagues established an association between PRV-1 and HSMI-like symptoms in Atlantic salmon net-pen farms (Di Cicco et al. 2017, Di Cicco et al. 2018) and PRV-1 and jaundice/anemia in Chinook salmon net-pen farms (Di Cicco et al. 2018). Although PRV-1 may play a role in these diseases (Polinski and Garver 2019), PRV-1 has not been established as the etiological agent of these diseases in these farms. Furthermore, despite the fact that in laboratory experiments naïve fish exposed to PRV-1 from the eastern North Pacific became infected, neither jaundice/anemia (Atlantic, sockeye, and Chinook salmon) nor HSMI (Atlantic and sockeye salmon) developed in these fish (Garver et al. 2016a, Garver et al. 2016b, Polinski and Garver 2019, Polinski et al. 2019). Unlike HSMI in Norway, jaundice/anemia and HSMI-like diseases are rare in British Columbia. Less than 10% of Atlantic and Pacific salmon farmed fish in British Columbia die within the net-pens, and of these less than 0.5% is associated with heart pathologies. Most of these heart pathologies are idiopathic; and only 0.05% are associated with jaundice in Pacific salmon farms (Polinski and Garver 2019). That is, for every 1000 Atlantic or Pacific salmon in net-pen farms, less than five die with associated heart pathologies, and in Pacific salmon, less than one fish is associated with jaundice.

Morton et al. (2017) reported that the incidence of PRV-1 in wild salmonid populations subjected to high exposure to Atlantic salmon net-pens was statistically greater compared with wild populations with low exposure to Atlantic salmon net-pens. These authors concluded that PRV was being transferred from the net-pens to wild populations and therefore infections in the farms influence infections rates in the wild populations. Garseth et al. (2013) concluded based on a phylogenetic analysis of sequences data that transmission of PRV from Atlantic salmon net-pens to wild populations in Norway is likely. These results are consistent with those of Morton et al. (2017). WDFW compared the prevalence of PRV-1 in Chinook, coho, and sockeye salmon from Alaska (no Atlantic salmon net-pens), Columbia River (no Atlantic salmon net-pens), and Puget Sound (Atlantic salmon net-pens present) (Purcell et al. 2018) with British Columbia prevalence data from Marty et al. (2015) and Morton et al. (2017). Considering only the Marty et al. (2015) and Purcell et al. (2018) data, the highest PRV-1 prevalence for Chinook salmon was from Columbia River fish and for coho salmon from Alaska fish, while for both of these species the two geographic areas with Atlantic salmon net-pens, Puget Sound and British Columbia, showed intermediate prevalence. The prevalence in sockeye salmon was zero for Alaska, Columbia River and Puget Sound, and 0.3% from British Columbia. PRV-1 prevalence reported in Morton et al. (2017) was considerably greater in all three species than the prevalence documented by Marty et al. (2015) and Purcell et al. (2018). Compared with the highest prevalence in the Marty et al. (2015) and Purcell et al. (2018) data, PRV-1 prevalence in Morton et al. (2017) was – 3x greater for Chinook, 1.5x greater for coho, and 31x greater for sockeye salmon. Although there could be an association between the incidence of PRV-1 in wild populations and exposure to net-pens in British Columbia, the conclusion from Morton et al. (2017) may be affected by how the authors defined prevalence and how they classified wild populations with respect to exposure to Atlantic salmon net-pens.

Morton et al. (2017) also concluded that the PRV infection may lower the fitness of wild fish by decreasing their capacity to complete a difficult migration from marine waters to freshwater spawning areas, thereby impacting population viability. The authors reached this conclusion by showing that "[f]ewer [PRV] infected adults of any species were detected at higher vs. lower elevations in the Fraser River, as well as tributaries of the Skeena and Nass rivers in northern BC. This association points to a cost of infection from PRV to the fitness of wild Pacific salmon" (Morton et al. 2017:12). However, Zhang et al. (2019) found that high PRV viral load had no effect on the oxygen affinity and carrying capacity of the red blood cells even for individuals with minor heart pathology.

In summary, PRV-1 in the eastern North Pacific is phylogenetically different from PRV-1 from the North Atlantic. Although the virus tends to be ubiquitous in both regions, their infection dynamics differ, and disease is rare and the pathogenicity of the virus is low or non-existent in net-pen aquaculture in the eastern North Pacific. Although the virus may be transmitted from the net-pens to wild populations in the eastern North Pacific, the infectious period is short (but the virus may be long-lived in marine waters) and disease does not develop in wild populations. PRV is common in both farmed and wild Atlantic salmon, but its prevalence in wild steelhead trout is low – 1 out of 375 samples (0.3%; Purcell et al. 2018).[10] However, we anticipate that prevalence among all-female triploid steelhead trout in Puget Sound net-pens may be more similar to the high prevalence of farmed Atlantic salmon than that in wild steelhead trout. **Based on these analyses, we considered PRV-1 transmission from Atlantic salmon net-pens to wild salmonid populations in Puget Sound to be a low risk. We consider PRV-1 transmission from all-female triploid steelhead trout net-pens to wild salmonid populations in Puget Sound to be the same as or possibly a lower risk, compared with Atlantic salmon net-pen aquaculture.**

Finally, the August 2017 Cypress #2 accident in Puget Sound resulted in an estimated release of about 250,000 Atlantic salmon. There is a high likelihood that most or all of these Atlantic salmon were positive with a PRV-1 strain from Iceland (Kibenge et al. 2019). The source of the PRV-1 was most likely from the broodstock in Iceland. This means that fish became infected within the freshwater hatchery and were planted in

[10] Morton et al. (2017) documented 4 out of 14 steelhead trout positive for PRV-1 but see above for concerns on how prevalence is defined in this study.

the net-pens when the virus was probably in its persistent non-infectious phase. In 2018 the WDFW Fish Health Unit implemented a surveillance program for PRV-1 at selected hatcheries in Puget Sound, Washington Coast, and Columbia River. To date, we have analyzed 648 samples from Chinook and coho salmon, and steelhead and rainbow trout. Of these samples 37 (6%) tested as strong-positive, 12 (2%) as positive, 34 (5%) as weak-positive, and 564 (87%) tested negative. We obtained readable sequences for 33 samples (5%) and these sequences represented two known strains, both part of the eastern North Pacific clade. **To date there is no evidence that the 2017 Cypress #2 accident resulted in the transmission of the Icelandic PRV-1 to wild populations in Washington.**

4.2.6.3. Sea Lice

Sea lice are ectoparasitic marine copepod crustaceans that are associated with infestations and economic loss in salmonid aquaculture (reviewed in Boxaspen 2006). The copepods undergo a life cycle that starts with a nauplius larva, a planktonic stage that ultimately molt into a planktonic and infectious copepodids. The distribution, abundance, and viability of sea lice is affected by sea temperature and salinity. Bricknell et al. (2006) showed that the survival and parasitic ability of planktonic *Lepeophtheirus salmonis*, a sea louse common in both the North Atlantic and North Pacific oceans, is severely compromised at salinities less than 29 ppt. Similarly, Crosbie et al. (2019) showed that *L. salmonis* nauplii from Norway completely avoided salinities less than 30 ppt, while copepodids tolerated salinities as low as 16-20 ppt. In the eastern North Pacific there are two predominate species of sea lice that affect salmonids, *L. salmonis* and *Caligus clemensi*. At a commercial Atlantic salmon farm near the Broughton Archipelago, British Columbia, the seasonal abundance of plankton larvae for both species of sea lice varied directly with water salinity, and consistent with the North Atlantic studies, larval abundance dropped when salinities fell below 30 ppt (Byrne et al. 2018).

Farm and wild fish populations in British Columbia have experienced infestations (e.g., Marty et al. 2010, Krkosek et al. 2011), although the link between farm and wild fish infestation is not clear, nor is the link well understood between number of sea lice at farms and wild fish productivity (Morton et al. 2004, Brooks 2005, Beamish et al. 2006, Brooks and Stucchi 2006, Krkosek et al. 2006, Morton et al. 2008, Marty et al. 2010, Krkosek et al. 2011).

Sea lice may be a problem for the salmonid net-pen industry in the North Atlantic and in British Columbia, and sea lice infestations at net-pen facilities in these regions may have a negative effect on wild salmonid populations; however, in Puget Sound, although sea lice do occur in net-pen facilities, and they are monitored; their numbers do not reach a level of concern. Water circulation is complex within Puget Sound, affected by a variety of factors, including the Strait of Juan de Fuca, river discharge, and bathymetry. Nevertheless, on average, through an entire year, surface water salinities with Puget Sound remain at or below 30 ppt (Khangaonkar et al. 2011, Sutherland et al. 2011; see Figure 1 below), which results in high mortality for sea lice pelagic larvae and minimizes the likelihood of significant sea lice infestations. **Cooke's net-pen facilities are not a nexus for the amplification and transmission of sea lice to native salmonids in Puget Sound**.

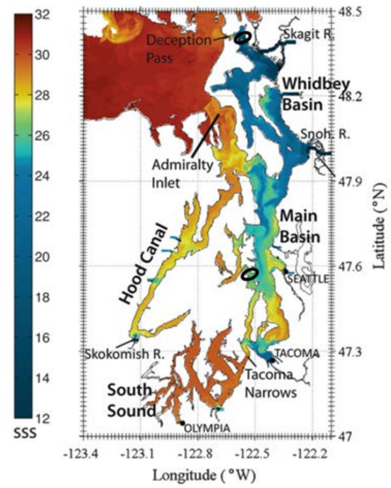

Figure 1. From Sutherland et al. (2011:Figure 1).showing sea surface salinity on 21 June 2006. Two ellipses added showing general location of Cooke's Hope Island and Rich Passage net-pen facilities.

4.2.7. Antibiotics and Medicated Feed

Antibiotics are administered to net-pen fish usually through medicated feed, referred to as Veterinary Feed Directives (VFDs). These are prescriptions written by licensed veterinarians that have established a Veterinary-Client-Patient-Relationship (VCPR) with the aquatic farmer and the fish. A veterinarian with a VCPR is formally recognized as the veterinarian of record (VOR) for a facility. VFDs, VCPRs, VORs, veterinary licenses, and the drugs that can be used for treatment of specific pathogens are all regulated by both Federal and Washington State rules. It is the VOR's obligation to adhere to these rules (i.e., violations of these rules can result in loss of license and livelihood). The "client" (owner of the fish, or the aquatic farmer) has the freedom to refuse treatment, but only a licensed veterinarian with a VCPR can order a VFD. It is the licensed veterinarian's and the VOR's license that are at risk if VFDs or other chemicals used on the fish are applied improperly or illegally, even if it is without the knowledge of the veterinarian. It is also the veterinarian's responsibility to adhere to the U.S. Food and Drug Administration's (FDA) Judicious Use of Antimicrobials policy.[11]

The most common pathogens (and their associated diseases) of the cultured Atlantic salmon in Puget Sound are: *Tenacibaculum maritimum* (yellowmouth); *Aeromonas salmonicida* (furunculosis); *Vibrio anguillarum* (vibriosis); *Piscirickettsia salmonis* (salmon rickettsia syndrome, SRS); and *Moritella viscosa* (winter ulcer) (J. Parsons, pers. comm 2020)[12]. Farm fish are particularly vulnerable to *T. maritimum* when they first enter salt water and are frequently given antibiotics to treat for yellowmouth. In fact, yellowmouth is the most

[11] See https://www.fda.gov/animal-veterinary/antimicrobial-resistance/judicious-use-antimicrobials

[12] Infectious hematopoietic necrosis virus (IHNV) is also a common pathogen of concern, but IHNV is managed through testing and a vaccine. The last outbreak of IHN in marine net-pens in Puget Sound was in 2012, prior to vaccination.

common disease for which antibiotics are applied to Atlantic salmon in Puget Sound. Experimental trials with culturing triploid steelhead trout in Puget Sound in 2012 showed that steelhead trout are more resistant to yellowmouth than are Atlantic salmon (J. Parson, pers. comm 2020), suggesting that Cooke's proposal to switch from Atlantic salmon to all-female triploid steelhead trout may result in less disease and fewer applications of antibiotics. Each of these bacteria, except for *A. salmonicida* and *P. salmonis,* are obligate marine or brackish water pathogens, and the fish become infected by these "wild" pathogens only after they enter the marine environment.

Love et al. (2020) provided a comparison of antibiotic use rates in Atlantic salmon net-pen in Norway, Scotland, Atlantic Canada, Maine, British Columbia, Washington, and Chile. Overall, from 2013 through 2017, a period prior to Cooke's ownership of the net-pens, antibiotic use based on kilogram of fish was highest in Washington followed closely by Chile. This publication does a good job documenting the relative amounts and trends of antibiotic use; however, the publication does not document the prevalence of specific pathogens in farm, freshwater hatchery, or ambient environments, and the therapeutic need for antibiotics. For example, in their risk assessment of environmental impact of Atlantic salmon farms in Norway Taranger et al. (2015) lists four viruses and sea lice, but no bacterial pathogens. Viral pathogens affect Atlantic salmon fish farms more than bacterial pathogens (see also Johansen et al. 2011), and there is a greater need in Norway to prevent viral outbreaks by using vaccines and biosecurity then there is treating bacterial outbreaks with antibiotics. Washington presents nearly the exact opposite situation than what is encountered in Norway. In the Puget Sound environment, bacterial pathogens are the dominant pathogens of concern, and therefore, we would expect greater use of antibiotics in Washington than in Norway. WDFW provides fish health services at 84 hatchery facilities and approximately 150 salmonid hatchery programs. In maintaining fish health for these programs, we emphasize biosecurity first, but VFDs are an essential part of our toolbox to treat and mitigate bacterial disease outbreaks.

In overall summary, there would be minimal differences between rearing Atlantic salmon and all-female triploid steelhead trout in Puget Sound net-pens in (1) the contraction, amplification, or transmission of pathogens; (2) their development of disease or their promotion of disease in wild finfish; or (3) their involvement in the increase virulence of existing pathogens or in the development of new pathogens. Furthermore, we consider the overall risk of these hazards to be relatively low.

4.3. Fish Escapes

Large-scale escapes (>10,000 individuals) resulting from infrastructure failure, such as the 2017 accident at Cooke Aquaculture's Cypress #2 facility (Clark et al. 2018), have happened wherever the farming of fish in open net pens is practiced. However, these events are relatively uncommon, accounting for only 19% of the fish escape incidents reported in Norway from 2001 to 2009 (Jensen et al. 2010). Across all species these large-scale incidents have been caused most frequently by mooring failure (e.g., Cypress #2), breakdown and sinking of steel floats, or major tears in the nets (Jensen et al. 2010). In December 2019 a fire destroyed part of a plastic float system in a pen in British Columbia and nearly all the 21,000 ready-for-harvest Atlantic salmon escaped (https://globalnews.ca/news/6328416/bc-fish-farm-fire-salmon/). There have been four large-scale Atlantic salmon net-pen escape events recorded in Washington; three events in four years, 1996 (107,000 salmon escaped), 1997 (369,000 fish), 1999 (115,000 fish); then no events for 18 years until the accident at Cooke's Cypress #2 net-pen in 2017 (250,000 fish) (Amos and Appleby 1999, Clark et al. 2018).

Other, often small-scale escapes, termed leakage, may occur due to errors during transfer of fish, maintenance errors, or small holes in nets caused by predators, floating debris, or vandalism (Jensen et al. 2010). Leakage of salmon from farms is typically undetectable (Britton et al. 2011, Fisher et al. 2014). There is a growing understanding that more gradual, low-level leakage of fertile fish can have a greater negative demographic and genetic impact on native species than the rarer, large-scale escape events (Baskett et al. 2013, Yang et al. 2019).

4.3.1. Structural Integrity of Net-Pen Infrastructure

Most large-scale escapes from salmon marine net-pens are a result of failures in the net-pen infrastructure (Jensen et al. 2010). The collapse of Cooke's Cypress #2 net-pen facility in 2017 that released an estimated 250,000 Atlantic salmon resulted from a failure of the mooring system and structural members of the raft's framing structure (Clark et al. 2018). Excessive biofouling by mussels and other marine organisms increased the drag force on the net-pen array, which likely resulted in the infrastructure failure. Following the Cypress #2 accident, management actions were taken that will lower the risk of net-pen infrastructure failure, compared with the risk that existed prior to the Cypress #2 accident.

- "In early in 2018, DNR and Cooke cooperatively developed a net hygiene monitoring protocol to improve net hygiene and document full compliance with the DNR Aquatic Land Leases. Since June 2018, Cooke has implemented the protocol at the Rich Passage and Hope Island facilities. [Cooke also implemented this protocol at Port Angeles until mid-2019, when rearing operations there concluded.] The protocol requires Cooke to score the cleanliness of each stock net containing fish at each farm on a weekly basis and submit those scores to DNR. Cooke substantiates the reported scores with video footage taken by Cooke divers of two stock nets per array randomly selected by DNR. DNR provides the numbers of the stock nets to be filmed the day before the filming must occur and the video is submitted to DNR within several days of being shot. The video dive footage follows a prescribed path that provides a representative view of biofouling. This video footage is required once per month during the peak vegetation growing season from May to October and every other month from November to April, when there is less vegetative biofouling. DNR's aquatic land manager, usually with a supervisor for a second set of eyes, reviews all video footage and cross checks biofouling observed with the net cleanliness scores submitted the preceding week. To date, the video verification has corroborated the net cleanliness scores reported by Cooke." (Dennis Clark, DNR; pers. communication October 2019).

- EHB 2957 requires that approximately every two-years, when net-pens are fallow, each of Cooke's facilities must be inspected by an independent marine engineering firm, approved by WDFW, and to receive fish the facility must be considered in good working order. In December 2019, a Consent Decree was reached between Cooke and Wild Fish Conservancy, where both parties agreed that before Cooke restocks any of their net-pen facilities, they are required to conduct a load analysis of the mooring and cage systems using environmental condition data that are consistent with the Norwegian aquaculture standard NS 9415. As part of the inspections mandated by EHB 2957, WDFW will require that Cooke provide an engineering analysis certifying that the net-pens conform to the parameters derived from the NS 9415 standard. Each net-pen facility will be evaluated independently as conformity to parameters derived from the NS 9415 standards require evaluation of the environmental conditions (e.g., currents, winds, waves, depth) specific to that net-pen facility. In Norway, the number of escaped Atlantic salmon declined from >600,000 fish per year (2001-2006) to <200,000 fish per year (2007-2009) after enactment of the NS 9415 standards in 2004 (Jensen et al. 2010).

4.3.2. Survival of escaped fish

The ability of escaped Atlantic salmon from fish farms in Norway to switch from pelleted feed to wild prey appears to depend upon their life stage at escape. Older, larger fish that escape often do not switch to live feed and survive poorly to sexual maturation (e.g., Skilbrei et al. 2015). Fish from the 2017 Cypress #2 event, that were harvest size at about ten pounds when the incident occurred, were found not to feed in the wild (e.g., only one of 71 fish examined (1.4%) had eaten possibly a small forage fish; WDFW, unpublished data). In contrast, fish that escape at early life stages appear to have a higher likelihood of adapting, feeding, and migrating to return as maturing adults. Jensen et al. (2013) captured Atlantic salmon, that had escaped early in the post-smolt stage, migrating and dispersing through the Arctic Ocean after one winter at sea; the growth and size of the escaped fish were similar to those of wild fish captured at the same time in the same area. Likewise, Skilbrei (2010) and Skilbrei et al. (2015) found that smolt and post-smolt escapees could survive and adopt the marine migratory pattern of their wild conspecifics. Blanchfield et al. (2009) and Patterson and Blanchfield (2013) studied survival of experimentally released diploid rainbow trout from open net-pen aquaculture in freshwater lakes in Ontario, Canada. In both studies annual survival of the released fish was approximately 50%, and although there was movement of fish away from the net-pens, most surviving fish showed continued reliance to the farm site.

Nearly all research on the behavior and survival of escaped farmed fish is based on diploid – fertile Atlantic salmon in Norway. However, in an experimental release of paired diploid and triploid Atlantic salmon from marine net-pens in Ireland, Cotter et al. (2000) and Wilkins et al. (2001) showed that significantly fewer triploid fish returned as adults to the coastal fisheries and to freshwater compared with their diploid siblings. These triploid Atlantic salmon may be less resistant to stressful environmental conditions and have significantly higher occurrence of lens cataracts than the diploid fish (Cotter et al. 2000, Wilkins et al.

2001, Cotter et al. 2002). Wilkins et al. (2001) and others (e.g., Glover et al. 2016) also postulated that the migration behavior of adult female triploid Atlantic salmon to freshwater was reduced by non-normal gonadal development. In the laboratory experiments pairing full-sibling diploid and triploid Atlantic salmon subjected to seawater challenges Leclercq et al. (2011) show that the triploid fish grow a suite of developmental deformities that may compromise their fitness in marine waters, including higher incidence and severity of lens cataracts, jaw malformation, vertebral deformities, and heart deformities possibly related to higher cardiac workloads.

Poorer survival and performance of triploid fish compared with diploid fish are not limited to Atlantic salmon. Scott et al. (2015) compared full-sibling diploid and triploid rainbow trout performance in the laboratory and showed that the triploid trout had significantly poorer hypoxia tolerance than their diploid siblings. The same result was observed in the five different strains of rainbow trout and three different brood years used in the experiment. Similar results were not seen in the adult, lake-reared trout, but Scott et al. (2015) considered that several factors may have confounded the analysis of the adult fish. Johnson et al. (2019) used hatchery rearing of full-sibling diploid and triploid steelhead trout and compared their survivorship and growth in both fresh- and salt-water. After 15 months in saltwater, the survivorship of the triploid fish was only 35% of their starting population, compared with 72% for the diploid fish. Withler et al. (1995) showed results similar to those in Johnson et al. (2019) using coho salmon.

Diploid Atlantic salmon in Norway may not be the best model to predict the survival of escaped all-female triploid steelhead trout from net-pens in Puget Sound. After four large-scale accidental releases and a number of intentional releases of Atlantic salmon in Puget Sound, there is no evidence that these fish survived for any extended period or were successful establishing spawning populations in Puget Sound (Amos and Appleby 1999). The literature on the marine survival of triploid Atlantic salmon and the relative survival of triploid rainbow and steelhead trout, and coho salmon in saltwater experiments suggest that triploid fish do poorly compared with their diploid siblings in marine waters. **We anticipate that in the unlikely event of a large-scale accidental release of all-female triploid steelhead trout from a net-pen in Puget Sound, the relative survival of the steelhead trout would be the same as or less than that previously seen with Atlantic salmon in this region**.

4.3.3. Genetic Issues

The genetic consequences of escaped diploid-fertile native species of farmed fish into open waters is a major concern with marine net-pen aquaculture (Hindar et al. 1991, Amos and Appleby 1999, Bolstad et al. 2017, Forseth et al. 2017, Glover et al. 2017, Yang et al. 2019). It is important to note that a wide variety of outcomes, ranging from no detectable effects (Glover et al. 2012) to substantial genetic introgression and even total displacement of wild populations by escaped farmed fish (Saegrov et al. 1997, Glover et al. 2012), were initially observed following escapes of Atlantic salmon from open net pens in Europe (reviewed in Hindar et al. 1991, Glover et al. 2017).

Escapes of fertile Atlantic salmon from open net pen aquaculture in the North Atlantic have been shown to have damaging impacts on the genetic variability both within and between native populations (Fleming et al. 2000, Houde et al. 2010, Karlsson et al. 2016, Bolstad et al. 2017, Glover et al. 2017). The most comprehensive data originate from Norwegian waters where five decades of farming Atlantic salmon was punctuated with escapes of millions of fish at different life stages (Diserud et al. 2019, Glover et al. 2019). Escaped, fertile and domesticated farm fish interbred with wild Atlantic salmon, thereby reducing fitness and placing more pressure on sometimes already dwindling wild populations (Fleming et al. 2000). Results show that invasions of escaped farm fish reduce reproductive fitness, reduce population productivity, disrupt local adaptations, and reduce the genetic diversity of wild salmon populations (Fleming et al. 2000, Bourret et al. 2011, Karlsson et al. 2016, Bolstad et al. 2017, Glover et al. 2017).

The impacts of escapes may vary depending on the status of the native stocks. In one example, Glover et al. (2012) studied introgression in 21 native populations of Atlantic salmon that had been exposed to large numbers of escaped farm fish and found that some populations were heavily introgressed (one native population was completely replaced with farm fish) while other populations were genetically intact. The authors concluded that healthy stocks of native fish that densely populated streams were resistant to introgression while depleted populations were much more vulnerable (see similar conclusions in Sylvester et al. 2018). This finding suggests that depleted stocks of steelhead trout and Chinook salmon in Puget Sound would be similarly vulnerable to genetic impacts from fertile con-specific farm escapees.

The use of triploid fish is recognized as normal aquaculture procedure that mitigates for the potential risks to the genetic structure and viability of wild populations from escaped farmed fish (e.g., Amos and Appleby 1999, Cotter et al. 2000, Naylor et al. 2005, Waples et al. 2012, Baskett et al. 2013, Rust et al. 2014, Glover et al. 2016, Hawkins et al. 2019). <u>Cooke is proposing to use all female triploid – sterile fish</u>

4.3.4. All female and Triploidy (Sterile) Fish

The most effective strategy to mitigate the risk of aquaculture large- or small-scale (leakage) escapes from open net pens is to limit farms to the use of sterile all-female fish (Thorgaard 1992, Cotter et al. 2000, Baskett et al. 2013, Lerfall et al. 2017). Sterile females are preferred because sterile males in many species may undergo sexual maturation and attempt to spawn even though these males produce no viable offspring (Hindar et al. 1991, Oppedal et al. 2003, Tiwary et al. 2004, Feindel et al. 2010). Such behavior from escaped males could lower the spawning success of native fish. For example, the release of sterile males has been used to reduce reproductive potential of wild populations in order to suppress populations of unwanted pests (Twohey et al. 2003, Bergstedt and Twohey 2007, Siefkes 2017). **Cooke's proposal is to use <u>all-female</u> triploid fish**.

Sterile females will be unable to successfully breed with native males and will eventually senesce and die (Tiwary et al. 2004, Lerfall et al. 2017). Introgression between escaped triploid females and the native populations will no longer be a risk to those native populations. Some results suggest that sterile salmon have a reduced instinct to enter freshwater (Cotter et al. 2000, Wilkins et al. 2001, Glover et al. 2016); thus escaped sterile females will also be less likely to compete for the spawning of wild males or dig up the redds of wild females. **Cooke's proposal is to use all-female triploid – sterile fish**.

Sterile lots of fish are most frequently produced by inducing triploidy--producing fish with three sets of chromosomes rather than the normal two. Biological regulation of chromosome sets is not as rigorously controlled in fish as in other vertebrates (Miller et al. 1994): triploidy is naturally common in some species (Qin et al. 2016, Zhigileva et al. 2017, Wu et al. 2019) and triploidy has been seen at low rates in wild salmonids (Thorgaard et al. 1982). Inducing triploidy, as in done in watermelons or bananas or oysters, for example, renders the organism largely "seedless" – that is, sterile.

The technology for producing triploid lots of fish is simple and easily applied on a commercial scale (Lerfall et al. 2017). Inducing triploidy to produce sterile Pacific salmon was optimized at Washington State University (Thorgaard et al. 1982, Parsons et al. 1986, Seeb et al. 1986, Thorgaard 1992). Triploids were raised in growth trials in net pens by the Squaxin Tribe more than 30 years ago (Seeb et al. 1993).

Triploidy can be induced at rates approaching 100% by shocking newly fertilized eggs with heat or pressure (Benfey and Sutterlin 1984); induced triploidy is practiced by some aquaculturists to reduce product loss due to precocious maturation prior to harvest (Janhunen et al. 2019) and used by management agencies who require sterile fish for sportfish stocking programs (e.g., over 9 million triploid rainbow trout have been stocked in freshwater by WDFW since 1995).

The efficacy of the methods used to create triploid fish is not 100%. This means that in every batch of triploid fish there may be fish that are fertile and can spawn with wild individuals of the same species. **Cooke's proposal is to receive all-female triploid fish from Troutlodge, a Washington State company based out of Bonney Lake. Troutlodge estimates that their average triploidy success rate is 99.83%, or a failure (error) rate of 0.17%. For the purpose of the following exercise, we will use a triploidy error rate = 0.20%**. Section 4.3.5 for discussion of triploid rates

4.3.4.1. Estimating genetic risk to native steelhead from steelhead escaping from Cooke's net-pens following a catastrophic failure of the net-pen infrastructure. To estimate risk of introgression between Cooke's net-pen steelhead and wild steelhead in Puget Sound, we estimated the following parameters and scenarios. These scenarios assume no mortality within the net-pens prior to escape. Mortality within the net-pens would reduce the number of both sterile and fertile fish that would escape.

- **Number of fish in net-pens: 1,000,000** [Clam Bay facility; Cooke Aquaculture Pacific (2019: B-33). Represents the largest facility and therefore the worst-case scenario.]
- **Triploidy error rate: 0.2%** [From above]
- **Proportion of the fish that escape: 0.82** [Based on the proportion of Atlantic salmon that escaped from Cypress #2 net-pen failure (Clark et al. 2018). We consider this to be a near worst-case scenario.]

- **Proportion of the escaped fish that elude recovery efforts: <u>0.77</u>** [Based on the number of Atlantic salmon that were recovered following the Cypress #2 net-pen failure (Clark et al. 2018). Depending on the number of fish that escape, age of fish that escape, when the fish escape, and the behavior of the fish when they escape, this proportion can be much different than what we present here.]
- **Proportion of diploid fish sexually mature at time of escape: <u>10-50% of fertile fish</u>** [50% is an extreme worst-case scenario, presented by Cooke Aquaculture Pacific (2019). Realistically, this proportion should be near zero because the fish will be harvested, on average, at less than two years of age, approximately 1-2 years prior to when they would reach sexually maturity (Cooke Aquaculture Pacific, 2019; J. Parson, pers. comm., 2019). We're using 10% as a low-end estimate, without justification other than it is greater than zero.]
- **Proportion of fish that will survive long enough to attempt to spawn: <u>50% of fertile fish</u>** [Blanchfield et al. (2009) estimated annual mortality of rainbow trout = 50%, while Patterson (2010; see also Patterson and Blanchfield 2013) estimated that 50% of the rainbow trout died within the first three months; however, Patterson and Blanchfield (2013) had unaccounted fish. Both studies consisted of experimental releases of relatively small sample sizes in freshwater. See section 4.3.2 for discussion of survival of escaped fish. We estimated that within a year of the Atlantic salmon release from Cypress #2, most if not all fish had either been recaptured or had died.]

Number of mature diploid-fertile steelhead from Cooke's net-pens that may be present in Puget Sound following an accidental release like that which occurred with Atlantic salmon at Cypress #2 in August 2017:

- 1,000,000 fish x 0.002 = 2,000 diploid-fertile fish in net-pen out of 1 million fish
- 2,000 fish x 0.82 = 1,640 diploid-fertile fish that will escape
- 1,640 x 0.77 = 1,263 diploid-fertile fish that elude recovery efforts
- 1,263 x 0.50 = 632 diploid-fertile fish that are sexually mature (higher estimate)
- 1,263 x 0.10 = 126 diploid-fertile fish that are sexually mature (lower estimate)
- 632 x 0.50 = 316 sexually mature diploid-fertile fish that survived (higher estimate)
- 126 x 0.50 = 63 sexually mature diploid-fertile fish that survived (lower estimate)

Number of mature diploid-fertile steelhead from Cooke's net-pens that may be present in Puget Sound following an accidental release like that which occurred with Atlantic salmon at Cypress #2 in August 2017, <u>but without a recovery attempt (see Section 4.3.6 and Mitigating Provisions)</u>:

- 1,000,000 fish x 0.002 = 2,000 diploid-fertile fish in net-pen
- 2,000 fish x 0.82 = 1,640 diploid-fertile fish that will escape
- ~~1,640 x 0.77 = 1,263 diploid-fertile fish that elude recovery efforts~~
- 1,640 x 0.50 = 820 diploid-fertile fish that are sexually mature (higher estimate)
- 1,640 x 0.10 = 164 diploid-fertile fish that are sexually mature (lower estimate)
- 820 x 0.50 = 410 sexually mature diploid-fertile fish that survived (higher estimate)
- 164 x 0.50 = 82 sexually mature diploid-fertile fish that survived (lower estimate)

We estimate conservatively that there will be 63 – 316 fertile female steelhead that would escape and survive to sexual maturity from Cooke Aquaculture's Clam Bay facility following an accident as described above (82 – 410 fertile female steelhead if a no recovery option was employed, or recovery was unsuccessful). If the accident were to occur at the Fort Ward site, with a total of 300,000 fish, there would be 19 – 95 fertile female steelhead that would escape and survive to sexual maturity (25 – 123) fertile female steelhead if a no recovery option was employed, or recovery was unsuccessful). Since these fish are all females, they will not spawn with each other, and to genetically affect Washington's steelhead populations, these fish would need to spawn with either hatchery- or natural-origin, natural-spawning steelhead. To do so, these domesticated fish would need to migrate into a steelhead spawning river, without homing or cues to enter a specific river, at the correct time of year, dig redds, and attract mates. These estimates are contingent on a catastrophic failure of the net-pen infrastructure, which are relatively uncommon (see Section 4.3). **Considering both the frequency of net-pen infrastructure failure and the low error rate producing triploid fish, we consider the risk to be low that domesticated all-female, triploid steelhead stocks cultured in Puget Sound net-pens will affect adversely the genetic structure of Washington's steelhead populations.**

4.3.5. Triploidy Rates

Appendix A in Cooke Aquaculture Pacific (2019) is a table provided to Cooke by Troutlodge, (the Washington State-based supplier of the all-female triploid eggs) that indicates that Troutlodge's proprietary procedure to create triploid-sterile fish has an error rate (i.e., rate of diploidy, and therefore rate of fertile fish) of 0.17%. This means that out of a lot of 1 million fish processed, there will be 998,300 triploid-sterile fish and 1,700 diploid-fertile fish. Troutlodges's table (Cooke 2019: Appendix A) shows 36 different lots of fish, each of unspecified size, that were subjected as eggs to the Troutlodge's triploidy procedure, one procedure per lot, from August 2013 through April 2018. The table includes the number of fish tested (N = 43 – 100), the number diploid results, number of triploid results, and the percent triploidy for each individual lot. Instead of treating each triploidy procedure as independent events Troutlodge treated the 36 triploidy procedures as a single continuous process, and therefore summed the total number of diploid results (five; one each in five lots) and divided that sum by the sum of all fish tested, across all 36 lots (2955), to produce an estimate of the error or diploidy rate of 0.17%. Cooke (2019: B-25) has adopted this error rate and justified Troutlodge's analysis stating that the "results in Appendix A are additive." We agree with Cooke Aquaculture that the procedure used by Troutlodge to generate triploid fish has a low error rate; perhaps as low as 0.17%. However, we disagree that the 36 lots, each containing an undisclosed number of eggs, subjected to Troutlodge's triploidy procedure over a 56-month period can be treated a single continuous process. The estimate is particularly problematic since potential variables in the procedure that may affect the procedure's efficacy were not provided (e.g., lot size; time between fertilization, water-hardening, and pressure treatment; duration and amount of high-pressure hydrostatic shock). In other words, we disagree with the method Cooke provided to estimate triploidy error rate.

Troutlodge used sample sizes ranging from 43 to 100 fish (mean = 82) per lot to calculate the triploidy error rate (Cooke 2019; Appendix A). Cooke suggested that for future lots to be transported into Puget Sound net-pen facilities, sampling rate be 100 fish per lot, reducing that sample size down to 60 if results appeared consistent with the data in Appendix A

(Cooke Aquaculture 2019; B-26). If the rate of diploidy (i.e., triploidy error rate) is indeed 0.0017 (0.17%) then the probability of drawing at least one diploid fish from a lot of 1,000,000 fish is 0.10 (n = 60 fish sampled) or 0.16 (n = 100 fish)[13]. That is, given an unbiased draw from the lot, there is a low expectation that a diploid fish will be selected using sample sizes of 60 or 100, even when there are diploid fish present in that lot. These sample sizes might underestimate the triploidy error rate, thereby underestimating the number of diploid or fertile fish in the lot. To achieve high confidence that at least one diploid fish is selected (probability > 0.90), a sample equal to 1500 is required[14]. Sample sizes of 60-100 are more appropriate for rate of diploidy equal to 5% rather than 0.17%.

Troutlodge effectively achieves a high sample size by pooling the results from all 36 lots in Cooke (2019:Appendix A) (n = 2955). To test the efficacy of this method, we modeled the procedure used by Troutlodge. That is, we assumed an actual rate of diploidy = 0.17%; drew from a hypergeometric distribution; sampled cumulatively from 36 independent lots, each with a total of 1 million eggs; and used sample sizes to test for triploidy success equal to 60, 150, 600, and 1500. We modeled each sample size 100,000 times to generate frequency distributions of calculated rates of diploidy (Figure 2). The mean rate of diploidy from 100,000 separate cumulative samples from 36 lots, for each sample size was unsurprisingly 0.17%; however, the rate of diploidy ranged from 0% to nearly 0.7% for the sample size = 60, with a large overall variance (Figure 2). That is, with a sample size of 60 for each of the 36 lots, there is a reasonable probability of calculating a rate of diploidy = 0.09%, 0.12%, or 0.19%, all close to 0.17%, and all quite low, but none equal to 0.17% (Figure 2). As we increased sample size from 150 to 1500, the precision of our estimate of the rate of diploidy increased.

If the probability of selecting one or more diploid fish from a pool of 1 million fish, with a triploidy error rate = 0.17% and a sample size = 100 is 0.16 (see above), then we would expect to draw at least one diploid fish from 16 lots out of 100 lots, or 5.8 lots out of 36 lots. Troutlodge's data showed one diploid fish from 5 lots out of 36 lots, with an average sample size per lot = 82. These results are what would be expected if the triploidy error rate was indeed close to 0.17%, as calculated by Troutlodge. We used the same modeling framework described above for Figure 2 to determine the percentage of times the calculated rate of diploidy from a cumulative sample from 36 lots was less than 0.2% out of 100,000 iterations, using a sample of only 60 fish, and actual rates of diploidy equal to 0.02%, 0.17%, 0.2% and 0.5%[15]. The calculated rates of diploidy were less than 0.2% (0.002) 100% of the time for actual rates of diploidy = 0.02%, 69% of the time for actual rates of diploidy = 0.17% (Troutlodge's estimate), and 57% of the time for actual rates of diploidy = 0.2%. The percentage of times the calculated rate of diploidy was less than 0.2% dropped dramatically to 2% for an actual rate of diploidy = 0.5%. **This suggests that the rate of diploidy for the Troutlodge triploidy procedure averages less than 0.5% and is probably close to 0.2%, essentially the same as the 0.17% provided by Cooke. However, drawing sample sizes of**

[13] Based on a hypergeometric distribution, which is equivalent to a binomial (e.g., triploid v. diploid) distribution, but sampling is without replacement, changing the probability after each trial.

[14] Unless you are sampling the entire population, you need to draw at least one diploid fish to assess triploidy error rate.

[15] Less than 0.2% is equal to the five left-most bars in Figure 2A.

60 or 100 from a lot of one million fish will not produce a precise measure of the triplody error rate. WDFW will require Cooke to have each individual lot they receive from Troutlodge tested for triploidy error rates using a sample size appropriate for the number of eggs in the lot. See Mitigating Provisions.

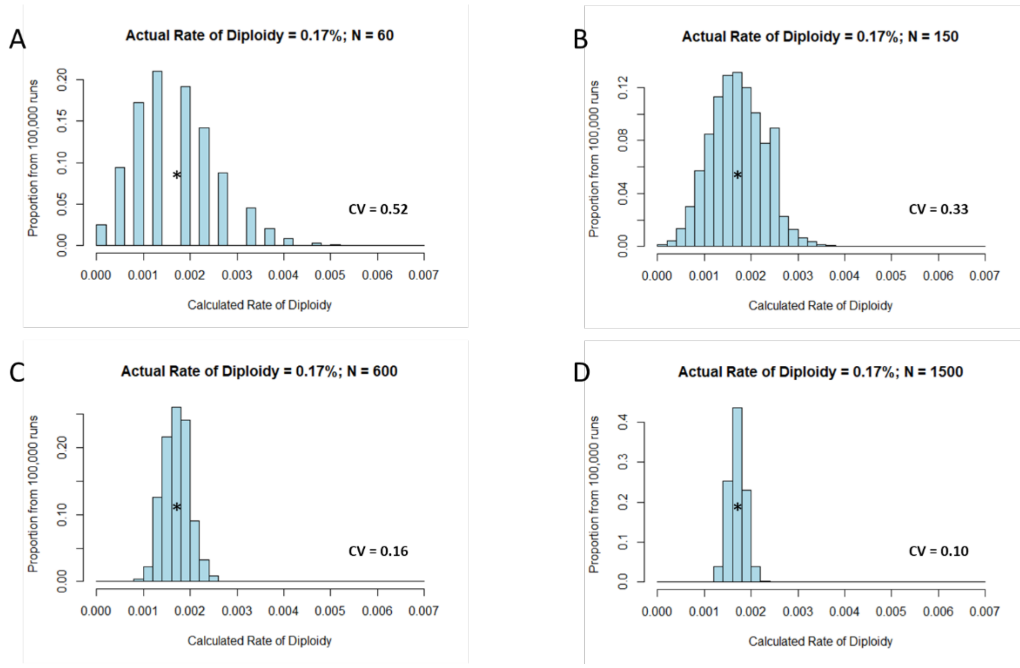

Figure 2. Frequency distributions of calculated rates of diploidy, across 100,000 iterations of the model for each of four samples sizes. Asterisk in each plot shows the frequency distribution category for 0.17%. As sample sizes increase there is an increase in the precision of the estimate for rate of diploidy, providing greater confidence that that estimate is accurate.

4.3.6. Recapture Efficacy of Escaped Fish

Recapture of fish that escape from net pens has been shown to be relatively ineffective in marine habitats, with rare exception (Dempster et al. 2018). Results show that recapture efforts must be immediate and widespread for best results, but recovery percentages are often still low (Skilbrei and Jorgensen 2010, Chittenden et al. 2011, Dempster et al. 2018). Suggestions that widespread and intense recapture efforts may show some success (Skilbrei and Jorgensen 2010) must be weighed against the risk of bycatch of native non-farm fish. Depending on the method, recapture may cause unacceptable harm in situations where ESA-listed stocks are present. The most effective and least destructive method for recapture is the use of live traps (Chittenden et al. 2011). Recapture efforts after escaped fish disperse, which could occur within hours, days or weeks, are not likely to be effective (Chittenden et al. 2011, Dempster et al. 2018), although the behavior of all-female triploid steelhead trout may differ from those species discussed in the referenced papers. However, the recapture of farmed fish within targeted rivers may provide some mitigation to prevent introgression when fertile fish escape from marine net pens (Glover et al. 2019).

4.4. Ecological effects of Net-Pen operations

4.4.1. Ecological Interactions

The published literature on the relative risks to wild populations from open net-pen aquaculture emphasizes mostly pathogen and parasite transmission and the effects from escaped farmed fish. The discussions concerning escaped farmed fish focuses on the genetic effects to wild populations, rather than ecological interactions between escaped and wild fish (see discussions above). For example, Forseth et al. (2017) developed a two-dimensional classification system of different anthropogenic factors to assess their relative risk to wild Atlantic salmon populations in Norway. The authors used 15 factors ranging from habitat alteration and hydropower to overpopulation and climate change. Included among the 15 factors were three aquaculture-related factors: sea lice, infections related to fish farming, and escaped farmed fish. The escaped farmed fish factor description was limited to the genetic risk to wild populations (Forseth et al. 2017).

Naylor et al. (2005) attempted a comprehensive assessment of the risks of escaped farmed fish to wild populations, including ecological, genetic, and socioeconomic concerns. Among the ecological risks were competition and predation. However, most of the discussion about competition and predation concerned interactions in the freshwater among juvenile fish, involving escapes from freshwater facilities or offspring from escapes. Naylor et al. (2005) stated that little is known about the competitive interactions between escaped farmed and wild fish in the marine environment, but then speculated that competition may exist since the fish show similar feeding patterns.

Two potential ecological risks to wild populations from net-pen aquaculture that have received limited attention in the literature are (1) the attraction of wild populations to the net-pen facilities, and (2) the potential entrapment and inadvertent harvest of wild fish with the net-pen cages.

Callier et al. (2018) provided a comprehensive review of the relationships between finfish and shellfish aquaculture structures and activities with the attraction (or repulsion) of wild populations. The authors indicated that these relationships are complicated and vary spatially and at several temporal scales. Many of the effects depend on fishery regulations and practices. That is, are the areas around net-pen facilities protected from fisheries, or deliberately avoided by or attract fishing activities? Callier et al. (2018) concluded that there may be effects to wild fish from finfish aquaculture structures and activities related their condition, growth, and reproductive success, and to their population's overall biomass and migratory patterns. However, these factors are poorly understood and the overall effect to population viability is unknown. Callier et al. (2018) reviewed 21 publications involving the aquaculture of eight finfish species, including Atlantic salmon and steelhead trout. The overall conclusions by these authors were consistent among the different farmed fish species. The interaction between Puget Sound net-pen facilities and aquaculture practices and the behavior of wild fish populations have not been studied, but we assume that such interactions occur. We also assume that the interactions in Puget Sound may be similar to those described by Callier et al. (2018), **and that there would be no difference in those**

interactions between the farming of Atlantic salmon and all-female triploid steelhead trout at Cooke's existing facilities.

Fish smaller than the mesh size of the net-pen cages can enter and pass through the cages. While in the cages the fish may forage and grow. Fjelldal et al. 2018 document the entrapment of eight wild species within seven Atlantic salmon net-pen facilities in Norway. The seven net-pens held a total of 4,182 Atlantic salmon, and 3,154 entrapped wild fish. The authors did not investigate if this was a normal occurrence in Atlantic salmon farms in Norway, or if there was a negative ecological effect of this bycatch. There exists the possibility that wild fish can become entrapped in Cooke's net-pen facilities in Puget Sound, and become bycatch mortalities when the farmed fish are harvested. The Canadian Government compiles and makes available the incidental finfish bycatch within British Columbia's marine finfish aquaculture farms[16]. From 2011 through September 2019 there were 1256 bycatch incidents reported by the Canadian Government that involved a total of 708,574 fish. However, two of these incidents were the deliberate depopulation of the net-pens to control the spread of IHNV outbreaks. These two incidents involved a single species (Pacific herring) and 406,366 fish, or 57% of the nine-year total. Overall, Pacific herring accounted for 638,950 (90%) of the total bycatch. The median number of fish caught as bycatch was eight fish per incident. A total of 308 Pacific salmon were caught in 87 incidents (median = 9 fish per incident), and no steelhead trout were caught. The population-level effects of this bycatch are not known, but the number of fish caught per incident is small absolutely, and small relative to their population sizes. WDFW will attempt to observe the harvest of fish from Cooke' net-pen facilities (see Section 6, Mitigating Provisions), but **there is no reason to assume that the bycatch, if any, would differ between the farming of Atlantic salmon and the farming all-female triploid steelhead trout at these facilities**.

4.4.2. Water Quality and the Benthic Environment

Washington Department of Ecology has the state's regulatory authority to protect water and sediment quality. The U.S. Environmental Protection Agency (EPA) authorized Ecology to administer the Federal Clean Water Act in Washington through National Pollutant Discharge Elimination System (NPDES) permits. RCW Chapter 90.48 defines Ecology's authority and obligations in administering the wastewater discharge permit program. On July 11, 2019, Ecology issued updated NPDES permits for the rearing of Atlantic salmon at four of Cooke's net-pen facilities, Hope Island, Clam Bay, Orchard Rocks, and Fort Ward. These updated permits require increased routine monitoring, inspections, and spill response planning and reporting.

WDFW has consulted with Ecology in making its SEPA determination and issuing to Cooke a Marine Finfish Aquaculture Permit to raise all-female triploid steelhead trout. Once Cooke has submitted to Ecology completed NPDES permit applications to raise all-female triploid steelhead trout at its existing net-pen facilities in Puget Sound, Ecology will begin a multi-step evaluation process with several opportunities for public comment. Ecology will evaluate if changing culture from Atlantic salmon to all-female triploid steelhead trout will

[16] https://open.canada.ca/data/en/dataset/0bf04c4e-d2b0-4188-9053-08dc4a7a2b03

change the character and degree of impact to water and sediment quality. WDFW defers to Ecology on their evaluation of the risk to the water and sediment from Cooke's proposed action. **As a mitigating provision (see Section 6), Cooke's Marine Finfish Aquaculture Permit to raise all-female triploid steelhead trout is contingent on receiving NPDES authorization from Ecology for this activity**.

5. NOAA Recovery Plan

The National Marine Fisheries Service (NMFS) published the final ESA Recovery Plan for Puget Sound steelhead trout on December 20, 2019. In the section describing pressures associated with ecological and genetic interactions between hatchery and natural-origin fish, NMFS included a paragraph on "Net pen Operations." The paragraph describes only the net-pen culturing of Atlantic salmon, although the paragraph ends with the mention of the potential replacement of Atlantic salmon with steelhead trout. The paragraph and a bulleted item on page 140 contain unreferenced statements about pollution and pathogen transmission risks from net-pen aquaculture. We have addressed in Section 4 of this document all the net-pen related risks discussed by NMFS in their ESA Recovery Plan.

6. Mitigating Provisions

Operations, including future finfish transport permits:

1. This Permit is for the marine cultivation of all-female triploid steelhead trout (*Oncorhynchus mykiss*) from embryos originating from Troutlodge, Bonney Lake, Washington.
2. Transgenic fish, as defined in WAC 220-370-100, are not permitted
3. In accordance with Washington State Law (2018 c 179 § 3; RCW 77.175.050) this permit is valid for existing marine net-pen facilities with valid leases of state-owned aquatic lands (Fort Ward, Orchard Rocks, Clam Bay, and Hope Island facilities). This permit will become valid for existing facilities without leases of state-owned aquatic lands (Cypress 1, Cypress 2, and Port Angeles) if these leases are restored, or new leases issued.
4. In accordance with WAC 220-370-100, this permit is valid for a maximum of five years, starting from the date of this correspondence and ending January 21, 2025 or on the date of termination of leases of state-owned aquatic lands, whichever is sooner (RCW 77.175.050).
5. Cooke must receive from the Washington Department of Ecology NPDES authorization to raise all-female triploid steelhead trout in their net-pen facilities in Puget Sound before Cooke can stock facilities with steelhead trout. All requirements and provisions stipulated by Ecology on NPDES permits must be followed.
6. All activities described in Cooke Aquaculture's Plan of Operation – All-female Triploid Rainbow Trout; Fish Escape Prevention, Response, and Report Plan; and Regulated Finfish Pathogen Report Plan must be followed as written, unless otherwise specified below. All plans must be updated annually and in consultation with WDFW Fish Health and Hatchery programs, with final drafts submitted to WDFW for approval no later than November 30 of the calendar year. The Fish Escape Prevention, Response, and Report Plan must be drafted in consultation with DNR, Ecology, WDFW, and effected treaty tribes.

7. All fish transported into net-pens must contain one or more visual marks, other than the shape of each fish, that *unambiguously* identifies each fish as commercial aquaculture fish, as opposed to hatchery- or natural-origin free ranging fish of Washington State. WDFW considers that commercial aquaculture steelhead marked with adipose fin clip only presents a risk of confusion with the state's hatchery-origin steelhead. Before July 2020 Cooke must implement an alternate method, approved by WDFW, to visually identify their fish.
8. For each lot of fish to be transported into marine net-pen facilities, Cooke must provide to WDFW a sample of tissue from 150 fish appropriate for genetic analyses, if the lot is derived from a single brood line. If the lot is composed of more than one brood line, Cooke must provide to WDFW samples of tissue from 150 fish from each brood line. The fish tissue can be from live or lethal sampling. WDFW will genotype samples using their baseline assay of SNP markers and will use the information only to determine if steelhead samples from hatchery- or natural-spawning fish are commercial aquaculture fish or F1 offspring of commercial aquaculture fish.
9. Prior to stocking net pens, Cooke must provide WDFW, DNR, and Ecology the approximate dates for stocking. Within one month after stocking is completed Cooke must provide to WDFW, DNR, and Ecology a report documenting the facility stocked, dates in which stocking occurred, the total number of fish stocked per day, and any complications that may have occurred during stocking. Cooke must report immediately if fish escaped during stocking. If requested by WDFW, DNR, or Ecology, Cooke must allow appropriately trained personnel from these agencies to monitor the stocking activities.
10. Prior to harvest, Cooke must provide WDFW, DNR, and Ecology the approximate dates for harvest. Within one month after harvesting is completed Cooke must provide to WDFW, DNR, and Ecology a report documenting the facility harvested, dates in which harvesting occurred, the total number of fish harvested per day, and any complications that may have occurred during harvesting. Cooke must report immediately if any live fish escaped during harvesting, or if any fish carcass, parts, or offal were discarded into the Puget Sound waters. The discard of carcasses, fish parts, or offal is also a violation of Cooke's NPDES permit. Cooke also must report the number and species of bycatch caught during harvesting. If requested by WDFW, DNR, or Ecology, Cooke must allow appropriately trained personnel from these agencies to monitor the harvesting activities.
11. The following monitoring data needs to be reported to WDFW, DNR, and Ecology as part of an expanded Monthly Feed, Biomass, and Disease Control Chemical Use Report, or as separate monthly report(s): (1) the feed conversion rates at each facility, (2) the estimated number of live individuals at each facility, and (3) the number of dead fish collected or observed (the greater of these two numbers) at each facility during the period since the prior reporting month.
12. For each of their facilities, Cooke must continue the net hygiene monitoring protocol developed cooperatively by Cooke and DNR (see Section 4.3.1 above).
13. WDFW Finfish Transport Permits are required when moving fish from freshwater facilities to marine net pens, or between aquatic farm sites.

Escape Prevention, Response, and Reporting:

1. In accordance with Washington State Law (2018 c 179 § 12; RCW 77.175.060) for each net-pen facility, Cooke must hire, at their own expense, a marine engineering firm approved by WDFW to

conduct inspections. Inspections must occur approximately every two years, when net pens are fallow, and must include topside and mooring assessments related to escapement potential, structural integrity, permit compliance, and operations. Analyses of the mooring and cage systems of each net-pen facility must use environmental condition data that are consistent with the Norwegian aquaculture standard NS 9415 (see Section 4.3.1 above).

2. Cooke must report to WDFW Fish Health Supervisor, Lead Veterinarian, or Aquaculture Coordinator within 24 hours of discovery of any fish that has been observed to have escaped from any net-pen facility or during transfer into or out of a net-pen facility, regardless of numbers of fish involved (i.e., the minimum reporting number is one).
3. It is conceivable that an attempt to recover fish after an escape event might negatively affect native Pacific salmonids more than no attempt to recover fish. Cooke is required to work with WDFW, Ecology, DNR, effected treaty tribes, and NOAA to include a no-recovery option in the 2021 Fish Escape Prevention, Response, and Reporting Plan, to be finalized December 2020. This option should include when, where, and under what conditions a recovery effort should not be attempted. A no-recovery option would be triggered by the state, in consultation with co-managers and federal agencies for the purpose of protecting native Pacific salmonids.
4. Both the Washington Department of Health and WDFW need to be notified if escaped fish were on medicated feed at the time of their escape or are within the required withdrawal period for the medicated feed used.
5. Before January 1, 2021, Cooke must have engineered mooring and anchoring plans and site-specific engineered drawings stamped by a structural engineer, for each net-pen facility.

Triploidy error rate

1. Cooke is to work with Troutlodge and WDFW to develop or implement an alternative method or employ a different sampling and statistical design to estimate the triploidy error rate. This method will be implemented on each lot of fish to be transported into marine net-pen facilities and provide the state with an estimated number of diploid-fertile fish in that lot. This alternative method or design must be implemented no later than July 2020, unless stated otherwise by WDFW. In the absence of the alternative method Cooke will be required to sample 600 fish from each lot to determine triploidy error rate (see Section 4.3.5 above)

Finfish Pathogen Reporting and Biosecurity:

1. Cooke must ensure that all state and federal Veterinary-Client-Patient-Relationship (VCPR), Veterinarian of Record (VOR), and Veterinary Feed Directive (VFD) rules and laws are followed (e.g., WAC 246-933-200, 21 CFR 514, 21 CFR 558).
2. In accordance with WAC 220-370-080 and 220-370-130 authorized WDFW employees shall have access to freshwater hatchery facilities and marine net-pen facilities to conduct inspections, to collect samples for disease surveillance, and to inspect net-pen infrastructure.
3. Net-pen facilities must remain fallow for 42 days after the last fish are harvested and the last containment net is removed for cleaning and repair. This number can be increased per determination of WDFW veterinarian due to disease prevalence just prior to or at the time of harvest.
4. Net-pen facilities must be managed as single-generation stocking.

5. Broodstock (parents) of embryos or fish going to Cooke Aquaculture freshwater rearing facilities will be sampled and tested at a certified lab for Washington Regulated Pathogens (see Table 1 below) at the 2% APPL annually within three months of transfer from Troutlodge to Cooke's freshwater facility.
6. Lots of pre-marine smolts prior to transfer from Cooke's freshwater faculties to marine net-pens will be sampled and tested at a certified testing lab for Washington State Regulated and Reportable pathogens (see #2 above) at the 2% APPL.
7. Cooke's freshwater and marine facilities are subject to inspections by WDFW to ensure proper biosecurity, fish health, and pathogen sampling. Sampling levels can be modified by WDFW in response to pathogen findings.
8. Under no conditions should fish carcasses be removed from the net-pens and returned into waters of Puget Sound. The discard of carcasses is also a violation of Cooke's NPDES permit.
9. All disease outbreaks, unexplained mortality, regulated, reportable, or exotic pathogen findings must be reported to the WDFW Fish Health Supervisor, Lead Veterinarian, or Aquaculture Coordinator within 24 hours.
10. A fish health evaluation report written by a certified fish health inspector must be submitted to WDFW each year, no later than January 31, summarizing fish health inspections, laboratory tests, and the presence of pathogens, for the previous calendar year, at each net-pen facility (one report that includes all net-pen facilities).

Table 1. Regulated and Reportable pathogens described in WAC 220-370 and in The Salmonid Disease Control Policy of the Fisheries Co-Managers of Washington State.

a. Regulated Pathogens:
   i. Infectious hematopoietic necrosis virus (IHNV)
   ii. Infectious pancreatic necrosis virus (IPNV)
   iii. Infectious salmon anemia virus (ISAV)
   iv. *Oncorhynchus masou* virus (OMV)
   v. Viral hemorrhagic septicemia virus (VHSV)
   vi. *Myxobolus cerebralis* (whirling disease only known in fresh water)

b. Reportable Pathogen:
   i. All viral replicating agents other than those listed as Regulated pathogens that are found on cell culture using procedures outlined in the AFS-USFWS Specific Procedures for Aquatic Animal Health Inspections or OIE Aquatic Code.
   ii. Strains of pathogenic bacteria resistant to antimicrobial agents approved for use in fish or used through an extra-label prescription or INAD permit.
   iii. *Piscirickettsia salmonis*
   iv. *Nucleospora salmonis*
   v. North Atlantic variants of PRV 1, and all variants of PRV 3

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SEPA ENVIRONMENTAL CHECKLIST

Purpose of checklist:

Governmental agencies use this checklist to help determine whether the environmental impacts of your proposal are significant. This information is also helpful to determine if available avoidance, minimization or compensatory mitigation measures will address the probable significant impacts or if an environmental impact statement will be prepared to further analyze the proposal.

A. Background

1. Name of proposed project, if applicable:

Conversion from Raising Atlantic Salmon to Raising Sterile All-Female Triploid Rainbow Trout/steelhead at Existing Marine Net Pen Sites in Puget Sound, Washington

2. Name of applicant:

Cooke Aquaculture Pacific, LLC

3. Address and phone number of applicant and contact person:

P.O. Box 669, Anacortes WA 98221
Kevin Bright, Permit Coordinator (360) 391.2409

4. Date checklist prepared: Revised July 23, 2019

A draft SEPA Checklist was submitted to WDFW on April 25, 2019. After review and comment by WDFW, this revised SEPA Checklist, dated July 23, 2019, was prepared and submitted to WDFW, WDOE and WDNR along with the SEPA Additional Information documents, a Threatened and Endangered Species Effects Analysis, and other permit application supporting information.
See Attachments A through E, identified in the list of environmental information prepared for the application (Section A.8 below).

5. Agency requesting checklist:

Washington Department of Fish and Wildlife

6. Proposed timing or schedule (including phasing, if applicable):

Permit approvals are sought by September 1, 2019 in order to transfer smolts currently growing at the Cooke Aquaculture hatchery in Thurston County to one or more of the marine net pen sites.

7. Do you have any plans for future additions, expansion, or further activity related to or connected with this proposal? If yes, explain.

No. Cooke Aquaculture has no plans for future additions or expansion of existing operations at its existing Puget Sound marine net pen sites.

8. List any environmental information you know about that has been prepared, or will be prepared, directly related to this proposal.

Finfish Aquaculture Plan of Operations, All-Female Triploid Rainbow Trout (*Oncorhynchus mykiss*). Cooke Aquaculture Pacific, LLC (updated January 18, 2019).

Fish Escape Prevention, Response & Reporting Plan. Cooke Aquaculture Pacific, LLC (updated October 12, 2018).

Regulated Finfish Pathogen Reporting Plan, Cooke Aquaculture Pacific, LLC (updated January 25, 2017).

SEPA Checklist Attachment A: Troutlodge Triploid Testing Results (2018).

SEPA Checklist Attachment B: Additional Information: Response to WDFW Questions (July 23, 2019).

SEPA Checklist Attachment C: Rainbow Trout Net Pen Aquaculture Annotated Bibliography (July 23, 2019).

SEPA Checklist Attachment D: Threatened and Endangered Species: 1990 PEIS Update (July 23, 2019).

SEPA Checklist Attachment E: Curriculum Vitae for Don Weitkamp, Ph.D. and Walton Dickhoff, Ph.D., Technical Experts who Prepared the Annotated Bibliography, Contributed to the Additional Information Document (SEPA Checklist Attachment B), and Prepared the Threatened and Endangered Species Effects Analysis (July 23, 2019).

9. Do you know whether applications are pending for governmental approvals of other proposals directly affecting the property covered by your proposal? If yes, explain.

No. There are no applications pending for governmental approvals of other proposals directly affecting the existing Cooke Aquaculture marine net pen sites.

10. List any government approvals or permits that will be needed for your proposal, if known.

Permit or Approval Required	Agency
Marine Finfish Aquaculture Permit Reapproval of existing permit (March 19, 2019) to change species cultured from Atlantic Salmon to all-female triploid (sterile) Rainbow Trout/steelhead	WDFW
NPDES Permit Modification for change of use to include alternate species such as sterile Rainbow Trout/steelhead and other possible native fish species at existing marine net pen sites[1]	Ecology
State-owned Aquatic Land Lease amendment to include culturing native fish species (all-female sterile Rainbow Trout/steelhead) at existing marine net pen sites	WDNR
Fish Transfer Permits to transport fish from the Cooke Aquaculture hatcheries to the marine net pen sites	WDFW

[1] Existing NPDES permits for the Fort Ward, Orchard Rocks, Clam Bay and Hope Island net pen facilities were renewed by the Washington Department of Ecology on July 11, 2019. Copies of the new NPDES permits are included by reference.

11. Give brief, complete description of your proposal, including the proposed uses and the size of the project and site. There are several questions later in this checklist that ask you to describe certain aspects of your proposal. You do not need to repeat those answers on this page.

Cooke Aquaculture Pacific, LLC (Cooke) is a Washington company that operates commercial marine net pens in Clallam, King, Kitsap and Skagit Counties, and two freshwater hatcheries in Thurston County. The company's facilities have been commercially raising and harvesting farmed salmon in the Puget Sound region since the 1970s. The net pen farms initially raised various types of salmonids including Coho Salmon, Chinook Salmon, Donaldson Rainbow Trout and steelhead trout. By the early 1990s, the Washington industry transitioned to raising primarily Atlantic Salmon. Cooke is proposing to change the fish species being cultured from Atlantic Salmon (*Salmo salar*) to domesticated stocks of all-female triploid Rainbow Trout/steelhead (*Oncorhynchus mykiss*).[2] While the common names Rainbow Trout and steelhead are often used interchangeably, this environmental review document and permit applications will refer to Rainbow Trout/steelhead, which is the single official common name given to this species by the American Fisheries Society several years ago. All-female triploid Rainbow Trout/steelhead have been selected to be raised in the existing net pen facilities because the use of mono-sex and sterile stocks of fish is recognized as means to significantly reduce potential genetic interaction with natural populations.

The Cooke Puget Sound marine net pen sites have all required State agency permits, and existing operations have been raising almost exclusively[3] only Atlantic Salmon for the past 30 years. A recent change in Washington law (RCW 77.125.050), however, requires phasing out the farming of non-native finfish in marine net pen aquaculture. This change means that commercial marine net pen production in Washington will need to find a commercially viable native species of fish to grow in order stay in the business of growing seafood for human consumption. Rainbow Trout/steelhead are a native species to the Pacific Northwest region and have been commercially raised in Washington, primarily in freshwater facilities, for over 80 years if not even longer. Cooke is requesting a re-approval of their Marine Finfish Aquaculture Permit (WAC 220-370-100) from the Washington Department of Fish and Wildlife (WDFW) that will allow the company to start growing domesticated stocks of a mono-sex (all-female) sterile (triploid) Rainbow trout/steelhead (*Oncorhynchus mykiss*) at their marine farms. Cooke received the renewal of their Marine Finfish Permit from WDFW on March 19, 2019 to continue raising Atlantic salmon at the four farm sites with a valid Aquatic Use Permit from WDNR (Clam Bay, Fort Ward, Orchard Rocks and Hope Island). Other than transitioning to the commercial cultivation of a different species of fish, the company is not planning any alteration to the existing fish pen physical structures, locations, supporting equipment, or general current practices, methods, maintenance and cultivation techniques currently used for growing Atlantic Salmon in net pens. Domesticated stocks of triploid Rainbow Trout/steelhead have very similar physiological and metabolic requirements to those of domesticated stocks of Atlantic Salmon. The basic difference is that all-female triploid Rainbow trout/steelhead are known to be reproductively sterile, and thus convert their energy almost entirely to growth. By comparison, diploid populations of Rainbow trout/steelhead and Atlantic salmon will reach a certain age and begin to expend growth energy toward the production of gametes and secondary sexual characteristics (sexual maturation).

Raising native-stock fish species in marine net pens is consistent with the recommendations of the *Final Programmatic Environmental Impact Statement: Fish Culture in Floating Net-Pens* (Washington Department of Fisheries, January 1990). The Preferred Alternative in the Programmatic EIS (PEIS)

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2 Genetically, Rainbow Trout and steelhead are the same species (*Oncorhynchus mykiss*), but they represent two separate life strategies. Rainbow Trout are the resident form that stay in freshwater. Steelhead trout are the anadromous form that migrates to the ocean and back to natal rivers and streams to spawn.

3 Other species of salmonids such as Coho and Chinook Salmon, and steelhead trout have also been raised at some of these marine net pens and other facilities in Puget Sound in the past.

concludes that *"In areas where WDF determines there is a risk of significant interbreeding (with indigenous species) or establishment of harmful self-sustaining populations, the agency should only approve the farming of sterile* ***or*** *monosexual individuals, or genetically incompatible species."* By farming a monosex (all-female) and sterile (triploid) stock of Rainbow Trout/steelhead, Cooke's proposed change of species incorporates both methods of reducing the risks of genetic interference to indigenous populations or from escaped farmed fish establishing self-sustaining feral populations. WDFW has requested that a SEPA Checklist be prepared, along with Additional Information (e.g., response to the Department's specific questions) for the Department's consideration during their review of the Cooke Aquaculture application for re-approval of the Marine Finfish Aquaculture permit to authorize the species conversion. The additional information provided in SEPA Checklist Attachments A though E update the 1990 Programmatic EIS, and provide specific detailed information regarding potential change in environmental impacts that may occur as a result of the Cooke Aquaculture species conversion proposal.

The 1990 *Final Programmatic Environmental Impact Statement: Fish Culture in Floating Net-Pens* is adopted herein by reference, in accordance with WAC 197-11-630. The SEPA Additional Information document (Attachment B), supported by an Annotated Bibliography (Attachment C), addresses the potential environmental effects of the species change proposal on the subjects of escapement, competition, potential genetic interactions, and minor operational differences. The current status of Federally-listed species under the Endangered Species Act since the 1990 PEIS was issued, as well as State-listed species, is described in Attachment D, along with an effects analysis for both Atlantic Salmon culture (which has occurred throughout the years since the 1990 PEIS was issued), and an effects analysis for the proposed species conversion to rear all-female, triploid Rainbow Trout/steelhead. These documents update the 1990 PEIS.

Because the existing Cooke Aquaculture net pens have previously obtained the facility construction permits as well as Marine Finfish Aquaculture Permits, Clean Water Act Section 402 NPDES Waste Discharge Permits, and State-owned Aquatic Land Leases, there is no site selection or construction required to implement the species conversion proposal. There will be only minor operational differences to farm all-female triploid Rainbow Trout/steelhead rather than farming Atlantic salmon, with insignificant impacts to elements of the Natural Environment, and no change in impact to elements of the Built Environment as a result of implementing the species conversion proposal.

The company will use local stocks of Rainbow Trout/steelhead produced by Troutlodge hatcheries in Pierce County. Brood stock are cultivated in Washington specifically for the production of ova to supply both private and public aquaculture operations. Troutlodge, a Washington-based company, has been producing Rainbow Trout/steelhead eggs for sale to farms and public enhancement hatcheries throughout the world since 1945. Brood fish are raised in regulated pathogen-free conditions for their entire life cycle. The company utilizes a comprehensive health testing and disease-free certification program that exceeds World Organization of Animal Health (OIE) standards at their Washington facilities, allowing them to export live salmonid eggs throughout the world.

Troutlodge has been producing mono-sex (all-female) populations of Rainbow Trout/steelhead eggs since the mid-1990s. The all-female (XX only) ova are subsequently fertilized with X-only mono-milt. Triploidy is induced by mechanical pressure shock. For a short period of time, a high-pressure hydrostatic shock is applied to the newly fertilized eggs at a specified time point post-fertilization. The post-fertilization pressure treatment forces the fertilized egg to retain the third set of chromosomes that is normally ejected at this time. Pressure is then released and the triploid (3N) eggs are allowed to continue development. Ploidy is confirmed using a fluorescent nucleic acid label on either embryo or blood tissue using a flow cytometer at the Washington State University School of Veterinary Medicine. Testing results of Troutlodge triploid fish and eggs over a period of five (5) years (from 2013 to 2018) demonstrate a high rate of success in triploid induction (99.83% – 2,950 of 2,955 fish and/or eggs sampled (see Attachment A).

Cooke operates two freshwater hatcheries in the Scatter Creek area of Thurston County. The hatcheries raise and produce the juvenile fish that are eventually transferred to the marine net pens for final cultivation to the desired harvest size. Eyed all-female triploid Rainbow Trout/steelhead eggs would be supplied to the Cooke hatcheries from the Troutlodge hatchery under a WDFW Fin Fish Transport permit. The eggs would be hatched and cultured to a certain size in the Cooke hatcheries, and then transferred to the marine net pens after undergoing the necessary fish pathogen screening protocols, subject to review and approval by WDFW. Thereafter, a Fin Fish Transport Permit would be required from WDFW for each specified lot of fish to be transferred from Cooke hatcheries to the marine net pens.

Marine net pen cultivation and production protocols for Rainbow Trout/steelhead are basically the same as those used for Atlantic Salmon. Maximum cage density levels are expected to be managed at the same levels (approximately 0.9 to 1.2 lb/ft^3 or 15 – 20 kg/m^3), resulting in comparable maximum biomass levels that have historically been attained at each of the existing Cooke Aquaculture sites. Depending upon fish size at harvest (targeted mean weights of approximately 7 to 9 lbs or 3.5 to 4.2 kg), the fish population sizes at each marine net pen site are expected to be similar to stocking levels for Atlantic Salmon. The fish feed composition for marine-reared trout diets will be the same or similar to the currently-used marine salmon diets. Modern salmon and trout feeds are composed of highly digestible ingredients that are specifically formulated for optimal growth and feed conversion rates. No differences in water quality or sediment quality are expected to result from this change in species or the accompanying Rainbow Trout/steelhead-specific feeds that would be used. Additional information on feed composition, expected feed conversion rates, projected growth rates, projected pen densities and production cycles is provided in Section D of the SEPA Additional Information document (Attachment B).

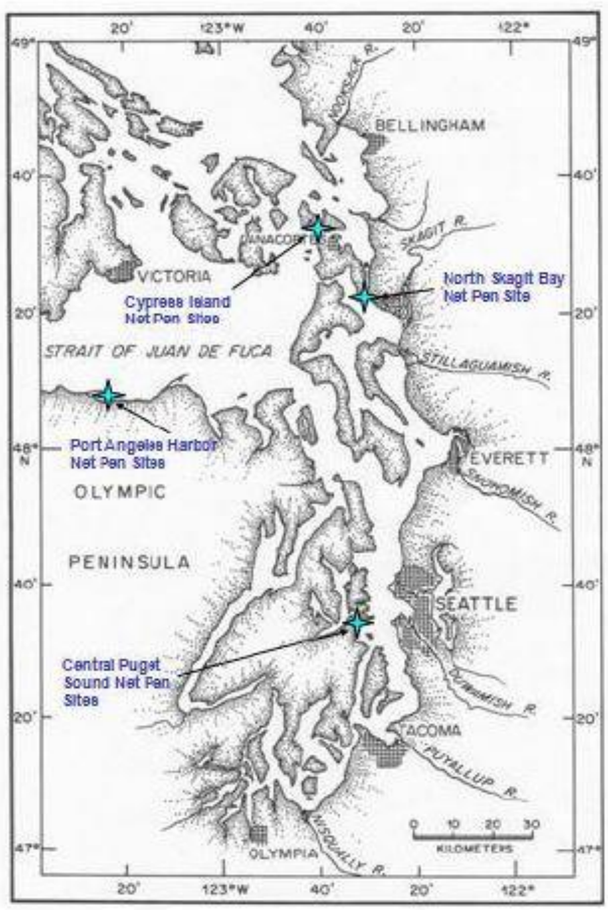
Equipment and all net pen support structures, stock nets, and predator exclusion nets will be the same or similar to what has been used for the cultivation and production of Atlantic Salmon. If the species conversion proposal is approved, the Rainbow Trout/steelhead produced in marine net pens will be harvested, processed, packaged and shipped fresh to seafood customers throughout the United States. Cooke is dedicated to producing a high-quality seafood in a sustainable and environmentally sound manner.

12. Location of the proposal. Give sufficient information for a person to understand the precise location of your proposed project, including a street address, if any, and section, township, and range, if known. If a proposal would occur over a range of area, provide the range or boundaries of the site(s). Provide a legal description, site plan, vicinity map, and topographic map, if reasonably available. While you should submit any plans required by the agency, you are not required to duplicate maps or detailed plans submitted with any permit applications related to this checklist.

Floating commercial marine net pens owned by Cooke Aquaculture Pacific, LLC are located in the following areas:

Fort Ward, Orchard Rocks and Clam Bay – Rich Passage in Kitsap County on the south end of Bainbridge Island, WA.

Hope Island – Skagit Bay in Skagit County on the northeast side of Hope Island, WA.[4]



Vicinity Map

4 Cooke Aquaculture Pacific is working with the Department of Natural Resources to resolve State-owned Aquatic Land Lease issues with the Cypress Island net pen and the Port Angeles net pen facilities. If the situation is resolved, these net pen sites would also be transitioned to native species aquaculture.

B. Environmental Elements

1. *Earth*

a. General description of the site:
(circle one): Flat, rolling, hilly, steep slopes, mountainous, other

Not applicable.

b. What is the steepest slope on the site (approximate percent slope)?

Not applicable.

c. What general types of soils are found on the site (for example, clay, sand, gravel, peat, muck)? If you know the classification of agricultural soils, specify them and note any agricultural land of long-term commercial significance and whether the proposal results in removing any of these soils.

Not applicable.

d. Are there surface indications or history of unstable soils in the immediate vicinity? If so, describe.

There have been no indications of unstable soils at the existing marine net pen sites.

e. Describe the purpose, type, total area, and approximate quantities and total affected area of any filling, excavation, and grading proposed. Indicate source of fill.

No new construction is proposed; therefore, there would be no fill, excavation, or grading at the existing marine net pen sites..

f. Could erosion occur as a result of clearing, construction, or use? If so, generally describe.

Not applicable. No new construction is proposed.

g. About what percent of the site will be covered with impervious surfaces after project construction (for example, asphalt or buildings)?

The "impervious surfaces" question is not applicable to the existing floating marine net pens, or to the change in species proposal.

h. **Proposed measures to reduce or control erosion, or other impacts to the earth, if any**:

Since no erosion or other impacts to earth would occur, no measures to reduce or control such impacts are proposed.

2. Air

a. What types of emissions to the air would result from the proposal during construction, operation, and maintenance when the project is completed? If any, generally describe and give approximate quantities if known.

Diesel-powered electrical generators (gensets) are used on the net pen sites to supply power to the feeding equipment. There is typically one genset for each net pen site. Typical hours of operation are from 7:00 AM to 5:00 PM. The generators may run continuously during that time or may be turned off during times when they are not in use. This represents no change from existing conditions, and would not be altered by the change in species proposal.

b. Are there any off-site sources of emissions or odor that may affect your proposal? If so, generally describe.

No.

c. Proposed measures to reduce or control emissions or other impacts to air, if any:

The proposal to change the species of fish reared at existing marine net pen sites would cause no air quality impacts; therefore, no measures are proposed to reduce or control such impacts.

The diesel generators that are used are new models that use EPA Tier IV type engines and run on clean fuels. Gensets are periodically shut off when they are not in use. This represents no change from existing conditions and would not be altered by the change in species proposal.

3. Water

a. Surface Water:

1) **Is there any surface water body on or in the immediate vicinity of the site (including year-around and seasonal streams, saltwater, lakes, ponds, wetlands)? If yes, describe type and provide names. If appropriate, state what stream or river it flows into**.

Cooke Aquaculture existing floating net pens are located in the marine waters of Rich Passage in Kitsap County, Skagit Bay in Skagit County, Deepwater Bay in Skagit County and Port Angeles Harbor in Clallam County (see Vicinity Map in Section A.12, above).

2) Will the project require any work over, in, or adjacent to (within 200 feet) the described waters? If yes, please describe and attach available plans.

The proposed project involves changing the species of finfish being reared at existing Cooke Aquaculture Puget Sound marine net pen facilities from Atlantic Salmon to an all-female sterile stock of Rainbow Trout/steelhead. These fish will be grown in the same existing net pen facilities that the Atlantic Salmon were grown in, using the same feeding and fish culturing practices that are currently employed at these facilities. There will be no in-water work or over-water work to construct or modify existing floating marine net pens.

3) Estimate the amount of fill and dredge material that would be placed in or removed from surface water or wetlands and indicate the area of the site that would be affected. Indicate the source of fill material.

None. No new construction is proposed.

4) Will the proposal require surface water withdrawals or diversions? Give general description, purpose, and approximate quantities if known.

No surface water withdrawals or diversions are required to implement the species change proposal, or to continue operations at existing floating net pen facilities.

5) Does the proposal lie within a 100-year floodplain? If so, note location on the site plan.

No, not applicable.

6) Does the proposal involve any discharges of waste materials to surface waters? If so, describe the type of waste and anticipated volume of discharge.

Existing permitted marine net pens will be used to rear all-female triploid Rainbow Trout/steelhead following the transition from Atlantic Salmon culture. Marine finfish rearing facilities in Washington State operate under the regulations and conditions of a National Pollutant Discharge Elimination System (NPDES) permit issued by the Washington Department of Ecology (Ecology). The Clean Water Act Section 402 NPDES Waste Discharge Permit establishes the basis for effluent limitations, water quality criteria, sediment management standards, and the technology-based standards for allowable discharges from the fish rearing facilities. Ecology issued updated the Cooke Aquaculture NPDES permits on July 11, 2019 for the Hope Island, Fort Ward, Orchard Rocks and Clam Bay net pen facilities. The permits set forth additional monitoring, reporting and recording keeping requirements for the protection of public and environmental health, *"This proposed permit (NPDES) includes all statutory requirements for Ecology to authorize a wastewater discharge. The permit includes limits and conditions to protect human health and aquatic life, and the beneficial uses of waters of the state of Washington."* Clam Bay NPDES Permit WA-0031526 Fact Sheet (dated July 11, 2019).[5]

Net pen operators manage nutrient waste with farm practices, efficient feeds and feeding practices, optimal pen configurations and farm orientation in order to optimize fish growth, waste distribution, and nutrient assimilation by the food web (Rust et al., November 2014). Impacts to water quality at salmon farm sites, including increased nitrogen, phosphorus, lipids, and turbidity or oxygen depletion have lessened significantly over the last 20 years as a result of a better understanding of siting requirements, improved feeding, better feed formulation, and better farm management practices (Soto and Norambuena 2004 in Rust et al., November 2014; McKinnon et al 2008 in Rust et al., November 2014; and Price and Morris 2013). Benthic impacts from U.S. net pens have been dramatically reduced over the last few decades, due to improved siting and better management practices (Rust et al., November 2014).

The primary discharge from marine salmonid net pen facilities, which includes Rainbow Trout/steelhead farms, is organic nutrients produced by the cultivated populations of fish that are being reared at the facility. These metabolic waste products are primarily composed of organic carbon, nitrogen and trace amounts of phosphorus. Switching from growing one species of salmonid (Atlantic Salmon) to a different closely-related species of salmonid (Rainbow Trout/steelhead) is not expected to result in changes to the composition or volume of waste discharge. Commercial salmon and trout diets contain the same or similar feed ingredients. Some commercial trout feeds tend to have slightly lower lipid- (fat) to-protein amounts in comparison to some salmon feeds. Cooke anticipates using the same commercial salmon feed for the all-female triploid Rainbow Trout/steelhead diets that

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[5] The NPDES Fact Sheets issued for all four locations with existing valid State-owned Aquatic Land Leases contain this same statement. The Fact Sheets are included by reference.

it is currently using to grow Atlantic Salmon at their marine net pen sites. Projected production cycles, the time from first fish in to last fish out (all harvesting completed), are anticipated to be less by several months compared to current Atlantic Salmon operations. This will be the result of both the expected growth rates for all-female triploid Rainbow Trout/steelhead being higher (faster growing compared to Atlantic Salmon), and the average harvest weight for Rainbow Trout/steelhead being smaller (approximately an 8-pound average weight for Rainbow Trout/steelhead compared to the approximately 11-pound average weight for Atlantic salmon). The overall amount of feed used during a production cycle and the period of peak biomass are anticipated to be less than with current Atlantic Salmon farming operations. In addition, because of the shortened production cycle for raising Rainbow Trout/steelhead, the frequency and number of fallowings will be increased over a given time period in comparison to the current Atlantic Salmon farming operations. Anticipated volumes of discharge from the marine net pens are therefore expected to be no more than and likely less than current peak levels. Additional information on feed composition, expected feed conversion rates, projected growth rates, projected pen densities and production cycles is provided in the SEPA Additional Information document (Attachment B, in the response to Information Request D.2).

b. Ground Water:

1) **Will groundwater be withdrawn from a well for drinking water or other purposes? If so, give a general description of the well, proposed uses and approximate quantities withdrawn from the well. Will water be discharged to groundwater? Give general description, purpose, and approximate quantities if known**.

No groundwater is used and no water will be discharged to groundwater as a result of the species change proposal.

2) **Describe waste material that will be discharged into the ground from septic tanks or other sources, if any (for example: Domestic sewage; industrial, containing the following chemicals. . . ; agricultural; etc.). Describe the general size of the system, the number of such systems, the number of houses to be served (if applicable), or the number of animals or humans the system(s) are expected to serve**.

No waste materials will be discharged into the ground.

c. Water runoff (including stormwater):

1) **Describe the source of runoff (including storm water) and method of collection and disposal, if any (include quantities, if known). Where will this water flow? Will this water flow into other waters? If so, describe**.

Marine net pen structures are a grid constructed of steel support structures from which the nets are suspended in which the fish are grown. Approximately 75% of the structure is open, unencumbered surface area. The steel walkway structures are composed of permeable surfaces (open steel grating for the walkway grid). There is no stormwater collection at the marine net pen sites, and none required for the species change proposal.

2) Could waste materials enter ground or surface waters? If so, generally describe.

See the response to SEPA Checklist Questions B.3.a.6 above, and B.7.a below.

3) Does the proposal alter or otherwise affect drainage patterns in the vicinity of the site? If so, describe.

No drainage patterns would be affected by the proposed action.

d. Proposed measures to reduce or control surface, ground, and runoff water, and drainage pattern impacts, if any:

Equipment used on the floating marine net pens has secondary containment provisions. Spill prevention, control and response procedures are posted at all sites. Spill kits are located on each site and employees are given instruction on spill response procedures.

Computerized feeding systems will be used to accurately keep track of how much the fish population is expected to eat, and how much feed is being fed (delivered) to the fish population in each pen. Fish feeding technicians will monitor each fish pen using both surface visual cues and underwater video cameras that allow them to ensure fish are being fed properly, without excess pellets sinking past the captive stocks and being lost into the environment. Fish feed is one of the most expensive costs in raising salmon. For this reason, the industry has researched and developed improved fish feeds, feed monitoring equipment, feed distribution equipment, computerized biomass growth programs, expected feed conversion rates, specific feed delivery rates, and several other tools and methods to maximize feed utilization for the growth of the fish stocks, while minimizing the chance of uneaten feed entering the environment. There would be no change to existing feed delivery systems or feed monitoring practices to implement this species change proposal.

4. Plants

a. Check the types of vegetation found on the site:

____ deciduous tree: alder, maple, aspen, other
____ evergreen tree: fir, cedar, pine, other
____ shrubs
____ grass
____ pasture
____ crop or grain
____ orchards, vineyards or other permanent crops
____ wet soil plants: cattail, buttercup, bullrush, skunk cabbage, other
____ water plants: water lily, eelgrass, milfoil, other
X other types of vegetation: Marine algae grows on the net pen structures such as floats, netting and anchor lines. This includes primarily green and brown algae species.

b. What kind and amount of vegetation will be removed or altered?

None.

c. List threatened and endangered species known to be on or near the site.

There are no threatened or endangered plant species on the existing marine net pen sites.

d. Proposed landscaping, use of native plants, or other measures to preserve or enhance vegetation on the site, if any:

Since there will be no impacts to plants or marine algae that grows on net pen structures as a result of the species change proposal, no measures are proposed to preserve or enhance vegetation on the existing marine net pen sites.

e. List all noxious weeds and invasive species known to be on or near the site.

None.

5. Animals

a. List any birds and other animals which have been observed on or near the site or are known to be on or near the site. Examples include:

birds: hawk, heron, eagle, songbirds, other: waterfowl, shorebirds
mammals: deer, bear, elk, beaver, other: marine mammals (see below)
fish: bass, salmon, trout, herring, shellfish, other: (see below)

Marine mammals, fish, birds and invertebrates can occur around existing marine net pen areas. These primarily include: whales, porpoise, seals, sea lions; several species of salmonids, baitfish, and ground fish; bald eagles, shorebirds and waterfowl. The occurrence of these animals will not be altered by the species change proposal.

b. List any threatened and endangered species known to be on or near the site.

Common Name	Federal Species Status	State Species Status
Humpback Whale	Endangered	Endangered
Southern Resident Killer Whale	Endangered	Endangered
Northern Sea Otter	None[6]	Endangered
Eastern Pacific Stellar Sea Lion	None[7]	None
Bald Eagle	None[8]	None
Marbled Murrelet	Threatened	Threatened
Chinook Salmon (Puget Sound ESU)	Threatened	Candidate
Chum Salmon (Hood Canal Summer-run)	Threatened	Candidate
Chum Salmon (Puget Sound/Strait of Georgia ESU)	None[9]	Candidate
Coho Salmon	Species of concern	None
Steelhead	Threatened	Candidate
Bull Trout	Threatened	Candidate
Eulachon	Threatened	Candidate
Flathead Sole	Species of concern	None
Green Sturgeon	Threatened	None
Pacific Cod	Species of concern	Candidate
Pacific Hake	Species of concern	Candidate
Pacific Lamprey	None	None

6 Northern Sea Otter presently have no Federal listing status. They are protected under the Marine Mammal Protection Act (https://www.fws.gov/wafwo/articles.cfm?id=149489657).

7 Eastern Pacific Stellar Sea Lion presently have no Federal or State listing status. They are protected under the Marine Mammal Protection Act (WDFW, August 2008 [updated January 2019]: Priority habitats and species list).

8 Bald Eagle presently have no Federal or State listing status. They are protected under the Bald and Golden Eagle Protection Act (WDFW, August 2008 [updated January 2019]: Priority habitats and species list).

9 WDFW, August 2008 (updated January 2019).

Common Name	Federal Species Status	State Species Status
River Lamprey	None	Candidate
Canary Rockfish	None[10]	Candidate
Bocaccio Rockfish	Endangered	Candidate
Yelloweye Rockfish	Threatened	Candidate
Pinto Abalone	Species of concern	Candidate
Pacific Harbor Porpoise	None	Candidate
Gray Whale	None	Sensitive
Pacific Herring	None	Candidate
Black Rockfish	None	Candidate
Brown Rockfish	None	Candidate
China Rockfish	None	Candidate
Copper Rockfish	None	Candidate
Quillback Rockfish	None	Candidate
Redstripe Rockfish	None	Candidate
Tiger Rockfish	None	Candidate
Yellowtail Rockfish	None	Candidate
Walleye Pollock (South Puget Sound)	None	Candidate
Olympia Oyster	None	Candidate

A narrative description of each of these species is provided in SEPA Checklist Attachment C: *Threatened and Endangered Species: 1990 PEIS Update*, along with an effects analysis for both Atlantic Salmon culture (which has occurred throughout the years since the 1990 PEIS was issued), and an effects analysis for the proposed species conversion to rear all-female, triploid Rainbow Trout/steelhead.

WDFW raised several specific questions about the potential effects of the species conversion proposal on threatened and endangered species (TES). Responses to these questions are provided in SEPA Checklist Attachment B: *Additional Information: Response to WDFW Questions*, supported by an *Annotated Bibliography* (Attachment C). The *Annotated Bibliography* was prepared by technical experts who also assisted with preparing the response to WDFW questions in Attachment B, and who prepared the TES effects analysis in Attachment D. Curriculum vitae for these experts are provided in SEPA Checklist Attachment E.

c. Is the site part of a migration route? If so, explain.

Yes. The existing marine net pens are located in the marine waters of Puget Sound and while not blocking any migration corridors it is likely that marine fish species, marine mammals and other wildlife may pass by in the water adjacent to the net pens.

d. Proposed measures to preserve or enhance wildlife, if any:

The following measures are current practices at Cooke Aquaculture existing marine net pen sites and would continue to be implemented with the species change proposal.

- Monitor for organic enrichment of the substrate beneath the farm. This is required by the Clean Water Act Section 402 NPDES Waste Discharge Permit for the project.
- Implement fallow periods at marine net pen operations to allow for sediment and benthic organism

[10] Genetic information collected by NMFS found that Canary Rockfish in the Puget Sound/Georgia Basin are not distinct from coastal Canary Rockfish, and therefore are not a Distinct Population Segment (DPS). To qualify for Federal ESA protections, a species must be discrete from the remainder of the species (NOAA Fisheries. 2017).

recovery periods from any excess nutrient enrichment. Fallowing for a period of 30 days or longer would occur after each single-stock generation is grown and harvested out. It is anticipated that the frequency of fallowing a site would increase as Rainbow Trout/steelhead grow-out cycles are anticipated to be shorter compared to Atlantic Salmon to reach the targeted market size.

- Use passive predator barrier netting surrounding each marine net pen for both avian and marine mammal deterrence.
- Tightly tension predator barrier nets and fish containment nets using a net weighting system to keep the net walls and floors tight. This minimizes the potential for underwater entanglement of marine mammals or diving birds.
- Efficiently plan vessel trips to minimize vessel traffic to and from marine net pen sites to reduce potential wildlife interactions or disturbance.
- Observe Federal regulations that implement a no-approach zone for killer whales and all other whales, dolphins, and porpoises when operating crew vessels approaching or leaving marine net pen sites.
- Implement the *Plan of Operation: All-Female Triploid Trout* (January 18, 2019), including updating the comprehensive Operational Procedures for Fish Escape Prevention, Reporting and Recapture and Regulated Fish Pathogen Reporting and Bio-security Plans.

e. List any invasive animal species known to be on or near the site.

None known.

6. Energy and Natural Resources

a. What kinds of energy (electric, natural gas, oil, wood stove, solar) will be used to meet the completed project's energy needs? Describe whether it will be used for heating, manufacturing, etc.

Electrical energy is supplied to existing marine net pen sites by diesel generators. The diesel generators run periodically during the work day to power feeding equipment or other small appliances. The species change proposal would require no change from these existing conditions.

b. Would your project affect the potential use of solar energy by adjacent properties? If so, generally describe.

No.

c. What kinds of energy conservation features are included in the plans of this proposal? List other proposed measures to reduce or control energy impacts, if any:

Electrical generators are operated only as needed and would be turned off when not in use. The species change proposal would require no change to this practice.

7. Environmental Health

a. Are there any environmental health hazards, including exposure to toxic chemicals, risk of fire and explosion, spill, or hazardous waste, that could occur as a result of this proposal? If so, describe.

Yes. All existing facilities utilize diesel fuel to operate various pieces of machinery located on the floating marine net pens and/or on support barges. There is a risk of spill when handling liquid fuels. Fuel is kept inside secondary containment structures and spill kits are kept on each site nearby these areas. Employees are trained in proper fuel handling, spill prevention, and cleanup procedures. The species change proposal would require no change to these practices.

1) **Describe any known or possible contamination at the site from present or past uses**.

None known.

2) **Describe existing hazardous chemicals/conditions that might affect project development and design. This includes underground hazardous liquid and gas transmission pipelines located within the project area and in the vicinity**.

None.

3) **Describe any toxic or hazardous chemicals that might be stored, used, or produced during the project's development or construction, or at any time during the operating life of the project**.

Primary hazardous chemicals used on the existing marine net pen sites are those contained within or used for the operation of various pieces of gasoline- or diesel-powered engines. These fuels, lubricants and coolants include diesel fuel, gasoline fuel, motor oils, hydraulic oil, antifreeze and other lubricants. Periodic maintenance activities on the cage structures are occasionally performed using paints or solvents. Necessary precautions and preventative measures are taken to keep these materials from entering the water. These materials are not stored on the floating net pens when not in use. The species change proposal would require no change to these current practices.

Medicated feed that contains antibiotics is used to treat specific disease events at the marine net pens. Medicated feed use is infrequent, and the prescribed medicated feed treatments occur in the form of either 5- or 10-day treatments depending upon the type of antibiotic used. There are currently only three types of antibiotics used at the marine net pens: Romet TC, Terramyacin and Aquaflor. Currently, 100% of the fish are vaccinated at the hatchery for common pathogens prior to being transported to the marine net pens which helps to minimize the incidence of disease and the need for antibiotic treatments at the net pens. These practices would continue to be applied to the stocks of Rainbow Trout/steelhead. The amount, frequency or types of medicated feeds are not expected to increase. Annual medicated feed use has trended downward over the past 10 years (from 2008 to 2018).

4) **Describe special emergency services that might be required**.

There is no anticipated need for new or additional emergency services associated with the proposed change of species to be reared at the marine net pens. Employees working at the farm sites are currently trained in first aid and CPR. Emergency communication devices are located at all existing sites and in crew support vessels. First aid equipment, Automatic Electrical

Defibrillators (AEDs), fire suppression and firefighting equipment, sea survival equipment, and emergency oxygen delivery kits are kept at the sites and/or on board the various support vessels. Minor employee injuries are treated on-site. Medical transport, if needed, can be provided by company crew vessels and by coordination with local land-based Emergency Medical Technicians (EMTs). An emergency meeting point at appropriate shore-side facilities can be arranged with local EMTs if needed. In the event of a serious dive accident, the U.S. Coast Guard would likely be called for assistance to provide rapid helicopter transportation directly from a marine net pen facility to a hyperbaric chamber at Harborview Hospital in Seattle.

Emergency spill prevention, response plans and procedures are posted at each of the existing marine net pen facilities along with emergency spill notification telephone numbers for State and Federal agencies. Emergency spill kits are stored and readily available near fueling areas. In the event of an accidental fuel spill, a marine fuel spill response vessel service would be immediately contacted for assistance, and State and Federal notification procedures would be implemented (Washington Department of Ecology and U.S. Coast Guard National Response Center). Certain employees are HAZWOPPER certified and capable of deploying the on-site spill prevention kits and clean-up materials for initial spill containment response. No change to these procedures would be required to implement the species change proposal.

5) **Proposed measures to reduce or control environmental health hazards, if any**:

Cooke Aquaculture has developed a Spill Prevention, Control and Response procedure for the existing marine net pen sites. Employees are trained and familiar with proper spill prevention procedures. Spill kits are located on each of the farm sites, and emergency spill notification contact numbers are posted in numerous locations. Double containment is used for fuel tanks and hazardous materials located on the floating marine net pen sites. None of these conditions would require any change to implement the species change proposal.

b. Noise

1) What types of noise exist in the area which may affect your project (for example: traffic, equipment, operation, other)?

None.

2) What types and levels of noise would be created by or associated with the project on a short-term or a long-term basis (for example: traffic, construction, operation, other)? Indicate what hours noise would come from the site.

Gasoline- and diesel-powered engines are the primary sources of noise associated with the operation of the existing marine net pen sites. The diesel generator engines are contained within sound enclosures and are considered the “run quiet” type of systems used for backup generators at hospitals and similar facilities. Hours of operation are typically normal working hours from 7:00 AM to 6:00 PM. None of these existing practices would be altered by the species change proposal.

3) Proposed measures to reduce or control noise impacts, if any:

Since there would be no noise impacts associated with the species change proposal, no measures are proposed to reduce or control such impacts.

Diesel generators associated with existing marine net pen facilities are operated only as needed.

8. Land and Shoreline Use

a. **What is the current use of the site and adjacent properties? Will the proposal affect current land uses on nearby or adjacent properties? If so, describe**.

The proposed change in species to be reared would utilize existing permitted commercial net pen aquaculture facilities that have been in operation since the early 1980s. Adjacent upland properties are typically developed with residential homes. Exceptions include the Clam Bay facility (in existence since the early 1970s), located adjacent to property owned by the U.S. Government; the Port Angeles net pens located near U.S. Government property and within a working harbor area; and the Cypress fish pens located adjacent to State-owned uplands (see the Vicinity Map in Section A.12 of this SEPA Checklist). Residential development along the waterfront and adjacent to the net pens has continued to occur over the past 30 to 40 years in areas where residential development is zoned. The pens are anchored offshore and do not interact with nearby land uses. The species change proposal would be undetectable to adjacent land uses.

b. Has the project site been used as working farmlands or working forest lands? If so, describe. How much agricultural or forest land of long-term commercial significance will be converted to other uses as a result of the proposal, if any? If resource lands have not been designated, how many acres in farmland or forest land tax status will be converted to nonfarm or nonforest use?

The Washington State Legislature has defined aquaculture as agriculture (RCW 15.85.010). As such, the existing marine net pen facilities could be considered working farmlands. The project application is to maintain the existing agricultural production of farmed fish by transitioning to a native sterile stock of fish species (Rainbow Trout/steelhead).

1) Will the proposal affect or be affected by surrounding working farm or forest land normal business operations, such as oversize equipment access, the application of pesticides, tilling, and harvesting? If so, how:

No.

c. Describe any structures on the site.

See attached drawings.

d. Will any structures be demolished? If so, what?

No structures will be demolished.

e. What is the current zoning classification of the site?

The existing net pen sites are all located in navigable waters that are designated Aquatic. Adjacent upland zoning is as follows:

- Hope Island Site: Adjacent upland zoning classification is Rural Residential (Skagit County).
- Fort Ward and Orchard Rocks Sites: Adjacent upland zoning classification is Shoreline Residential (City of Bainbridge Island).
- Clam Bay Site: Adjacent upland zoning classification is Military (Kitsap County).
- Port Angeles Site: Adjacent upland zoning classification is City (Clallam County).

- Cypress Island Site: Adjacent upland zoning classification is Public (Skagit County).

f. What is the current comprehensive plan designation of the site?

Adjacent upland comprehensive plan designations are:

- Hope Island Site: Shoreline of Statewide Significance
- Fort Ward and Orchard Rocks Sites: Shoreline Residential
- Clam Bay Site: Military
- Port Angeles Site: City
- Cypress Island Site: Public Open Space.

g. If applicable, what is the current shoreline master program designation of the site?

- Hope Island Site: Aquatic
- Fort Ward and Orchard Rocks Sites: Shoreline Residential
- Clam Bay Site: Military
- Port Angeles Site: Aquatic
- Cypress Island Site: Aquatic.

h. Has any part of the site been classified as a critical area by the city or county? If so, specify.

Unknown by applicant.

i. Approximately how many people would reside or work in the completed project?

Each net pen site is staffed with approximately 8 full time employees. There would be no change in the number of workers as a result of implementing the species change proposal.

j. Approximately how many people would the completed project displace?

None.

k. Proposed measures to avoid or reduce displacement impacts, if any:

Since there would be no displacement impacts, no measures are proposed to avoid or reduce such impacts.

l. Proposed measures to ensure the proposal is compatible with existing and projected land uses and plans, if any:

Existing net pens where the change of species is proposed have all necessary land use permits and in-water permits in-hand, having operated at these locations since the mid-1970s and mid-1980s. The proposal does not create new impacts to existing or projected land uses or land use plans.

m. Proposed measures to reduce or control impacts to agricultural and forest lands of long-term commercial significance, if any:

Since there would be no impacts to agricultural or forest land, no measures are proposed to reduce or control such impacts.

9. Housing

a. Approximately how many units would be provided, if any? Indicate whether high, middle, or low-income housing.

None.

b. Approximately how many units, if any, would be eliminated? Indicate whether high, middle, or low-income housing.

None.

c. Proposed measures to reduce or control housing impacts, if any:

Since there would be no impacts to housing, no measures are proposed to reduce or control such impacts.

10. Aesthetics

a. What is the tallest height of any proposed structure(s), not including antennas; what is the principal exterior building material(s) proposed?

The Hope Island and Clam Bay net pen sites have an associated feeding barge that is moored to the fish pen structure. The approximate height of the feed barges is 14 feet above sea level. The net pen structures all have a perimeter barrier net that is approximately 6 to 8 feet above the water level. No change to these structures is required to implement the species change proposal.

b. What views in the immediate vicinity would be altered or obstructed?

The existing fish pens are located from approximately 600 feet to 1,500 feet offshore. They do not obstruct views. View obstruction would not be an impact of the species change proposal.

c. Proposed measures to reduce or control aesthetic impacts, if any:

Since there would be no aesthetic impacts associated with the species change proposal, no measures are proposed to reduce or control such impacts.

The net pen structures use dark or grey materials to help minimize visual impacts. The farm sites are kept clean and orderly.

11. Light and Glare

a. What type of light or glare will the proposal produce? What time of day would it mainly occur?

Existing marine net pen facilities are required by the U.S. Coast Guard to maintain navigation warning signal lights under a Private Aids to Navigation (PATON) permit. Each site has a small flashing yellow navigation light that is automatically activated by darkness. The light flashes on every 7 seconds and only operates at night. There are no other surface lights used on the fish pens. No change to existing navigation lights would be required to implement the species change proposal.

Underwater lights (visible as a glow in the water above the sea surface were used seasonally in Atlantic Salmon culture to suppress sexual maturation of the winter entry smolts during their second sea-winter in the 18- to 24-month rearing cycle. Underwater lights will not be used with the species change proposal to rear Rainbow Trout/steelhead because the all-female triploid fish do not go into sexual maturation.

b. Could light or glare from the finished project be a safety hazard or interfere with views?

No.

c. What existing off-site sources of light or glare may affect your proposal?

None.

d. Proposed measures to reduce or control light and glare impacts, if any:

The use of lights on support vessels could be minimized if these vessels are used during the evening hours. There would be no change to this practice as a result of implementing the species change proposal.

12. Recreation

a. What designated and informal recreational opportunities are in the immediate vicinity?

Informal recreational opportunities nearby existing net pen facilities include recreational boating, fishing, wildlife viewing and beach combing.

b. Would the proposed project displace any existing recreational uses? If so, describe.

No. The proposal to change the species reared at existing marine net pen facilities would have no effect on existing recreational uses. There will be no change to the size and/or location of existing net pen structures in order to implement the species change proposal; therefore, there would be no change to existing recreational opportunities adjacent to these facilities.

While existing facilities occupy space on the surface of the water, there is a massive amount of water surface area in comparison that is available for navigation and recreational opportunities around the fish pens.

c. Proposed measures to reduce or control impacts on recreation, including recreation opportunities to be provided by the project or applicant, if any:

Since the species change proposal would result in no impacts to recreation, no measures are proposed to reduce or control such impacts.

Potential conflicts between existing marine net pens and aquatic recreation opportunities are minimized by the use of navigation marker lights and the identification of these lights on NOAA navigation charts.

13. Historic and cultural preservation

a. Are there any buildings, structures, or sites, located on or near the site that are over 45 years old listed in or eligible for listing in national, state, or local preservation registers ? If so, specifically describe.

No.

b. Are there any landmarks, features, or other evidence of Indian or historic use or occupation? This may include human burials or old cemeteries. Are there any material evidence, artifacts, or areas of cultural importance on or near the site? Please list any professional studies conducted at the site to identify such resources.

No.

c. Describe the methods used to assess the potential impacts to cultural and historic resources on or near the project site. Examples include consultation with tribes and the department of archeology and historic preservation, archaeological surveys, historic maps, GIS data, etc.

There would be no change to existing floating marine net pen sites to implement the species change proposal; therefore, there would be no cause for any impact to cultural or historic resources.

d. Proposed measures to avoid, minimize, or compensate for loss, changes to, and disturbance to resources. Please include plans for the above and any permits that may be required.

Since there would be no impacts to cultural or historic resources as a result of the species change proposal, no measures are proposed to avoid, minimize, or compensate for loss, changes to, or disturbance to such resources.

14. Transportation

a. Identify public streets and highways serving the site or affected geographic area and describe proposed access to the existing street system. Show on site plans, if any.

Employees use public roads to get to work and meet in the morning at nearby marinas or at a private company dock to access existing fish pens by boat. Young fish (smolts) are transported from existing Cooke hatcheries to a vessel in Olympia Harbor for transport to existing marine net pen sites. These practices will be the same with the species change proposal as they are with existing operations that rear Atlantic Salmon.

b. Is the site or affected geographic area currently served by public transit? If so, generally describe. If not, what is the approximate distance to the nearest transit stop?

No.

c. How many additional parking spaces would the completed project or non-project proposal have? How many would the project or proposal eliminate?

None, not applicable.

d. Will the proposal require any new or improvements to existing roads, streets, pedestrian, bicycle or state transportation facilities, not including driveways? If so, generally describe (indicate whether public or private).

No.

e. Will the project or proposal use (or occur in the immediate vicinity of) water, rail, or air transportation? If so, generally describe.

See the response to SEPA Checklist Question B.14. a, above.

f. How many vehicular trips per day would be generated by the completed project or proposal? If known, indicate when peak volumes would occur and what percentage of the volume would be trucks (such as commercial and nonpassenger vehicles). What data or transportation models were used to make these estimates?

The Fort Ward, Orchard Rocks and Clam Bay sites are accessed from both the Bainbridge Island side of Rich Passage as well as the Manchester side of Rich Passage. Approximately 18 vehicle trips per day, 7 days per week occur on the Bainbridge Island side of Rich Passage. The company owns a dock facility located on Bainbridge Island as well as adjacent upland parking. Employees commute to work and park in the company parking lot where they can then be taken out to the Fort Ward, Orchard Rocks and Clam Bay net pens by company work vessels. A smaller number of employees at these facilities arrive via the Manchester side. Approximately 8 vehicle trips per day, 7 days per week occur on the Manchester side. The number of vessel trips to and from the Rich Passage cages each day varies but on average it is estimated to be 5 to 8 round trips per day. Trip generation at these sites would not change with implementation of the species change proposal.

The Hope Island employees arrive and park their vehicles in a parking lot at the Cornet Bay Marina located on the north end of Whidbey Island. The employees then board the company work vessel and are taken out to the Hope Island net pen site. Approximately 8 vehicle trips per day, 7 days per week occur. The number vessel trips to and from the Hope Island farm is estimated to be 2 to 4 round trips per day. Trip generation at this site would not change with implementation of the species change proposal.

The Port Angeles facility is accessed by boat. Employees park their vehicles in a parking lot adjacent to a small public boat launch located on Ediz Hook. A crew boat picks up the employees and takes them to the farm sites located just offshore of Ediz Hook. Trip generation at these sites would not change with implementation of the species change proposal.

The Cypress Island facilities are accessed by boat. Employees park their vehicles in a marina parking lot located in Anacortes. A crew boat picks up the employees and takes them to the farm sites which are located adjacent to Cypress Island. Trip generation at these sites would not change with implementation of the species change proposal.

Normal work hours on the existing marine net pen sites are 7:00 AM to 6:00 PM, seven days per week.

g. Will the proposal interfere with, affect or be affected by the movement of agricultural and forest products on roads or streets in the area? If so, generally describe.

No.

h. Proposed measures to reduce or control transportation impacts, if any:

Employees are encouraged to ride share to work in their personal vehicles. Once on the fish pens, the number of vessel trips are used efficiently. Multiple employees ride out to the fish pens at the same time and then return to shore at the same time. The efficient use of the vessels is encouraged. These existing practices will not be altered by the species change proposal.

15. Public Services

a. Would the project result in an increased need for public services (for example: fire protection, police protection, public transit, health care, schools, other)? If so, generally describe.

No. The existing marine net pen facilities do not require typical land-based public services, and none would be required to implement the species change proposal.

b. Proposed measures to reduce or control direct impacts on public services, if any.

Since there will be no impact on public services as a result of the species change proposal, no measures are proposed to reduce or control such impacts.

16. Utilities

a. Circle utilities currently available at the site: electricity, natural gas, water, refuse service, telephone, sanitary sewer, septic system, other <u>(See below)</u>.

The existing marine net pen facilities are self-sufficient. There are no public utilities installed on these sites. Port-O-Lets are maintained on the farm sites for sanitary wastes. A work vessel brings the cleaned and serviced Port-O-Lets out to the farms each week and exchanges them with the used ones. The used Port-O-Lets are taken to an upland support facility where they are cleaned and serviced. The work vessel removes all solid waste (garbage) materials from the farm sites and transports it to nearby upland dock facilities. The shore-side facilities are serviced by municipal waste and recycling collection services. Electricity is provided by diesel electric generators. Bottled drinking water is provided on the sites for the employees. None of these existing practices will be altered as a result of the species change proposal.

b. Describe the utilities that are proposed for the project, the utility providing the service, and the general construction activities on the site or in the immediate vicinity which might be needed.

None, not applicable.

C. Signature

The above answers are true and complete to the best of my knowledge. I understand that the lead agency is relying on them to make its decision.

Signature:

Name of signee: Kevin Bright

Position and Agency/Organization: Permit Coordinator, Cooke Aquaculture Pacific, LLC

Date Submitted: July 24, 2019

References

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Price, C.S. and J.A. Morris, Jr. 2013. *Marine Cage Culture and the Environment: Twenty-First Century Science Informing a Sustainable Industry*. National Oceanic and Atmospheric Administration. Technical Memorandum NOS-NCCOS-164. Silver Spring, MD.

RPS ASA. January 2016. *Port Angeles-East Fish Pens Biological Evaluation*. Prepared for Icicle Seafoods, Inc. Seattle, WA.

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Soto, D. and F. Norambuena. 2004. *Evaluation of Salmon Farming Effects on Marine Systems in the Inner Seas of Southern Chile: A Large-Scale Measurative Experiment*. Journal of Applied Ichthyology 20: 493-501. *In* Rust et al., November 2014.

Attachment A

Troutlodge Triploid Testing Results

Flow Cytometer Results - WSU (Thorgaard Lab)
Troutlodge Triploid Testing

Lot ID	Date Sampled	# of Samples	Results			
			2N Diploid	3N Triploid	Samples Not Run*	3N Triploid %
5233JSN (brow Well)	8/26/2013	100		100		100.0%
7183KCN (pacific)	9/16/2013	100	1	99		99.0%
8222KSN	11/4/2013	100		100		100.0%
8292KPN	11/4/2013	100		97	3	100.0%
10243SSN	1/6/2014	100		100		100.0%
2143SSN	5/12/2014	100		98	2	100.0%
2213SSN	5/12/2014	100		100		100.0%
8073KSN	5/29/2014	95		95		100.0%
9173KMIXN	12/1/2014	100		99	1	100.0%
9253KCN	12/1/2014	100		100		100.0%
9253SMIXN	12/1/2014	100	1	99		99.0%
12233SON	3/2/2015	100	1	92	7	98.9%
12183SON	3/2/2015	100		70	30	100.0%
12173SON	3/2/2015	100		91	9	100.0%
11142STL14TL	3/11/2015	81		81		100.0%
121153	6/10/2015	61		61		100.0%
128153	6/10/2015	70		70		100.0%
219153	6/17/2015	65		65		100.0%
304153	6/17/2015	65	1	64		98.5%
416153	7/14/2015	84		84		100.0%
10063	3/8/2016	100		99	1	100.0%
11183	2/8/2016	100		97	3	100.0%
8113	12/14/2016	90		90		100.0%
10063	12/14/2016	91		91		100.0%
9303	12/20/2016	87		85	2	100.0%
10273	3/1/2017	99		99		100.0%
11093	3/1/2017	89		89		100.0%
DT 17	5/11/2017	60		60		100.0%
WIN	5/11/2017	60		43	17	100.0%
DT5 FEB20170217	7/7/2017	64		63	1	100.0%
DT6 MAY20170412	7/7/2017	63		57	6	100.0%
02222SSFAM	10/16/2017	60		60		100.0%
912KPN	1/9/2018 Aug	80	1	79		98.8%
TERF1118PA3NG2	4/17/2018	60		55	5	100.0%
NOV2017AV3N	4/17/2018	60		59	1	100.0%
NOV2017PA2G2	4/17/2018	60		59	1	100.0%
		Totals	**5**	**2950**		
		2N and 3N Results	**0.17%**	**99.83%**		

***Samples not run. Typically due to insufficient volume of blood from swimup fry to process through flow cytometer.**

Attachment B

Additional Information:
Response to WDFW Questions

Attachment B to SEPA Checklist
Additional Information: Response to WDFW Questions

Cooke Aquaculture Pacific Marine Aquaculture Permit Application to Transition from Raising Atlantic Salmon to Raising Sterile All-Female Triploid Rainbow Trout/Steelhead at the Cooke Existing Marine Net Pen Sites in Puget Sound, Washington

INTRODUCTION

Cooke Aquaculture Pacific, LLC (Cooke) proposes to change the fish species being farmed at its seven (7) existing marine net pen aquaculture sites from Atlantic Salmon (*Salmo salar*) to domesticated stocks of mono-sex (all-female) sterile (triploid) Rainbow Trout (*Oncorhynchus mykiss*). While the common names Rainbow Trout and steelhead are often used interchangeably, this environmental review document and permit applications will refer to Rainbow Trout, which is the single official common name given to the species (*O. mykiss*) by the American Fisheries Society several years ago. All-female triploid Rainbow Trout proposed to be raised in the marine net pen facilities are a sterile, mono-sex stock of fish which reduces the risks of genetic interference to native populations. Sterile mono-sex fish stocks are also used in numerous public agency recreational fish stocking programs for this same reason.

Four of the existing Cooke Puget Sound marine net pen sites have valid permits for the commercial rearing of Atlantic Salmon in marine net pens. The salmon farms in Washington have been raising almost exclusively Atlantic Salmon for the past 30 years. A recent change in Washington State law (RCW 77.125.050), however, requires phasing out non-native finfish in marine net pen aquaculture. This change means commercial marine net pen production in Washington will need to find a commercially viable native species of fish to grow in order stay in the business of growing seafood for human consumption. Rainbow Trout are a native species to the Pacific Northwest region and have been commercially farmed in Washington, primarily in freshwater facilities, for more than 80 years, if not longer. Cooke is requesting re-approval of their Marine Finfish Aquaculture Permit (WAC 220-370-100) from the Washington Department of Fish and Wildlife (WDFW) that will allow the company to start growing domesticated stocks of a mono-sex, sterile Rainbow Trout (*Oncorhynchus mykiss*) at their marine farms. Cooke received the renewal of their Marine Finfish Aquaculture Permit from WDFW on March 19, 2019 allowing Cooke to continue raising Atlantic Salmon at the four farm sites (Clam Bay, Fort Ward, Orchard Rocks and Hope Island) with a valid Aquatic Use Permit from the Washington Department of Natural Resources (WDNR). Other than transitioning to the commercial farming of a different species of fish, the company is not planning any alteration to the existing fish pen physical structures, site locations, supporting equipment, or general current practices, methods, maintenance and cultivation techniques being used for growing Atlantic Salmon at the farms. Domesticated stocks of triploid Rainbow Trout/steelhead have very similar physiological and metabolic requirements to those of domesticated stocks of Atlantic Salmon. The basic difference is that all-female triploid Rainbow Trout are known to be reproductively sterile, and thus convert their energy almost entirely to growth. By comparison, diploid populations of Rainbow Trout and Atlantic Salmon will reach a certain age and begin to expend growth energy toward the production of gametes and secondary sexual characteristics (sexual maturation).

Raising native-stock fish species in marine net pens is consistent with the recommendations of the *Final Programmatic Environmental Impact Statement: Fish Culture in Floating Net-Pens* (Washington Department of Fisheries, January 1990). The Preferred Alternative in the Programmatic EIS (PEIS) also concludes that *"In areas where WDF determines there is a risk of significant interbreeding (with indigenous species) or establishment of harmful self-sustaining populations, the agency should only approve the farming of sterile* ***or*** *monosexual individuals, or genetically incompatible species."* By farming a mono-sex (all-female) and sterile (triploid) stock of Rainbow Trout/steelhead, the proposed species change in Cooke Puget Sound net pens incorporates both methods of reducing the risks of genetic interference to indigenous populations or from escaped farmed fish establishing self-sustaining feral populations. WDFW requested that a SEPA Checklist be prepared along with Additional Information (e.g., information to augment the 1990 Programmatic EIS) to compare the change in environmental impacts that may occur as a result of the change of species to be farmed in Cooke Aquaculture marine net pens, and to update the 1990 PEIS with information derived from studies that have been conducted since that time on elements of the environment that are relevant to the analysis of effects that may result from the Cooke species change proposal. The 1990 *Final Programmatic Environmental Impact Statement: Fish Culture in Floating Net-Pens* is adopted herein by reference, in accordance with WAC 197-11-630. This SEPA Additional Information document addresses the potential environmental effects of the species change proposal on the subjects of Escapement, Potential Interactions with Other Salmonid Species in the Natural Environment, Genetic Interactions, and Operational Differences. Potential effects on threatened, endangered, and candidate animal species Federally-listed under the Endangered Species Act since the 1990 PEIS was issued, as well as potential effects on State-listed and candidate species, are provided in SEPA Checklist Attachment D.

A project-specific SEPA Checklist was submitted to WDFW on April 25, 2019, and subsequently revised concurrent with preparation of this Additional Information document to respond to comments and questions identified by WDFW. Because the existing Cooke Aquaculture net pens have previously obtained the facility construction permits as well as Finfish Aquaculture Permits, Clean Water Act Section 402 NPDES Waste Discharge Permits, and Aquatic Land Leases, there is no site selection or construction required to implement the Cooke species change proposal. There will be only minor operational differences to farm triploid Rainbow Trout/steelhead rather than Atlantic Salmon, with insignificant impacts to elements of the Natural Environment. There will be no change in impact to elements of the Built Environment as a result of implementing the species change proposal.

Cooke will use local stocks of Rainbow Trout/steelhead produced by Troutlodge hatcheries in Pierce County. Troutlodge brood stock are cultivated in Washington specifically for the production of ova to supply both private and public aquaculture operations. Troutlodge, a Washington-based company, has been producing Rainbow Trout/steelhead eggs for sale to private fish farms and public enhancement hatcheries since 1945. Brood fish are raised in regulated pathogen-free conditions for their entire life cycle. The company utilizes a comprehensive health testing and disease-free certification program at their Washington facilities which exceed World Organization of Animal Health (OIE) standards, allowing them to export live salmonid eggs throughout the world.

Troutlodge has been producing mono-sex (all-female) populations of Rainbow Trout/steelhead eggs since the mid-1990s. The all-female (XX only) ova are subsequently fertilized with X-only mono-milt. Triploidy is induced by mechanical pressure shock. For a short period of time, a high-pressure hydrostatic shock is applied to the newly fertilized eggs at a specified time point, post-fertilization. The post-fertilization pressure treatment forces the fertilized egg to retain the third set of chromosomes that is normally ejected at this time. Pressure is then released and the triploid (3N) eggs are allowed to continue development. Ploidy is confirmed using a fluorescent nucleic acid label on either embryo or blood tissue using a flow cytometer at the Washington State University School of Veterinary Medicine. Testing results of Troutlodge triploid fish and eggs over a period of five (5) years (from 2013 to 2018) demonstrate a high rate of success in triploid induction (99.83% − 2,950 of the 2,955 fish and/or eggs sampled); see SEPA Checklist Attachment A. Further discussion on the production, testing and physiology of female triploid Rainbow Trout/steelhead is provided in response to Genetic and Interbreeding questions (Section C) below.

Cooke operates two freshwater hatcheries in the Scatter Creek area of Thurston County. The hatcheries raise and produce the juvenile fish that are eventually transferred to the marine net pens for final cultivation to the desired harvest size. Eyed all-female triploid Rainbow Trout/steelhead eggs would be supplied to the Cooke hatcheries from the Troutlodge hatchery under a WDFW Fin Fish Transport permit. The eggs would be hatched and cultured to a certain size in the Cooke hatcheries, and then transferred to the marine net pens after undergoing the necessary fish pathogen screening protocols, subject to review and approval by WDFW. Thereafter, a Fin Fish Transport Permit would be required from WDFW for each specified lot of fish to be transferred from Cooke hatcheries to the marine net pens. Additional information on disease biosecurity practices, disease control and health certifications for Fin Fish Transport Permits is provided in response to Disease Risk questions (Section B, Questions B.7 and B.8) below.

Marine net pen cultivation and production protocols for Rainbow Trout/steelhead will be basically the same as those used for Atlantic Salmon. Maximum cage density levels are expected to be managed at the same levels (approximately 0.9 to 1.2 pound/ft^3 or 15 to 20 kg/m^3), resulting in comparable maximum biomass levels that have historically been attained at each of the existing Cooke Aquaculture sites. Depending upon fish size at harvest (targeted mean weights of approximately 7 to 9 pounds or 3.5 to 4.2 kg); the fish population sizes at each marine net pen site are expected to be similar to stocking levels for Atlantic Salmon. The fish feed composition for marine-reared Rainbow Trout/steelhead diets will be the same or similar to the currently-used marine salmon diets. Modern salmon and trout feeds are composed of highly digestible ingredients that are specifically formulated for optimal growth and feed conversion rates. No differences in water quality or sediment quality are expected to result from this species change or the accompanying Rainbow Trout/steelhead-specific feeds that would be used. Additional information on feed composition, expected feed conversion rates, projected growth rates, projected pen densities and production cycles is provided in response to Operational Differences questions (Section D, Question D.2) below.

Equipment and all net pen support structures, stock nets, and predator exclusion nets will be the same or similar to what has been used for the cultivation and production of Atlantic Salmon at these farms. If approved, the Rainbow Trout/steelhead produced in the marine net pens will be harvested, processed, packaged and shipped fresh to seafood customers throughout the United States. Aquaculture is a form of agriculture, and is designated as a preferred use of the aquatic environment by the State's Shoreline Management Act ([RCW 90.58.020, WAC 173-26-201[2][d] and WAC 173-26-241[3][b][i][A]). Cooke is dedicated to producing high quality seafood in a sustainable and environmentally-sound manner.

PURPOSE OF THE ADDITIONAL INFORMATION DOCUMENT

Additional Information provided in this document supplements the project-specific SEPA Checklist and updates the 1990 Programmatic EIS. If WDFW decides to publish this information as an addendum to the PEIS, certain SEPA procedures will apply. An Addendum is an environmental document used to provide additional information or analysis that does not substantially change the analysis of significant impacts and alternatives in the existing environmental document. … An addendum may be used at any time during the SEPA process (WAC 197-11-706). An addendum shall clearly identify the proposal for which it is written and the environmental document it adds to or modifies. An agency is not required to prepare a draft addendum, or to circulate an addendum for public review and comment, though it is encouraged to do so.[1]

This Additional Information document addresses issues originally evaluated in the *Final Programmatic Environmental Impact Statement: Fish Culture in Floating Net-Pens* (Washington Department of Fisheries, January 1990), as well as responding to comments and questions identified by WDFW during the Cooke Marine Finfish Aquaculture Permit renewal process. The 1990 PEIS provides a detailed analysis of all issues considered relevant to net-pen aquaculture at the time it was published. Additional

[1] WAC 197-11-625(1), (2), and (5).

information provided herein is focused on updating sections of the PEIS applicable to the Cooke Aquaculture species change proposal with more recently available scientific information, and addresses the proposed change of farmed species at the existing marine net pen aquaculture facilities from Atlantic Salmon to an all-female triploid Rainbow Trout/steelhead stock.

SEQUENCE OF EVENTS IN THE MARINE FINFISH AQUACULTURE PERMIT RENEWAL PROCESS

On November 28, 2018, Cooke Aquaculture Pacific (Cooke) submitted application materials to the Washington Department of Fish and Wildlife (WDFW) for the renewal of their existing Marine Finfish Aquaculture Permits for Cooke Puget Sound salmon farms in Washington State. The application materials submitted included information necessary for the renewal of Atlantic Salmon *(Salmo salar)* Marine Aquaculture Permits and for the approval to start farming the all-female triploid Rainbow Trout/steelhead (*Oncorhynchus mykiss)* at the same net pen facilities.

Cooke received an email message from Dr. Kenneth Warheit (WDFW Supervisor, Fish Health Section) on January 2, 2019, requesting additional information on the proposal to begin farming all-female triploid Rainbow Trout (aka steelhead trout) in the marine net pens. Cooke provided additional information to the Department on January 9, 2019, in the form of a Fact Sheet on all-female triploid Rainbow Trout/steelhead production, including stock origin and other information. After submitting the All-Female Triploid Rainbow Trout Fact Sheet, Cooke was advised by WDFW that in order for the Department to process the renewal of the Atlantic Salmon Marine Aquaculture Permit in a timely manner, Cooke would need to revise their original application for rearing both *S. salar* and *O. mykiss* and resubmit it as two separate applications, one for approval of growing *S. salar* and one for growing *O. mykiss* in the marine net pens. Revised applications, one to farm Atlantic Salmon and one to farm all-female triploid Rainbow Trout, were subsequently submitted to WDFW on January 18, 2019.

On March 19, 2019 Cooke received correspondence from WDFW informing the company that the Department had renewed and issued a Marine Aquaculture Permit to raise Atlantic Salmon, but the application for a permit to raise a new species (*O. mykiss*) in the net pens would require the Department to perform an environmental review through the SEPA process. Under the State Environmental Policy Act (SEPA), local governments and State agencies use the SEPA environmental checklist to help determine whether a proposal's impacts are likely to be significant. This helps determine:

- If an applicant can put measures in place to avoid, minimize, or counter adverse effects.
- Whether compensatory mitigation measures can offset significant impacts.
- If an environmental impact statement (EIS) needs to be prepared for further analysis.

On April 25, 2019 Cooke submitted their SEPA Environmental Checklist and preliminary outline of additional support information to Ms. Lisa Wood, WDFW SEPA Lead Coordinator.

On May 21, 2019, Cooke received e-mail correspondence from Dr. Kenneth Warheit and Ms. Lisa Wood, after the Department had reviewed the SEPA documents. Dr. Warheit and Ms. Wood listed comments and additional questions to be addressed by Cooke for the species change proposal in the Additional Information documents. Those May 21 requests have been integrated into the Cooke original outline for the Additional Information and supporting documents to be provided to augment the SEPA Checklist and 1990 PEIS. Responses are provided below on the subjects of Escapement Issues, Potential Interactions with Other Salmonid Species in the Natural Environment, Genetic Interactions, and Operational Differences.

A. ESCAPEMENT ISSUES

Behavior

A.1. What is known about the species-specific behavior of cultured Rainbow Trout; in particular, cultured all-female (AF) triploid Rainbow Trout?

Cooke Aquaculture Pacific Response: The behavior of triploid all-female Rainbow Trout/steelhead raised in captivity differs from that of naturally or enhancement hatchery-produced Rainbow Trout/steelhead because the captive rearing experience alters physical and phenotypic traits. Reinbold, D., G. H. Thorgaard, and P. A. Carter. (2009) found domesticated populations of Rainbow Trout selected for high growth rates and large body size, resulted in reduced sprint swimming performance. They found significantly higher body masses and significantly slower swim speeds in highly domesticated progeny groups compared to semi-wild hybrid progeny groups.

Effects of domestication on the behavioral traits of Rainbow Trout/steelhead in natural conditions has been extensively studied to determine the degree of divergence of hatchery from their wild donor populations. Steelhead bred in captivity quickly (within one to two generations) lose reproductive fitness (Araki et al. 2008). The magnitude of this fitness loss has been estimated as 40% per generation in captivity (Araki et al. 2007). Johnsson and Abrahams (1991) compared the foraging behavior of laboratory-reared juvenile steelhead and steelhead/domesticated Rainbow Trout hybrids. Both strains suffered identical mortality rates and therefore were considered to be equally susceptible to predation. This confirmed that the hybrid trout were significantly more willing to take risks than the wild steelhead. Cultivated stocks raised in aquaculture facilities are protected from the selective predatory pressures that naturally-produced fish would experience almost constantly in the natural environment. As a result, captive stocks have generally been found to exhibit less predator avoidance behavior than wild origin stocks. In a study by Alvarez et al, (2003) on brown trout *(S. trutta)* second generation hatchery fish and the offspring of wild fish raised under hatchery conditions were insensitive to predation risk. This indicates that behavioral divergence between wild and domesticated individuals can arise from a process of direct or indirect selection on reduced responsiveness to predation risk, or as a lack of previous experience with predators (Alvarez et al. 2003).

The farming experience of the captive-reared Rainbow Trout/steelhead is also much different from that of naturally-produced Rainbow Trout/steelhead in that the captive-reared fish experience only pelleted food nourishment during their cultivation and are not reliant on finding natural prey items for their survival and growth. This results in fish that do not readily recognize their normal natural prey items and a reduced ability to effectively capture wild prey items. It is generally thought that farmed fish are maladapted to feeding in the wild (Blanchfield 2009) and the stomach contents of adult Rainbow Trout/steelhead that had escaped from marine farms consisted primarily of indigestible items such as small pieces of wood and other articles that resembled commercial food pellets (Rikardsen and Sandring 2006).

The triploid genetics of the captive all-female Rainbow Trout/steelhead eliminates the reproductive physiology of these fish due to the absence of a functional ovary. Studies of all-female triploid Rainbow Trout/steelhead released into streams showed that female triploids did not exhibit any migratory or reproductive behavior, while some of the male triploid trout would still exhibit reproductive behavior despite being physiologically sterile (Piferrer et al. 2009). Hatchery reared and aquaculture facility reared trout would also have no experience with a natural migration from their natal freshwater rearing location or marine experience other than at the aquaculture location. Thus, the cultivated triploid Rainbow Trout/steelhead would have none of the imprinting, migrating, and prey experience that provides wild and hatchery-released steelhead the experience that supports spawning migrations to their natal streams. Most escaped farmed fish have low fitness for the wild and quickly become easy victims of predators such as marine mammals, other fish, and birds (Amos et al 2002).

A.2 How do cultured Rainbow Trout behave in the wild if they escape from a marine net pen? To what extent do they exhibit "site fidelity" behavior to remain in proximity to the net pen?

Cooke Aquaculture Pacific Response: Rainbow Trout/steelhead reared in net pens have been shown to initially exhibit "site fidelity" that is, to remain near their rearing site when released. After several days, they begin to disperse into the surrounding environment, but they do not have an imprinted natal site, and tend to disperse over a substantial area. However, Bridger et al. (2001) found high site fidelity among triploid steelhead experimentally released during the growing season within a Newfoundland bay. Bridger found 75% of the released triploid steelhead remained within a 500 m radius of the summer grow-out site 32 days after release.

In an experiment to determine the degree to which released domesticated stocks of all-female triploid steelhead exhibited fidelity to an aquaculture facility, Bridger et al. (2001) released all-female triploid steelhead from a cage towed approximately 1 km away from the original rearing site. They observed strong site fidelity among the domesticated steelhead stocks released during the growing season. Fidelity was only slightly larger for the on-site releases (fish released directly from the rearing site) compared to the off-site releases. Off-site released domesticated steelhead made a rapid return to their rearing sites, suggesting imprinting to the rearing site and homing behavior. Released steelhead eventually dispersed from the release site. Bridger found that 65% of AF triploid steelhead released approximately 1,000 m outside of the summer rearing site returned to the site within two days of release. Dispersed triploid steelhead were also detected in the vicinity of other aquaculture sites throughout the Canadian bay. Their observations indicate that the domesticated steelhead will actually return to the rearing site and that some level of orientation exists based on cues or imprinting that was established while growing on the site Bridger et al. (2001).

Westley et al. (2013) found that naturally-produced steelhead stray at rates of 0.30 to 2.3%. They conclude that these patterns are largely the result of species-specific behavioral and endocrine factors during the juvenile life stages, but analyses also suggest that environmental factors can influence straying rates during the adult upstream migration. All-female triploid Rainbow Trout/steelhead reared in net pens do not have the opportunity to encounter behavioral experiences that are gained by wild and hatchery-reared juveniles during their downstream out-migration. They also do not have the capacity to develop the same endocrine factor as hatchery and wild steelhead due to the absence of functional gonads. Lacking both the environmental and hormonal cues, an aquaculture facility all-female triploid Rainbow Trout/steelhead would not be imprinted for a particular natal stream.

Rikardsen and Sandring (2006) found that hatchery Rainbow Trout/steelhead at post-smolt (120 to 340 g) and adult stages (800 to 3,400 g) adapted differently to natural marine prey after escaping from two fish farms in northern Norway. Young domestic Rainbow Trout/steelhead more easily adjusted to natural feeding after escape than the older, larger fish, which often fed on indigestible items similar in shape to the commercial pellets to which they were accustomed. Lindberg et al. (2009) simulated the escape of 48 adult diploid Rainbow Trout/steelhead from a net cage fish farm in Lake Övre Fryken, Sweden. The post release dispersal of Rainbow Trout/steelhead was fast, showed long range dispersal behavior, low winter survival and the fish lacked the ability to find suitable spawning habitats. These results suggested that reproducing for the first time may be an obstacle to the establishment of escaped farmed Rainbow Trout/steelhead.

A.3 In the event of incremental or significant escape, what is the potential for cultured all-female (AF) triploid Rainbow Trout to interact/co-mingle with wild and/or hatchery-reared Rainbow Trout in the natural environment?

Cooke Aquaculture Pacific Response: The effect is determined in part by the age of the triploid Rainbow Trout/steelhead at release and their absence of rearing/migratory experience that both hatchery and wild steelhead encounter during their migration to estuarine and marine habitats (see the response to Question A.1 and 2 above). The Cooke triploid Rainbow Trout/steelhead will be raised from eggs to smolt size on ground water (well water) at the Cooke freshwater hatchery. The smolt will are carried in fish transport trucks from the hatchery and then taken by a fish transport vessel to the marine net pens. The fish will be reared in the marine net pens from smolt to adult size in about 14 to 16 months.[2] During their early rearing freshwater phase, they have no opportunity to imprint on the migratory path to a natal stream.

Selective breeding of salmonids for commercial aquaculture has resulted in domesticated strains possessing a divergent physiological and behavioral phenotype from that of wild conspecifics (Martens et al. 2014). This is particularly true for triploid Rainbow Trout/steelhead that have been intentionally produced to have different physiology and behavioral characteristics than wild steelhead. Martens et al. (2014) concluded that rapid growth of domestic-strain Atlantic Salmon was achieved through the combination of enhanced feed consumption (increased by ~40%) and feeding efficiency (up to 60% improved feed conversion ratio) relative to the conspecific wild fish.

Scott et al. (2014) found that triploid Rainbow Trout/steelhead released into native environments (e.g., protected lakes) had reduced survival and suggested that a lower tolerance to hypoxia in triploids could explain part of the lowered survival. Intentional releases of hatchery-reared diploid Rainbow Trout/steelhead have been shown to result in substantial portions of naturally-spawning fish to be from hatchery-reared parents. Kostow et al. (2003) found that hatchery-produced fish made up 60 to 82% of the natural spawners in the Clackamas River, Oregon. This high percentage of hatchery fish naturally spawning in the wild would not be possible from a net pen release of the all-female triploid Rainbow Trout/steelhead that have a mean sterility rate of 99.8 %.

A.4 What is their chance of survival in the wild?

Cooke Aquaculture Pacific Response: Substantial numbers of Rainbow Trout/steelhead have been reared in hatcheries and released in Washington waters since early in the 20th century. In recent years, roughly 6 million hatchery-produced juvenile steelhead have been released annually by State, Tribal and Federal hatcheries in Washington State waters (Puget Sound, Coastal, and Columbia Basin). Nearly all of these fish are reproductively competent (e.g., fertile) fish that can spawn with either wild or other hatchery-produced steelhead. Many of these hatchery-produced steelhead return to spawn naturally, either with other hatchery-produced fish, or some with wild fish. However, the offspring of hatchery-produced adults have reduced fitness for survival and reproduction in natural environments as compared to wild fish (Chilcote et al. 1986; Kostow et al. 2003; McLean et al. 2008; Araki et al. 2007 and 2008; Blouin et al. 2014; and Berntson et al. 2011). Kostow et al. (2006) found that surviving hatchery females produced only 4.4 to 7.0% the number of offspring produced per wild female.

Araki et al. (2008) concluded that diploid hatchery fish have lower fitness in natural environments than wild fish. This fitness decline can occur very quickly, sometimes following only one or two generations of captive rearing. The all-female Rainbow Trout/steelhead produced by Troutlodge have been reared in hatchery conditions for many years (and multiple generations) with the specific purpose of producing fish that survive and grow rapidly in artificial rearing conditions. The production of triploid all-female Rainbow Trout/steelhead produces fish that grow rapidly, have slower swimming speed, and do not have

[2] The freshwater phase from egg to smolt takes about 10 to 12 months, after which the smolts are reared in saltwater pens for another 14 to 16 months before being harvested. Total time from egg to harvest is approximately 24 to 28 months. These fish have no chance to imprint on a river system since the hatchery runs on well water and they are taken directly from the hatchery to the net pens by transfer vessel.

reproductive capability (average sterility rate of 99.83%). The absence of functional reproductive organs eliminates their capacity to produce the hormones that stimulate spawning migration and behavior (Kobayashi et al. 1998, Piferrer et al. 2009). The life history that involves only pellet food and eliminates exposure to feeding on wild prey renders the triploid Rainbow Trout/steelhead unfit for long-term survival under wild conditions. Escaped fish may survive for months on fat reserves but are likely not capable of out-competing wild or hatchery fish for prey resources, or reproductive habitat.

Chilcote et al. (1986) found that the success of diploid hatchery fish in producing smolt offspring was only 28% of that for wild fish. With the triploid Rainbow Trout/steelhead having a sterility rate of greater than 99.8%, and most or all escaped fish not reaching spawning habitat, only a few escapes would likely produce any smolt offspring.

The proposed aquaculture production of triploid Rainbow Trout/steelhead will use only fish that are produced in a disease-free facility. All-female triploid Rainbow Trout/steelhead are not functionally reproductive in that they do not produce viable gametes. Thus, the all-female triploid steelhead that might be inadvertently released from existing aquaculture facilities have little risk of contributing to the reproducing population of steelhead in Washington waters or diminishing the fitness of offspring that already includes a large component of hatchery-produced fish. If 10,000 triploid all-female triploid steelhead were released and survived to the spawning period, they would potentially contribute 20 fish to a spawning population that already includes several million hatchery-produced fish, if they were released during the period of natural spawning and if they did successfully migrate to a stream in which wild and/or hatchery fish spawn.

A several-year study (Blanchfield 2009) using radio telemetry on open-cage freshwater-raised Rainbow Trout/steelhead intentionally released out of a net pen and into a lake environment showed that the fish suffered high annual mortality after the first year, with none surviving beyond 3 years. The study also found that upon initial release from the net pen, most fish tended to spend a significant amount of time near the cage site. A main finding of this study was the low survival of escaped farmed fish (Blanchfield 2009).

Previous telemetry studies of farmed Rainbow Trout/steelhead and Atlantic Salmon in the marine environment have shown a mix of post-escape movement patterns (Blanchfield 2009). Rainbow Trout/steelhead in coastal Newfoundland exhibited fidelity to the cage sites, movement among the various cages sites within the area, and gradual dispersal (Bridger et al. 2001); while Atlantic Salmon showed rapid dispersal away from the farm sites (Whoriskey et al. 2006). There appears to be some attraction to the cage sites by escaped fish, but a wider geographic dispersion of some of the fish is likely to occur. Tidal currents, site-specific geography, marine mammal and other predator interactions and the initial fitness of the escaped fish to begin with, would all play a part in how widely and quickly escaped fish disperse.

Initial survival outside the protection of the net pen environment is dependent on the level of predation experienced by the escaped stocks. Whole-lake experimental comparisons have shown that domesticated Rainbow Trout/steelhead are more often found in high-risk pelagic habitats and as a result suffer greater mortality compared to their wild counterparts (Biro et al. 2004, 2006). Farmed salmon stocks have not had to utilize predator avoidance traits in order to survive and reproduce. Brood fish spend their entire life in the protected environment of the brood stock hatchery before being artificially propagated to create the next generation of production eggs. With selective pressures removed, there is no competitive advantage for beneficial phenotypic predator avoidance traits to be passed on to the next generation.

In addition, it is generally thought that farmed fish are maladapted to feeding in the wild (Blanchfield 2009). The diets of adult Rainbow Trout/steelhead that had escaped from marine farms primarily consisted of indigestible items such as small pieces of wood and other articles that resembled commercial food pellets (Rikardsen and Sandring 2006). The Spokane Tribal Hatchery and WDFW Sherman Creek Hatchery annually releases approximately 750,000 triploid Rainbow Trout/steelhead into Lake Roosevelt (Peone 2016; WDFW website). In Rufus Woods Lake, where triploid Rainbow Trout/steelhead are raised for aquaculture in net pens, some triploid Rainbow Trout/steelhead are released intentionally and some escape from net pens (Keleher and Cross 2016). Triploid Rainbow Trout/steelhead stomachs (n=409) collected from fish captured in gill nets and creel surveys in Lake Roosevelt and the Rufus Woods area indicated no piscivory with fish primarily consuming *Daphnia*, copepods, ostracods, dipterans, snails, and arthropods (Richards 2011).

Survival and fitness of artificially-propagated salmon and trout in the wild has been studied extensively in the Pacific Northwest. General indications are that salmonids reared in an artificial environment have the potential to rapidly lose fitness characteristics important to their survival in the wild. Accumulating data (Araki et al. 2009) indicate that hatchery fish (Salmonidae) have lower fitness in natural environments than wild fish. This fitness decline can occur very quickly, sometimes following only one or two generations of captive rearing. Although marine aquaculture is a relatively new enterprise, artificial propagation of fish populations has been conducted for centuries, and the major genetic risks associated with salmon hatcheries and marine stock enhancement (loss of fitness, loss of diversity within and among natural populations) have been characterized for several decades (Waples et al. 2010).

Observations and recoveries of escaped pen-reared Atlantic Salmon indicate that escaped fish that manage to survive, do so only temporarily once outside the protection of the net pen environment. Despite being reared in, and having periodically escaped from open net pen aquaculture operations in Washington and British Columbia since the 1980s, escaped Atlantic Salmon have never established a feral population in the Pacific Northwest. Even deliberate releases of Atlantic salmon, eggs, fry and smolts during historical attempts to establish recreational fisheries failed to ever establish self-sustaining populations in the Northern Hemisphere that was outside of their native range, Waknitz et al. (2002). The majority of escaped farmed Atlantic Salmon that have been recovered in Washington and British Columbia are found with empty stomachs and in the state of becoming increasingly emaciated over time outside of the net pen facility (i.e., in the natural environment). Net pen-reared Atlantic Salmon stocks appear to rapidly lose prey recognition and foraging skills, predator avoidance behavior, and reproductive traits. Captively-reared fish stocks become increasingly naïve to the natural selective pressures outside of the artificial rearing environment with each successive generation raised in captivity. Wild fish populations on the other hand, are constantly under the natural selective conditions that encourage adaptive phenotypic and genotypic fitness traits that can be passed on to the next generation.

The Troutlodge stocks of Rainbow Trout/steelhead have been raised in captivity, domesticated and selectively bred for more than 60 years in freshwater raceways and ponds. These stocks are the definition of a domesticated fish stock, having never spawned naturally, fed naturally, or competed in the natural environment for more than 30 generations. Their ability to compete and survive to reproduce in the wild is greatly reduced because of the loss of natural fitness traits resulting from the domestication process. Additionally, triploidy in Rainbow Trout/steelhead is known to reduce the overall tolerance to physiological challenges. Triploid salmonids have been reported to be more sensitive to environmental changes than diploid fish, which may relate to their altered physiology and cellular morphology (Preston 2013). The proposal to use domesticated all-female triploid Rainbow Trout/steelhead is a safeguard that reduces the likelihood of the fish to survive outside of the net pen environment. Using a sterilized, mono-sex stock further reduces the risk of genetic interference from escaped fish that may survive.

With regard to survival and ultimately the genetic risk of escaped fish in the natural environment, Waples et al. (2012) concludes the following; *"These considerations indicate that successful containment of genetic risks associated with marine aquaculture should focus on two general strategies: 1) prevent escapes, and 2) ensure that individuals that do escape have a low probability of surviving to reproduce in the wild. The magnitude of genetic effects of marine aquaculture is determined primarily by how effective* *programs are at these two control points. Regarding the first point, a variety of methods can be used to reduce the probability of escapes, but it is generally recognized that no marine containment system will be 100% effective. The realized genetic effects of marine aquaculture on natural populations, therefore, will depend heavily on the subsequent fate of individuals that escape into natural habitats. According to one view, significant genetic introgression due to escapes from aquaculture is unlikely, because stocks used for aquaculture will either be sterile or so highly domesticated that survival and reproduction in nature is greatly compromised. The extent to which (and time frame over which) this actually occurs, however, is likely to vary considerably among species."* (Waples et al. 2012).

Life Stage and Seasonal Differences

A.5 In the event of an escapement of cultured ALL-FEMALE triploid Rainbow Trout from a marine net pen site, should the amount of recovery effort be influenced by the life stage of the escaped fish in relation to the life stage at the time of wild and hatchery-reared populations in the natural environment?

Cooke Aquaculture Pacific Response: The potential for by-catch of non-target species exists during most fisheries. Fishery managers utilize multiple tools such as spatial-temporal restrictions, gear restrictions, and resource planning to manage the fishery resource and reduce the by-catch of non-target or protected species. Even with perfect execution of these measures by fishery managers and the fishers themselves, unintentional by-catch of non-target species can still occur. The severity of negative consequences from by-catch is dependent on the total amount of by-catch, the survival rates of any released by-catch and the population status of the by-catch species. As with other fisheries where interception of non-target species can occur, gear types and fishing methods play a key role in reducing by-catch impacts.

If a large escapement of all-female triploid Rainbow Trout/steelhead were to occur from a net pen site, fishery managers would need to assess the probability and consequences of ecological impacts of the escaped fish themselves against the potential ecological impacts of those recovery efforts. The same fishery management tools can be used in this instance, and can mitigate the degree of unintended impacts. Similarly, some of the risks of potential negative impacts can be reduced by using a non-reproductively viable (all-female, triploid) stock of fish. This measure reduces the chance of escaped fish from becoming a reproducing feral population and can reduce the probability of genetic introgression on native populations. As discussed later in this document, sterile all-female Rainbow Trout/steelhead stocks have been found to not show signs of sexual maturation or reproductive spawning behavior. It is possible that a small fraction of the farmed population may be diploid females, with the potential to sexually mature and attempt to spawn if they were to survive long enough in the wild. Because the number of those individuals surviving to the point of maturation would likely be low, it can be characterized as a low number of chances for a low probability sequence of events to occur (female diploid > female diploid escapes > female diploid survives to sexual maturation > female diploid locates a spawning stream at the same timing as native fish > female diploid pairs up and successfully mates with a native fish). In making a decision as to whether to recover the escaped fish, the probability of this sequence of events occurring should be weighed against the potential for unintended impacts occurring from the recovery effort itself.

Co-occurrence of similar life stage (adult or juvenile) escaped farmed Rainbow Trout/steelhead with wild or hatchery-stock steelhead, and/or other listed salmonid species, in the marine environment at the same time of year, could occur. As discussed below in response to questions about physiological distinctions and marking techniques, Cooke believes the farm-raised stock of all-female triploid Rainbow Trout/steelhead will have substantial external morphological differences compared to naturally-occurring wild stocks or the enhancement-produced stocks. These physical differences will make an escaped farm raised fish distinguishable if captured in the same environment. Cooke has proposed to externally mark the fish they raise by removal of the adipose fin prior to the fish being transferred to the net pens. This will make the farmed steelhead stocks easily identifiable from wild stocks of salmonids which have intact adipose fins.

Atlantic Salmon that have been raised in Puget Sound net pens over the past 35 years were reproductively viable animals that had the potential to reproduce in the wild and become established. Past large Atlantic Salmon escapement events from net pens in Washington and British Columbia, as well as the intentional planting of millions of Atlantic Salmon eggs, fry and smolt in the past by fishery managers into west coast freshwater systems in their attempts to create sport fisheries, all failed to result in self-sustaining populations.[3] In other words, the probability of successful reproduction was low based on prior knowledge. Despite this fact, the consequences (e.g., the possible establishment of a non-native species) raised the level of concern and effort that was given to remove escaped Atlantic Salmon from the wild by fishery managers. Similarly, the probability of a sterilized mono-sex stock of Rainbow Trout/steelhead establishing a self-sustaining population is also very low, but there is a possibility that some of the escaped Rainbow Trout/steelhead could be diploid females. Based on the efficacy of the triploid process used by Troutlodge and the triploid testing results, the average number of individual diploids in a given population is significantly less than 1% (0.13%); however, some of those fish could survive, mature, find a spawning stream, pair with and then reproduce with native steelhead trout. The likelihood of this sequence of events occurring is low but cannot be dismissed completely. The probability (likelihood) and the consequences (negative impacts) of that type of event must be assessed against the possibility of negative impacts (such as by-catch) occurring from the recapture efforts themselves.

Physiological Distinctions

A.6 Describe potential marking techniques used to specifically identify Cooke Aquaculture fish: fin clipping during vaccination, otolith marking.

Cooke Aquaculture Pacific Response: Cooke proposes to both internally mark the ALL-FEMALE triploid steelhead it raises by thermal marking the otolith at the fry stage and to externally mark the fish by adipose fin removal (clipping) prior to them being stocked into the marine net pens. The adipose fin removal will make the Cooke-reared steelhead visually distinct from any naturally-produced wild steelhead which have an intact adipose fin. The thermal otolith mark will uniquely identify the Cooke Rainbow Trout/steelhead to the hatchery grower (Cooke).

Thermal Otolith Marking

Cooke Aquaculture Pacific uses a thermal otolith marking procedure on 100% of the Atlantic Salmon reared at their hatcheries and marine net pen facilities. Every year-class of Atlantic Salmon fry undergo a brief series of temperature changes that uniquely mark the otolith (ear bones) with a specific series of different sized growth rings. The otolith marks are observable under a microscope. The specific temperature regime for the Cooke hatchery was developed in cooperation with WDFW staff that use the same technique to mark hatchery enhancement fish originating from State and Tribal hatchery programs. Samples of each year-class of fish raised at the Cooke hatchery are sent to WDFW where the otolith marks are both verified and recorded. Washington commercial marine net pen farms growing Atlantic Salmon began the thermal marking program in 2002. It has been a successful technique for identifying an escaped farm- raised Atlantic Salmon to an individual grower. Thermal marking also allows the ability to

[3] WDFW released Atlantic Salmon smolts for the purpose of establishing runs in 1951, 1980, and 1981. Many releases were also made in lakes; however, none of these resulted in the return of adult Atlantic Salmon. For colonization to occur, Atlantic Salmon would need to be successful in each step of a complicated life history, and complete the life history in numbers sufficient to perpetuate the stock. Attempts throughout the United States and world to introduce and establish Atlantic Salmon outside the Atlantic Ocean have failed (Amos and Appleby, WDFW, September 1999).

distinguish offspring if they should arise from a reproductive event in the wild by feral farm fish as those offspring would lack the same thermal markings.

Cooke would use the same thermal marking program for the triploid Rainbow Trout/steelhead stock it is proposing to raise at the company's existing marine net pen facilities. This will individually identify each population of fish being reared as coming from a Cooke Aquaculture farm. The company would again work cooperatively with WDFW staff to develop the unique thermal regime to mark each generation of fry being raised at Cooke hatcheries, destined for transport to the marine net pens. Annual samples would be submitted to WDFW for verification and record keeping.

External Marking by Adipose Clipping

Cooke proposes to externally mark all of the Rainbow Trout/steelhead raised at the hatchery by removing the adipose fins at the time the fish are being vaccinated. The absence of an adipose fin creates a means to identify, by external observation, whether the fish is an escaped farmed Rainbow Trout/steelhead or an unmarked (e.g., adipose fin intact) and presumably wild native steelhead trout during recapture efforts and/or in the midst of any other fisheries that are occurring at the time.

Mass marking by adipose fin clipping is recognized as an effective management tool by State, Federal and Tribal fisheries managers in protecting wild salmon and steelhead and allowing mark-selective fisheries. External morphological differences that are known to occur between farm-raised domesticated stocks of all-female triploid Rainbow Trout/steelhead and presumably wild native steelhead or a fisheries enhancement program steelhead are described below. Genetic records of the Troutlodge stock of Rainbow Trout/steelhead and/or tissue samples for genetic testing would also be provided to WDFW upon request. It is presumed that the DNA analysis would allow the Department to further distinguish between an escaped farmed fish and native, naturally-reproducing fish or fisheries enhancement program fish if necessary.

A.7 Explore a secondary mark, in addition to adipose fin clip, that would distinguish visually Cooke steelhead from tribal and state produced hatchery steelhead.

Cooke Aquaculture Pacific Response: Adipose fin clipping is a method that is used to externally mark enhancement hatchery-produced steelhead in Washington. It should be noted that adipose removal is also used on steelhead released by public enhancement hatcheries in California, Oregon, Alaska and British Columbia. Since both the commercially-reared Cooke all-female triploid steelhead and the enhancement-hatchery-reared Rainbow Trout/steelhead would have the same external identification mark (clipped adipose fin), there is the possibility to incorrectly identify the origins of a steelhead recovered in the natural environment. The chance of encountering an enhancement hatchery steelhead and commercially-reared escaped Rainbow Trout/steelhead in the marine or freshwater environment at the same time and; for both types of fish (farm-reared and wild-reared) to be at the same life stage (age); and of similar approximate sizes and exhibiting the same coloration and other external characteristics such as fin condition, body length, body depth, etc., would seem to be unlikely or of very low probability for the following reasons:

The life histories and selective pressures that would occur between a farm-raised Rainbow Trout/steelhead and wild-reared steelhead are extremely different and likely result in many morphological differences (Taylor 1986; Swain et al. 1991; Fleming et al. 1994; Hard et al. 2000; Belk et al. 2008). The early freshwater life stages between the two types of fish result in substantially different juvenile smolts. For instance, the majority of steelhead juveniles reside in fresh water for two years prior to migrating to sea as smolts. Two-year-old naturally-produced smolts range in size from 140 to 160 mm in length (Wydoski and Whitney 1979, Burgner et al. 1992). Commercially-raised all-female triploid Rainbow Trout/steelhead will reach that same length (140 to 160 mm) in 5 months and can be ready for seawater entry within 10 months of hatching. The expected growth rate of the net pen-reared all-female triploid Rainbow Trout/steelhead forecasts the fish reaching the targeted harvest size of 3.5 to 4.5 kilograms (~7 to 10 pounds) in only 14 to 16 months after entry into the marine net pens (~ 24 to 26 months post-hatch). This means the farmed all-female triploid Rainbow Trout/steelhead will be approximately only 2 to 2.5 years in age by the time they are reaching harvestable size, being harvested (e.g., removed from net pens) and taken to the processing plant. Both wild and hatchery-produced, free ranging steelhead typically spend the first 2 years of their life in the freshwater environment and then another 2 years at sea (2/2) before returning to the river at sexual maturation to spawn (see Figure 1 below). Some steelhead stocks take even longer at sea reaching 5 years of age (2/3) prior to their first spawning. Steelhead can spawn multiple times and have been known to reach up to 45 pounds in weight. Typically however, free-ranging naturally produced adult steelhead will be approximately 10 pounds when they return to the river.

Figure 1.

Table 2c. Age structure of Puget Sound steelhead: frequencies of life-history patterns. Age structure indicates freshwater age/ocean age. Reproduced from Busby et al. (1996). Populations in italics are representative of adjacent ESUs.

		Life History (frequency)				
Population	Run	Primary		Secondary		Reference
Chilliwack River	*WSH*	2/2	0.31	2/3	0.31	Maher and Larkin 1956
Skagit River	WSH	2/2	0.48	2.3	0.33	WDFW 1994b
Deer Creek	SSH	2/1	0.95	3/1	0.05	WDF et al. 1993
Snohomish River	WSH	2/2	0.47	2/3	0.36	WDFW 1994b
Green River	WSH	2/2	0.52	2/3	0.17	Pautzke and Meigs 1941
Puyallup River	WSH	2/2	0.61	2/3	0.28	WDFW 1994b
Nisqually River	WSH	2/2	0.51	2/3	0.28	WDFW 1994b
Hoh River	*WSH*	2/2	0.74	2/3	0.14	Larson and Ward 1952

In theory, an escape of the all-female triploid Rainbow Trout/steelhead could occur at the exact same time and location as the migration of naturally-occurring steelhead in the same area. While there is some chance that the escaped fish could be the same size and age class, it is unlikely they would have the same physical characteristics as wild or enhancement hatchery-produced steelhead. As described above, farm-raised all-female triploid Rainbow Trout/steelhead grow very rapidly in comparison to naturally-produced or enhancement hatchery-produced diploid steelhead. The rapid growth rate of all-female triploid Rainbow Trout/steelhead results in a deeper-bodied fish that will have a much higher condition factor (shorter fork length to weight ratio) compared to naturally-occurring or enhancement-produced steelhead stocks.

Triploid Rainbow Trout/Steelhead

Wild Sea Run Steelhead

In the event of an escapement from a farm site and any subsequent targeted recovery efforts, the farmed fish would be distinguishable by their known age class, average weight, and length and condition factor at the time of the escape. This information coupled with the clipped adipose fin would allow the escaped fish to be identified and separated from by-catch of non-targeted wild and/or tribal and state steelhead stocks. Assuming that the escaped farmed fish survived in the wild for an extended period of time, they have been found to become emaciated because of their limited ability to successfully find wild prey items. This could make them less easily recognizable based on the condition factor alone as the undernourished escaped fish becomes more lean and potentially similar in appearance to a natural free-ranging steelhead. However, there are differences in the physical appearance of a fish that is lean and lengthy from surviving in the natural environment, compared to an emaciated fish which becomes disproportionately long (snake-like) in relation to the size of the head and fins.

In addition, creel counts or steelhead enhancement hatchery brood fish collection facilities would likely be capable of distinguishing the external morphological differences in the fish besides just the lack of an adipose fin. While there is a low probability that an escaped all-female triploid Rainbow Trout/steelhead could be misidentified as a Tribal or State enhancement steelhead, the risk of genetic impact to either wild or enhancement stocks would be further mitigated by rearing only all-female triploid stocks of Rainbow Trout/steelhead at the marine net pens. Opportunities for recreational and commercial fishing for steelhead are very limited in Washington. The risk that one of these fisheries would capture extra enhancement hatchery-reared steelhead because they were misidentifying them as being possible escaped farmed all-female triploid Rainbow Trout/steelhead from a net pen facility would not seem to be very high.

Other possible mass marking methods were looked into for a different technique that could further easily distinguish marine net pen escapees from enhancement stocks. There are multiple methods that have been developed over the years by fisheries scientists and enhancement programs to expand the options for externally marking salmonids. Methods such as freeze branding (FB), Visible Implant Filament (VIF), Visible Implant Elastomer (VIE) tags, tattooing and ventral fin removal are methods that have varying levels of success depending upon the type of application. Literature suggests that external marks such as freeze branding, VIF and VIE are only short-term external marks that eventually disappear over the life span of the fish (Figure 2). Ventral fin removal is another option but is less effective than adipose fin removal because of the capability for ventral fin regeneration (up to 47%) after removal (Skalski et al. 2009). Removal of the adipose fin has the most long-term stability with a very low frequency (0 to 4%) of regeneration (Skalski et al. 2009). Of these, only adipose fin removal appears to be the most suitable for mass marking programs, and for the ability for this external mark to be recognizable for the entire life span of the fish.

Figure 2.

Table 1 Summary of marking techniques for small fish, the availability of unique codes for ease of identifiability, permanency (stability) of the mark, and minimum fish size or life stage requirements

Mark technique	Unique codes possible	Suitable for mass mark	Category of detection*	Stability	Minimum fish size or life stage
External marks					
Fluorescent elastomer (VIE)	240	No	IV or ISD	Variable	26 mm
Fluorescent filament (VIF)	3-character alpha-num	No	IV or ISD	Variable	50 mm
Dye marking	Limited (i.e., ≤30)	Yes	IV or ISD, NS	Variable	25–50 mm
Adipose clip	None	Yes	IV	0–4% regeneration	50 mm
Ventral clip	None	Yes	IV	0–47% regeneration	50 mm
Adipose or ventral clip and CWT (coded wire tag)	Unlimited	Yes	DD	Variable	<2.1 g HLCWT >2.1 g FLCWT
Tattoos	Limited	No	IV, NS	Low	100 mm
Freeze branding	Limited	No	IV, NS	Low	50 mm
Internal marks					
Half-length CWT (HLCWT)	Unlimited	Yes	DD, S	Variable	<2.1 g
Full-length CWT (FLCWT)	Unlimited	Yes	DD	Variable	>2.1 g
Genetic	Unlimited	Yes	DD, NS	Permanent	None
Molecular/laser	Limited	Yes	ISD, NS	30 months	8 days post-yolk absorption
Strontium isotope ratios	None	Yes	DD, NS	Permanent	None
Oxytetracycline	Limited	Yes	DD, S	High	None
Strontium chloride	Limited	Yes	DD, S	High	None
Calcein immersion	Limited	Yes	ISD, S or NS	4–12 months	None
Tetracycline	Limited	Yes	DD, S	High	None
Otolith thermal	Unlimited	Yes	DD, S	Permanent	Emergent fry–advanced yearling
Dry mark otolith (eggs)	Unlimited	Yes	DD, S	Permanent	Only for eggs
PIT	Unlimited	Yes	ISD, NS	85–100%	50 mm

*Detection categories.
IV: Immediate Visual—marks that can be easily and immediately seen by the unaided eye.
ISD: Immediate Specialized Detection—marks that can be immediately detected with the proper equipment. Every fish must be analyzed because these fish do not have a visual identifier.
DD: Delayed Detection—marks that require sacrificing the fish or sampling harvested fish to obtain the tag or tissue for specialized laboratory analysis.
S: Sacrificing the fish is required.
NS: No sacrifice of the fish is required.

A.8 If a recovery effort was implemented to capture escaped cultured all-female triploid Rainbow Trout, what are the observable morphological differences that would distinguish them from wild and/or hatchery-reared populations captured in the natural environment?

Cooke Aquaculture Pacific Response: Initial identification of a potentially escaped all-female triploid Rainbow Trout/steelhead would be made by external observation of the missing adipose fin. Further distinction can then be made by observation of the body shape, general size, fork length and weight of the fish; external markings and coloration; and observation of the condition of the pectoral, dorsal, pelvic and caudal fins. Several external morphological differences are known to occur between farm-raised and wild fish. A range of studies conducted, mainly in temperate countries, has demonstrated the existence of wide morphological differences between wild and farmed fish (Taylor 1986; Swain et al. 1991; Pakkasmaa et al. 1998; Hard et al. 2000; Ojanguren and Brana 2003; Cramon-Taubadel et al. 2005; Berejikian and Tezak 2005; Solem et al. 2006; Belk et al. 2008). The all-female triploid Rainbow Trout/steelhead stocks and the wild or enhancement steelhead stocks found in the natural environment would exhibit distinctive traits based on genotypic and phenotypic differences. In relation to morphology, polymorphisms between alternative environments may occur in the same species, in response to different selective pressures (Robinson and Wilson 1998; Langerhans et al. 2003). The all-female triploid Rainbow Trout/steelhead that Cooke proposes to raise in its existing marine net pens will originate from brood fish that have undergone a selective breeding program over multiple generations, specifically focused on rapid growth and ability to thrive in the hatchery and ultimately in a commercial rearing farm. Wild populations live under the natural processes for survival that continually selects for the ability to successfully forage and compete for prey, avoid predation, thrive and then successfully reproduce in the natural environment. Alternatively, domesticated or farmed stocks of Rainbow Trout/steelhead express traits that are either beneficial or the by-product of being raised in the controlled farm environment, first at the hatchery and then later in net pens, and share few (if any) of the same selective pressures that wild steelhead undergo. Selective pressures present in the fish farming tank are quite different from those existing in the natural environment, which may result in very different morphologies generated by phenotypic plasticity (Taylor 1986; Swain et al. 1991; Fleming et al. 1994; Hard et al. 2000; Belk et al. 2008).

The all-female triploid Rainbow Trout/steelhead proposed to be reared in the Cooke Puget Sound marine net pen facilities would come from fry hatched from incubation trays that are fed a pelletized fish feed from the moment they begin actively feeding. The fry are raised in tanks with controlled flow rates, water temperature, and dissolved oxygen levels. They are protected from predators by the walls of the hatchery building and their rearing tanks, and eventually by the barrier netting around marine net pens. The selective pressures for predator avoidance behavior, physical prowess, body shape or coloration that would increase their chance of survival in the wild are no longer present. Farmed triploid Rainbow Trout/steelhead will exhibit a much higher condition factor (body weight to length ratio) than a wild or hatchery-reared steelhead of the same age. This body shape difference alone would likely make distinguishing a recaptured escaped farm-raised fish from a wild or hatchery origin steelhead captured in the wild, obvious to fisheries managers as well as others. Several studies demonstrate that morphological and physiological differences are observable from the first (F1) generation in a hatchery compared to the same stock of Rainbow Trout/steelhead that reproduced in the wild. Wild juvenile steelhead were found to possess significantly more superficial lateral line neuromasts, normal aragonite-containing otoliths, and significantly larger brains than hatchery-reared steelhead (Brown et al., 2013). Additionally, these differences together predict reduced sensitivity to biologically important hydrodynamic and acoustic signals from natural biotic (predator, prey, conspecific) and abiotic (turbulent flow, current) sources among hatchery-reared steelhead, in turn predicting reduced survival fitness after release (Brown et al. 2013). It should also be pointed out that net pen-reared fish typically show some signs of fin erosion stemming from mechanical damage during their hatchery rearing process and then later the rearing phase in a net pen. This could also be a way for fisheries managers and others to distinguish pen-reared fish from wild or hatchery free-ranging steelhead.

The external appearance of hatchery-raised triploid Rainbow Trout/steelhead (*O. mykiss)* is often compared to a "football" because of their deep body relative to their fork length and the smaller head size compared to diploid Rainbow Trout or sea-run steelhead (*O. mykiss)*. See the photographic comparison in the response to A.7 above. The deep body shape is the result of the rapid growth rates and a tendency for compression of the vertebrae due to the rapid growth rates in comparison to natural or hatchery-origin sea-run steelhead. In the event of an escapement of fish from a marine net pen, information that would be provided by the aquatic farmer to State agencies and Tribes would include the estimated number of fish that had escaped; and the location, age class, average size, health status and medicated feed use history which is required by WAC 220-370-120(b). Cooke proposes that, in the event of an escapement, the Condition Factor (CF) of escaped fish also be provided to State and Tribal fishery managers in the initial fish escape report. The Condition Factor along with the clipped adipose fin would be a way to quickly distinguish between an escaped farmed fish and wild or hatchery-raised steelhead found in the natural environment. Knowledge of the average weight of the escaped fish along with the Condition Factor would 1) help in decisions regarding recapture efforts and coordinating the most effective fishing gear types that could be used, and 2) create a means for the fishers to quickly distinguish an escaped farmed fish from possible by-catch of wild or fisheries enhancement program steelhead stock populations. Photographs would also be provided to the agencies of cohort fish for dissemination to the public that would give further visual characteristics of the escaped fish such as coloration, head shape, fin condition, body shape and scale size.

Lastly, it should be pointed out that all-female triploid Rainbow Trout/steelhead stocks show no signs of ovarian development and maturation compared to normal diploid females (Sumpter et al. 1991; Carrasco et al. 1998). The fact that an escaped fish would be sterile and therefore incapable of reproducing themselves or presenting a risk to the genomics of native stocks, should be considered by fisheries managers in contrast to the possible unintended "take" of protected species and the level and type of escaped fish recovery efforts to be undertaken.

B. Potential Interactions With Other Salmonid Species in the Natural Environment: Both Federally-Listed Threatened and Endangered Salmon Species, and Non-Listed Species

Competition

B.1 In the event of incremental or significant escape of cultured ALL-FEMALE triploid Rainbow Trout into the natural environment, to what extent would they compete with wild, Federally-listed salmonid species for food, space and other habitat requirements?

Cooke Aquaculture Pacific Response: It is unlikely that incremental or significant escape of triploid steelhead would result in detectable competition with wild and the numerous hatchery-reared diploid steelhead. Younger all-female triploid Rainbow Trout/steelhead would have no experience with the wild prey resources consumed by wild and hatchery-reared Rainbow Trout/steelhead. The all-female triploid steelhead would be small in number compared to the approximately six million hatchery diploid fish released into Washington waters each year. The all-female triploid steelhead would tend to be surface feeders due to their feeding experience with pellets rather than natural prey. The small number of net pen escapees that would be sexually functional are unlikely to have many individuals that would successfully find spawning habitat that is currently underutilized. Although all-female triploid steelhead are likely to be of larger size than wild and hatchery steelhead of similar age, they tend to be weaker swimmers and thus not competitive with the wild and hatchery fish.

Predation

B.2 In the event of incremental or significant escape of cultured AF triploid Rainbow Trout into the natural environment, what would be the potential for them to prey upon the young of wild, Federally-listed salmonid species?

Cooke Aquaculture Pacific Response: Studies to date suggest that escaped cultured fish do not easily transition to feeding on native fauna (Abrantes et al. 2011). Cultured Rainbow escapes often feed on pellets, leaves, and woody debris and only approximately 0 to 2% have been observed to have fed on fish. The potential for escaped all-female triploid Rainbow Trout/steelhead to feed on young of the wild, Federally-listed salmonid species is exceedingly low. Escapes would have to occur when listed young of the year were in their vicinity, which is only a fraction of the year. In addition, the relative abundance of large numbers of hatchery releases of non-listed fish would further reduce the likelihood that a federally listed fish would be consumed.

By-Catch

B.3 In the event of incremental or significant escape of cultured all-female triploid Rainbow Trout, what are the potential risks of accidental "take" of wild, Federally-listed salmonid species as by-catch in a recovery effort?

Cooke Aquaculture Pacific Response: Recovery methods could be modified to reduce the likelihood of accidental take of wild, Federally listed salmonid species. For example, if an escape event occurred during juvenile outmigration or adult spawning migration of listed species, then recovery gear could be limited to more selective methods, e.g., hook and line, to reduce by-catch.

Also see the response to Questions A.5, A.6 and A.7, above.

B.4 What mitigation measures and plans can be incorporated to reduce the risk of accidental take of listed salmonid species in escaped fish recovery efforts?

Cooke Aquaculture Pacific Response: The prevention of any escapement from the pens in the first place is the best method of reducing the risk of accidental take of listed salmonid species since no recovery effort would be necessary. After the Cypress Site 2 fish escapement, Cooke worked closely with both WDFW and the Washington Department of Ecology (Ecology) to update and improve the company's fish escape prevention, response and reporting plans. These improvements were developed from the lessons learned in the Cypress incident, and include an improved and expanded emergency communications plan, improved inspection programs, increased reporting responsibilities for Cooke that include a list of individual Tribal resource managers to be contacted in the event of an escapement, Incident Command Training for key Cooke staff members, and an emergency response vessel contact list.

Cooke has also worked with the Department of Natural Resources to develop a net hygiene scoring program that tracks the net cleaning progress from week to week at each of the farm sites and is reported to WDNR. Improved containment technologies reduce escapement risks and carry obvious economic, environmental and sociological incentives for the aquatic farmer.

B.5 Please estimate rate of escapes during stocking and through small tears in net.

Cooke Aquaculture Pacific Response: The rate of escapes during stocking is zero. The risk of this type of event occurring has been reduced substantially by the incorporation of the single-generation stocking program. There are few live fish transport or handling events with this type of production compared to the prior multi-generation production strategy which had fish being pumped from pen to pen, periodically size-graded and transferred between different farm sites. Single-stocking means the young fish are size-graded into groups at the hatchery, vaccinated and then transferred to an individual pen at the marine site where they remain until they are harvested. The employees are trained in Fish Escape Prevention, and the proper techniques for inspecting the stock net for holes, securing the transport vessel and discharge pipe during fish stocking and harvesting events. The transport vessel is securely tied to the side of the net pen facility immediately adjacent to the pen that is going to be stocked or destocked. During stocking, the fish are pumped from the hold of the vessel by a vacuum fish pump. The discharge hose from the pump chamber is mechanically clamped to the fish pump chamber and the hose is supported by the vessels' crane. The outlet end of the discharge hose is placed into the fish pen designated to receive the fish, and is secured in place with mooring lines. The vacuum pump has to be running for fish to be pumped through the pipe and into the pen. The reverse is true when fish are harvested from the pen. They are pulled through the vacuum pump chamber and discharged into a machine that humanely dispatches them. The dispatched harvested fish slide down an enclosed chute and into the hold of the vessel. Employees at the farm site assist and supervise these handling events. For all of these reasons, there is low risk of fish escapes during stocking or destocking of the fish pens.

The estimated rate of escapes through small tears in the net is zero. There is no constant rate of escapement of fish through the netting or a leakage rate as suggested by the B.5 information request. There is, however, always going to be some potential risk of escapements from accidents and damage to the fish containment nets. The job of a fish farmer is to minimize the probability of accidents by using multiple prevention procedures. Risks are identified and plans are developed and implemented to reduce those risks. Employees are trained in the company's Fish Escape Prevention Plans. There are various methods, materials and procedures that are incorporated that reduce the risk of small holes or breaches occurring in the netting that could lead to an escape. Stronger stock netting materials, improved predator barrier nets, surface chafe guards on the stock and predator nets, improved net designs, testing break strength, repair and replacement procedures, routine net inspections by divers during mortality dives, and many other practices are incorporated in the procedures to prevent both large and small fish escapes.

B.6 Describe in detail planned recovery actions in the event of a spill. Steelhead do not school to the extent that salmon school, which may mean recapture efforts may not the same as for Atlantic salmon. The Fish Escape Prevention, Response, and Reporting Plan submitted with your marine finfish aquaculture permit application was designed for Atlantic salmon. What are the risks of negatively affecting federally-listed salmonids during recovery after a spill event?

Cooke Aquaculture Pacific Response: The response to this information request is provided above in Section A and in prior Section B responses. Cooke worked with both WDFW and Ecology to update the Cooke Fish Escape Prevention and Response Plans in October 2018, taking into account lessons learned from the Cypress Site 2 fish escapement and response. Specifically, the plans were updated with improvements to the communication channels and identifying the potential resources that could be used for an escape recovery process. The updated plans have been approved by WDFW and Ecology. These plans, actions, procedures and policies are directly applicable to raising any salmonid species from smolt to adult in the Cooke Aquaculture Puget Sound marine net pens. The basic principles of risk identification of the facilities, procedures and equipment, and the subsequent development and implementation of accident prevention procedures are all very much the same. Copies of these plans were submitted with the original Marine Finfish Aquaculture Permit Application materials. The company will be working with both WDFW and WDOE in reviewing and updating those plans with respect to the new NPDES permit condition requirements that are expected to be issued in July 2019. In addition, any permit conditions specific to raising all-female triploid Rainbow Trout/steelhead that are required by WDFW to issue a new Marine Finfish Aquaculture Permit would also be incorporated into updated versions of the plan.

Briefly, if a fish escapement were to occur, the company must notify WDFW, Ecology, WDNR, NMFS and individual Tribal resource managers and begin implementation of the Cooke Aquaculture − Fish Escape Prevention, Reporting and Response Plans (dated October 12, 2018). Excerpts from the current plan are inserted below, describing fish recovery procedures and actions that would be implemented in the event of an escape.

EMERGENCY PROCEDURES FOR DETERMINING AND REPORTING A SIGNIFICANT FISH RELEASE

Investigation

- Site Managers, Assistant Site Managers and/or the General Manager will investigate to determine whether a Significant Fish Release has occurred based on one or more of the following factors: (1) observations of fish behavior in the net-pen; (2) observations of fish stocks outside the containment net; (3) unexplained decrease in daily feed intake; (4) review of fish inventory information; (5) observations of a large breach in the containment net at the surface or by the dive team below the surface; and (6) an unusually high presence of seal or sea lion activity nearby a fish pen.
- If a pen is suspected of having a breach in net integrity, the Area Manager, Site Manager and/or Assistant Site Manager will deploy the dive team as immediately as possible to inspect the pen for a breach and make any repairs if necessary. If immediate deployment conflicts with the safety of the dive team because of night fall, hazardous weather, tidal or other unusual conditions, the safety of the dive team is given priority. The dive inspection is to be carried out as soon as safer conditions allow.
- If a breach in the containment net is discovered in a pen by the divers or from surface observations that is large enough to have allowed a large number of fish to potentially escape then the pen will be suspected of possibly having had an accidental fish escape. The accidental fish escape reporting, recovery and re-inventory procedures will be initiated by the General Manager, Site Manager and/or the Permit Coordinator.

- If an accidental fish escape is suspected, state agencies are to be notified within 24 hours (see Emergency Reporting and Contact Lists).
- The following sections cover the emergency response and reporting procedures in the case of a significant escapement.

Emergency Reporting Procedures

A positive determination that a significant fish release has occurred will initiate the emergency reporting and recovery procedures. An Accidental Fish Release Report will be submitted to the Washington Departments of Fish and Wildlife and the Washington Department of Ecology within 24 hours in the format below.

Accidental Fish Release Report Information

Fish Release Report Date: ______	Age Class of Fish: _______________
Location of Escape: ___________	Disease History: ________________
Date of Release: _____________	Medicated Feed History: __________
Number of Fish: ______________	Species of Fish: ___________
Avg. Weight of Fish: ___________	Cause of Release: ____________
Employee Name: _____________	Employee Position: ___________

Incident Command Structure (ICS) Training and Cooke Staff Responsibilities

In the event of a significant emergency situation at the fish pens that requires the activation of an Incident Command Structure (ICS), an ICS certificated Cooke employee or representative will actively participate with the ICS. The employee will have technical knowledge of the net pen operations and be a resource to the ICS. The following members of the Cooke Emergency Management Team will have received training in the Incident Command process by January 1, 2019, as described below.

- ICS Training: The Area Managers and Site Managers from each marine farm location, the Business Support Analyst and the Permit Coordinator will complete the online ICS-100 training course and have received the appropriate ICS-100 course completion certificate. The ICS-100 online course is available at:

    https://training.fema.gov/is/courseoverview.aspx?code=IS-100.c

Cooke Communications and Responsibilities During a Significant Emergency Event

General Manager and/or Permit Coordinator will contact the following and provide the preliminary Accidental Fish Release Report information described below:

> **Key Contacts** – WDOE, WDNR, WDFW, NWIFC and nearby tribal Natural Resource Managers
>
> **Information** – Compile and provides the fish inventory information for Emergency Reporting Procedures as described below including: species of fish, age, size, fish health history, date of last medicated feed use, estimated number of escaped fish and other pertinent information.

Contact the Washington Department of Health − if the escaped fish are within the required withdrawal period for recent medicated feed use. Information to the Department of Health will include the type of medication and the date when withdrawal will be met.

General Manager and Site Managers contacts the following:

Emergency response vessels, tug-boats, harvest vessels, and any other support vessels as needed to respond to the situation.

PROCEDURES FOR RECOVERY OF ESCAPED FISH

Recapture Procedures

- The first priority shall be to determine and attempt to correct the cause of the accidental fish release through the repair of the breach.
- In the event of a catastrophic failure of the equipment, securing of the net pen structure or other appropriate immediate response actions may be necessary first in order to stabilize the site before other actions on the fish pen site can be taken. The safety of Cooke employees and contractors takes priority at all times.
- Cooke management maintains an Emergency Work Vessel Contact list. Copies of the contact list are attached to the end of this plan. The vessel contact list will be updated annually along with other parts of this plan.
- Concurrently with the actions to stop and reduce further fish escapements from the facility, the Cooke Emergency Management Team will begin the process of implementing the rapid recovery of escaped farmed fish. This will include utilizing internal resources as well as contacting outside resources to aid in the recapture process.
- If there is reason to suspect an accidental fish escape has occurred (see above section − Emergency Procedures for Determining and Reporting a Significant Fish Release), the Cooke Emergency Management Team will contact the Washington Department of Fish and Wildlife (WDFW) regarding the feasibility and approval of possible fish recovery measures in the area of the escapement.
- Recovery efforts are dependent upon approval from WDFW and written authorization by WDFW must be obtained by the Cooke Emergency Management Team before commencing any recapture efforts.
- Upon receiving authorization from WDFW, the company will commence recovery of escaped fish through one or more of the following actions: (1) use of company skiffs and seine nets; (2) contacting the Northwest Indians Fishery Commission and nearby tribal Natural Resource managers to help facilitate the recapture of escaped fish; (3) contacting and engaging the services of local commercial fishing boat operators to facilitate the recapture escaped fish.

Recapture Gear, Boats and Methods

The company owns and operates several large work vessels with cranes. These vessels have the capacity to pull and refit anchors, transport supplies/equipment to the farm sites, lift stock nets to the surface and swap stock nets with new ones. These vessels can be relocated to any site in an emergency situation. The name of the vessel and their home port location is as follows: F/V Clam Digger −Anacortes, Cypress and Hope Island; F/V Elsie Em and F/V Supplier − Bainbridge Sites; F/V Farm Hand − Port Angeles.

Gear − Each farming area typically has 2 to 3 braided nylon seine nets approximately 15 fathoms long and 9 fathoms deep with 1" mesh. These nets are used for harvesting fish but can be used as beach seine nets in an emergency. Company work skiffs would be used to deploy the beach seines and crew along the nearby shorelines for attempted escaped fish recovery.

Methods − The most effective method to recover escaped fish would be to hire outside contractors with commercial fishing gear and vessels designed for capturing and killing wild and hatchery raised fish. There are multiple factors that would go into the decision process for which type of gear and fishing methods could be used to recapture escaped fish. The time of year, location and the size of the escaped fish will all need to be considered before deploying commercial fishing boats which could result in by-catch of non-target species.

Depending on approval from fisheries resource managers however, the immediate area around the farm could be "fenced off" using large purse seine vessel(s). These vessels could be used to reduce the dispersion of the escaped fish and facilitate recovery. Depending on the situation the nets could be pursed and the fish pumped into a harvest vessel using the vacuum pump. Any non-target salmonids could be removed manually by visually observing the fish on a de-watered table and releasing them over the side. The most effective method for capturing and killing escaped fish would be the use of gill-netters if the fish are within the size range that gill net vessels target. Typically gill-nets are made for harvesting fish that are over 3 pounds and up to 15 pounds. While large fish may get tangled in the netting, the size of the heads precludes them from getting gilled in the net. Smaller escaped fish could easily swim through the mesh openings or not be fully gilled and fall off as the nets are retrieved.

- Cooke will work with nearby area Tribes to annually review appropriate fisheries, gear types and identify the key natural resource contacts in the areas near each of the marine net pen farming locations. The tribal contact list has been developed with the names and department contact phone numbers for the nearby tribal entities.
- Within five working days of terminating the fish escape recovery actions, the company will submit a Fish Recovery Response Report to WDFW and WDOE.

We have found very little literature that discusses any behavioral differences in schooling between farm raised Atlantic Salmon and all-female triploid Rainbow Trout/steelhead raised in marine net pens. Both fish school in the pens however, all-female triploid Rainbow Trout/steelhead reared in net pens seem to be especially surface orientated in the net pen, whereas Atlantic Salmon will generally settle into a schooling pattern mid-pen or deeper once they have finished feeding. Both species of net pen reared fish are fed pelletized feed each day, which is distributed across the surface of the pens during the feeding periods. When being fed, the all-female triploid Rainbow Trout/steelhead crowd the surface of the pen to the point that they are substantially breaking the surface or "boiling" the surface water of the pen while they are feeding (personal observation).

Wild steelhead live a more isolated life in comparison while foraging in the freshwater and little is actually known about the social behavior of sea-run steelhead while they are in the marine phase of their life cycle. Likely some schooling and regrouping behavior is exhibited as the fish return to their natal streams at distinct times of year to reproduce. Personal observations and information gathered during previous large escapements of Atlantic Salmon from marine net pens indicate the fish tend to hug the shoreline and congregate, at least initially, in shallow bays and shoreline environments. It is likely that the fish are conditioned to feel more secure when there is an object in their peripheral vision (such as the shoreline) after they initially escape because that is the environment they have been raised in their entire life. Both farmed species of fish are raised for their entire life beginning in fiberglass tanks and then later in the net pens for the production fish which likely leads to substantially different behavior than their wild counterparts in the natural environment.

Disease Risk

B.7 What would be the potential risk of pathogen and/or parasite transmission from cultured AF triploid Rainbow Trout to wild and/or hatchery reared salmonid species when passing by the net pens, or in the event of an escape?

Cooke Aquaculture Pacific Response: Outbreaks of pathogenic virus or bacteria are rare and would be treated, and/or fish would be removed from net pens. During the time of on-site infection with pathogens or parasites, there would be a chance of transmission from cultured fish to wild and/or hatchery salmonids passing by the net pens. However, the transmission of pathogens or parasites would be reduced due to dilution with greater distance from the net pens. The risk of transmission might be greater with escapes from the standpoint of a greater chance for co-mingling of cultured fish with wild/hatchery fish. On the other hand, dispersal of escaped fish would reduce the local concentration of infective organisms being shed.

The use of triploid Rainbow Trout/steelhead would not increase disease concerns relative to the use of Atlantic Salmon. Triploid and diploid Rainbow Trout do not differ in immune responses to infectious diseases (Weber et al. 2013, Wiens et al. 2018).

Should escaped all-female triploid Rainbow Trout/steelhead carry a disease agent, the risk of them being the source of an outbreak in wild fish is low for the following reasons (Amos, Thomas and Stewart 2001, as cited in Rust et al. November 2014):

1) Native pathogens are already a part of the environment where wild fish are routinely exposed and have developed some natural immunity
2) Escapees are unlikely to generate an infectious dose (or infective pressure) sufficient to result in disease in a healthy wild population
3) The mere presence of a pathogen alone will not cause disease without environmental factors that play a large role in triggering disease events (McVicar 1997; Moffitt et al. 1998; and Amos, Appleby et al. 2001 as cited in Rust et al., November 2014), and
4) Most escaped farmed fish have low fitness for the wild and quickly become easy victims of predators such as marine mammals, other fish, and birds.

B.8 Describe disease control methods: Biosecurity of brood stock facilities, eggs, smolt and marine farm sites; disease screening, reportable and regulated pathogen reporting plans, vaccination, and antibiotic use.

Cooke Aquaculture Pacific Response: WDFW and the Washington Department of Agriculture (WDOA) have regulatory authority with regard to aquaculture products. Chapter 220-370 WAC establishes rules promoting the health, productivity and well-being of aquaculture products and wild-stock fisheries. These rules identify the conditions that are required for the transfer and importation of live aquaculture products in Washington and the circumstances when action will be taken to control disease (WAC 220-370-020).

WDFW regulates the movement of live private-sector aquaculture products through the Fish Transport/Import Permit. Private fish hatcheries are required to maintain fish health laboratory screening records and to routinely screen the stocks for regulated pathogens. Fish Transport permits are required for the movement of fish from the freshwater hatchery to the marine net pen facilities. The brood stock producing the AF triploid Rainbow Trout/steelhead stocks are raised in isolation on regulated pathogen-free water. These brood fish are screened for pathogens when they are spawned, and the resulting gametes are incubated on regulated pathogen free ground water at the hatcheries. Negative disease screening results are necessary in order for a transport permit to be issued by WDFW. The smolts will undergo an additional regulated pathogen screening test just prior to transfer to the marine net pens. A Fish Transport Permit is necessary for the movement of the smolts from the hatchery to the marine sites and again, testing requirements for regulated pathogens must be met prior to the issuance of a transfer permit. Additional disease screening requirements may be added by the Department to ensure the health of aquaculture products and wild stock fisheries. Licensed veterinary services and accredited veterinary labs are used by aquatic farmers to certify that brood stock fish and resulting fry are free of regulated pathogens. The movement of live salmonids or gametes across State or international borders (importation) is strictly controlled by the additional Federal regulations enforced by the U.S. Fish and Wildlife Service (USFWS) under Title 50 of the Code of Federal Regulations (Regulation 50 CFR, Part 16.13). A Title 50 accredited fish health inspector must certify that the live aquaculture products have undergone the necessary disease screenings that meet Title 50. Inspection by and approval from USFWS is necessary for the importation or transport of live salmonid products into Washington State. The fish health regulations are designed to control risk and reduce the incidence of serious fish pathogens from negatively affecting private, public and/or Tribal aquaculture facilities, and to protect the wild stock fisheries.

Managers of aquaculture facilities prevent and control the risk of disease events by implementing structured disease prevention programs. The ability to maintain a captive brood stock program in the controlled hatchery environment using pathogen-free ground water is the foundation for growing disease-free stocks. Strict biosecurity measures including isolation of each generation and their life stages at the egg, fry, and smolt stage further enhance the health of the production fish stocks in the marine net pens. Technological advancements in fish farming equipment, feeds and fish rearing procedures have evolved significantly since the 1990 PEIS was written. Facilities routinely utilize ozone and/or UV sterilization at the hatcheries to ensure pathogen-free water supplies at the hatchery. Vaccination of 100% of the smolts prior to transport to the net pens is a routine practice along with the development of improved vaccines.

Effective disease control programs include: 1) routine health exams by aquatic animal health specialists; 2) health inspections prior to movement of fish between regions or health management zones; 3) accurate record keeping by the farmer to include mortalities and presumptive causes, growth rates, and feed rates, and feed conversion rates; 4) implementation of a bio-security plan for each farm site; and 5) use of preventative medicine such as 100% stock vaccination and the use of probiotics (Rust et al., November 2014).

Washington marine finfish farms employ full-time fish health professionals who routinely monitor the health status at the farm sites. Aquatic farmers have economic incentive to maintain the health of their fish stocks through the use of strict biosecurity measures, disease control practices, 100% vaccination of the juvenile fish prior to transfer to marine net pens, and maintaining growing conditions conducive to healthy and fast-growing fish populations. Cooke Aquaculture will use local stocks of triploid Rainbow Trout/steelhead produced by Troutlodge hatcheries in Pierce County. Troutlodge, a Washington-based company, has been producing Rainbow Trout/steelhead eggs for sale to farms and public enhancement hatcheries throughout the world since 1945. Brood stock are raised in regulated pathogen-free facilities for their entire life cycle. The company utilizes a comprehensive health testing and disease-free certification program that exceeds World Organization of Animal Health (OIE) standards at their Washington facilities, allowing them to export live salmonid eggs throughout the world.

Troutlodge has been producing mono-sex (all-female) populations of Rainbow Trout/steelhead eggs since the mid-1990s. Eyed all-female triploid Rainbow Trout/steelhead eggs delivered to Cooke hatcheries from the Troutlodge hatchery under a WDFW Fin Fish Transport permit would be hatched and cultured in isolation on regulated pathogen-free water to a certain size in the Cooke hatcheries, and then transferred to the marine net pens. Prior to transfer to the net pens, the smolts would undergo the required fish pathogen screening requirements subject to review and approval by WDFW and necessary for issuance of a Fish Transport Permit. The Fish Transport Permit is required from WDFW for each specified lot of fish to be transferred from the Cooke hatchery to the marine net pen farms.

C. GENETICS, INTERBREEDING

C.1 Describe production techniques for all-female and triploid stocks of Rainbow Trout and ploidy quality assurance data.

<u>Cooke Aquaculture Pacific Response</u>: High rates of triploid induction in salmonid species are achieved by thermal shocking and pressure shocking the fertilized eggs at a specific stage during early cellular development. Physiological stress is associated with temperature shocks resulting in lower survival to hatch than pressure shocking. It has become the consensus that hydrostatic pressure shock is the methodology of choice for triploid induction in salmonids due to the relative ease to standardize the shock, as compared to temperature, and to obtain consistent results (Piferrer et al. 2009).

Diploid Rainbow Trout/steelhead ova contain two (2) sets of chromosomes while the milt from diploid male trout contains only one (1) set of chromosomes. Under normal post-fertilization conditions between a diploid male and female, the extra set of female chromosomes contained in the ova are ejected during meiosis and the formation of the second polar body. The resulting zygote contains a set of two chromosomes that will develop into diploid organism. A physical or chemical shock applied during meiosis can suppress the formation of the second polar body and the ejection of the extra set of female chromosomes. The resulting zygote contains (3) sets of chromosomes (3n), which will then develop into a triploid organism. In diploid organisms, the condition of triploidy or polyploidy results in the suppression or complete loss of reproductive traits and viability. Ploidy is confirmed using a fluorescent nucleic acid label on either embryo or blood tissue using a flow cytometer at the Washington State University School of Veterinary Medicine. Testing results of Troutlodge triploid fish and eggs over a period of five (5) years (from 2013 to 2018) demonstrate a high rate of success in triploid induction (99.84% − 2,950 of 2,955 fish and/or eggs sampled; see SEPA Checklist Attachment A.

Triploidy in Rainbow Trout/steelhead and other salmonids has also been achieved by developing a population of tetraploid (4n) males that produce 2n milt and crossing them with diploid (2n) females. Survival rates and growth however, has been shown to be severely depressed in the tetraploid individuals in comparison to the diploid controls, and makes brood stock production difficult (Chourrout et al. 1986). It is the Cooke's understanding that hydrostatic pressure treatment on diploid crosses represents the most consistent and technologically available method for the consistent production of triploid Rainbow Trout/steelhead. In addition, the utilization of feminized brood populations (producing X-only gametes) in combination with induced triploidy further ensures that sexual maturation is significantly inhibited. Female triploid Rainbow Trout/steelhead are functionally sterile with no observable development of oocytes (Lincoln and Scott 1984). Sterilization of populations, even if only partially effective, can substantially reduce both genetic and demographic consequences of cultured escapees (Baskett et al. 2013). Studies carried out in Britain on public and private river stocking programs looked carefully at the use of all-female triploids as a means to protect naturally spawning stocks. Results indicated that the stocking of all-female triploids did not noticeably impact wild stocks, and all-female triploids did not exhibit reproductive migration to spawning grounds (Piferrer et al. 2009).

C.2 For the past two years, based on Appendix A of the preliminary outline, your sample size for testing the efficacy of the triploidy procedure was as low as 60 individuals. What would be the sample size to validate triploidy for the lot of fish being moved into a net pen site (n ~ 800,000 for example)? If the number tested is as low as 60, the testing rate would be 0.008%, which seems inadequate to estimate the true triploidy rate.

<u>Cooke Aquaculture Pacific Response</u>: The results provided in Appendix A are additive, and confirm the hydrostatic shock process used at Troutlodge to induce triploidy. Ploidy was confirmed using a fluorescent nucleic acid label on either embryo or blood tissue using a flow cytometer at the Washington State University School of Veterinary Medicine. Testing results of Troutlodge triploid fish and eggs over a period of five years, from 2013 to 2018, and a total of 2,955 fish and/or eggs being tested, demonstrated a mean success rate of 99.84% triploid induction with low variance. Cooke suggests continuing the procedure of randomly sampling 100 fish from each individual lot of eggs/fish from a cohort that is being reared at the Cooke hatchery. Results of the triploid testing for each lot would be submitted to WDFW along with the fish health screening results as part of the Fish Transport Permit Application for that group of fish. If over a period of time, the results of the triploidy testing continues to show a low level of variance (verifying the triploid induction process), then this testing number could possibly be reduced to 60 fish samples.

In addition to the triploid screening process, Cooke suggests that WDFW personnel could perform visual inspections over a large number of adult fish while they are being processed at the processing plant, observing for both external and internal signs of sexual maturation and ovarian development. This could be a cooperative and informational process between Cooke and WDFW that could help give the public confidence that the technology to produce all-female triploid fish results in fish that are not developing reproductive capacity.

C.3 What is the evidence that triploid female steelhead do not mature, or at least become sexually active?

Cooke Aquaculture Pacific Response: Most species with cells that have a nuclei are diploid (2n), having one set of chromosomes from each parent. A triploid organism contains three sets of chromosomes (3n) instead of the normal number of two. The occurrence of triploidy in normally diploid organism is known to result in organisms that are not reproductively viable because they lack homologous chromosomes, preventing chromosomal pairing and segregation during meiosis. Comparative studies between diploid and triploid Rainbow Trout/steelhead have been carried out demonstrating a lack of sexual maturation occurring in female triploid Rainbow Trout/steelhead. In diploid female controls, the ovaries were packed with oocytes while those from female triploids, although showing the typical lamellar structure of an ovary, contained no oocytes, thus indicating that female triploids are sterile (Lincoln and Scott 1983). Diploid females had large, well developed ovaries containing yolk filled secondary oocytes whereas the triploids had only string like ovaries containing nests of oogonia. No primary oocytes were present (Lincoln and Scott 1984).

Triploid organisms with three sets of homologous chromosomes are found spontaneously in both wild and cultured populations, and can be easily induced in many commercially-relevant species of fish and shellfish (Piferrer 2009). The major consequence of triploidy is gonadal sterility, which is of advantage in the aquaculture of mollusks since it can result in superior growth. In fish, the induction of triploidy is mainly used to avoid problems associated with sexual maturation such as lower growth rates, increased incidence of diseases and deterioration of the organoleptic properties (Piferrer 2009). The induction of triploidy in cultured fish species is a well-known method to prevent the establishment of fertile populations, non-native reproducing populations, and to reduce risks of genetic introgression with naturally-occurring populations. The process is widely used by public fisheries enhancement programs across the United States for creating public fishing opportunities while protecting the genetic integrity of naturally-reproducing fish stocks in lakes and rivers.

The Oregon Department of Fish and Wildlife has incorporated the production of triploid trout for their recreational trout stocking programs in Oregon lakes and river systems. Behavioral studies carried out by the Oregon Hatchery Research Center (OHRC) on the spawning interactions in experimental stream channels between triploid Rainbow Trout/steelhead and control diploid Rainbow Trout/steelhead were completed as part of their review. The ORHC determined that the triploid female trout did not become sexually mature, did not show any indication of spawning activity, and did not interact with control fish. The triploid males in the same study, on the other hand, did show spawning behavior and interaction with the diploid control fish. Some triploid males do not show any indications of sexual maturation, but a significant proportion of triploid males develop sexually dimorphic morphological characters, behave aggressively towards control males, and attempt to spawn with control females (OHRC research summary publication).

C.4 A mean triploidy rate of 99.83% (median = 100%) is excellent, but even with that rate, and with all sites fully operable and stocked (~3.5 M fish), there will be over 6,000 fertile female fish. The lowest triploidy rate in the data you provided was 98.5%, which would generate over 50,000 fertile female fish. If the last escape event (~250,000 fish) occurred with triploid steelhead, the number of fertile female (425 – 3,750, based on the rates discussed above), would have exceeded the number of wild steelhead returning to spawn in many rivers in Puget Sound. You state on page 2 of your preliminary outline that using all-female, triploid fish would "eliminat[e] risks of genetic interference." Based on the above, these methods would reduce, but not eliminate the risk. Please evaluate this risk in a manner that doesn't simply dismiss the risk.

Cooke Aquaculture Pacific Response: Cooke has initiated many new improvements with regard to preventing escapes over the past two years and believes that by changing to cultivating sterilized all-female stocks of Rainbow Trout/steelhead, the company is taking additional steps to reduce the risk of effects related to escaped farmed fish. Answers provided by Cooke in the response to SEPA Checklist questions state, *"All-female triploid Rainbow Trout have been selected to be raised in the existing net pen facilities because the use of mono-sex and sterile stocks of fish is recognized as means to significantly reduce potential genetic interaction with natural populations."* The draft SEPA Checklist has been revised to clarify that Cooke does not believe all risk would be eliminated by the use of all-female triploid Rainbow Trout/steelhead, nor does the company believe that zero risk is possible. There is always some level of uncertainty, especially when dealing with highly variable and complex ecological systems. Assessing risks involves evaluating how likely potential problems are to occur (probability); how severe those potential problems might be (negative consequences); and what measures can be taken to keep the problems from occurring or correct them if they do occur (safeguards/mitigation). Risk management is an essential tool that is used in almost every human activity, from just crossing the road to building a skyscraper. WDFW uses a risk management strategy when they perform disease screening protocols for regulated fish pathogens at State-operated hatcheries. The Department manages fish health risks by screening the returning adult salmon brood stock fish for regulated fish pathogens and other common pathogens; however, there is always some level of risk for a disease occurrence. For this reason, other biosecurity measures are taken to further reduce risks and pathogen contingency plans are developed to address potential negative consequences if they occur.

As stated on page 3 of the SEPA Checklist:

> "Raising native-stock fish species in marine net pens is consistent with the recommendations of the *Final Programmatic Environmental Impact Statement: Fish Culture in Floating Net-Pens* (Washington Department of Fisheries, January 1990). The Preferred Alternative in the Programmatic EIS (PEIS) concludes that *"In areas where WDF determines there is a risk of significant interbreeding (with indigenous species) or establishment of harmful self-sustaining populations, the agency should only approve the farming of sterile* ***or*** *monosexual individuals, or genetically incompatible species."*

Cooke points out in Section A.11 of the SEPA Checklist that the proposal to use an all-female triploid stock of native fish combines both methods identified in 1990 by WDF for reducing risks of genetic introgression on native species;["By farming a monosex (all-female) and sterile (triploid) stock of Rainbow Trout/steelhead, Cooke's proposed change of species incorporates both methods of eliminating risks of genetic interference to indigenous populations or the establishment of self-sustaining runs."] The company's request to raise a sterile and monosex population of fish is consistent with the 1990 PEIS Preferred Alternative. Both of these measures were recognized (by WDF at the time) to reduce risks of negative impacts from interbreeding with indigenous species. Public fisheries managers have been for many years and are currently using these same methods (e.g., the use of all-female triploid trout stocking programs) in public recreational fisheries throughout the world (including in Washington State) as means to eliminate the risks of genetic introgression on naturally-reproducing populations. We have replaced the term "eliminating" with the word "reducing" in the revised SEPA Checklist (July 2019) to clarify that risks will not be completely eliminated by the species change proposal, but will be reduced by implementing this risk management action.

While we agree that 99.83% is not the same as 100%, and leaves the potential for the occurrence of a comparatively small number of diploid females in a given population, there are several factors that would reduce the probability of escaped diploid female fish interbreeding and thereby negatively impacting local stocks. As suggested in the WDFW concern that hypothetically, there could be from 6,000 to 50,000 fertile females in the population of approximately 3.5 million fish stocked in Cooke Puget Sound marine net pens, this is an oversimplification. It is not reflective of the biological reality of the fish or the physical reality of the operations– factors that need to be considered to assess actual potential risks of genetic interference from an escapement. There is low to no risk that either of these calculated numbers (6,000 or 50,000 fertile females escaping, surviving, and interbreeding) would ever occur. First, it is very unlikely that there would be 3.5 million fish (total) in the Cooke Puget Sound marine net pens all at the same time. Production is staggered so that some sites are harvesting fish while others are fallow, or in the process of planting or growing the next crop. This operational procedure also means that the fish at the different farm sites will be of different year classes compared to one another; therefore, there would be no risk that all of them would reach sexual maturity at the same time.

More importantly, there is low to no likelihood even if there were 3.5 million fish (total) in all Cooke Puget Sound marine net pens, that all of them would escape at the same time or over a short time span. The different net pen sites are located in spatially distant areas of Puget Sound (Rich Passage, Hope Island, Strait of Juan de Fuca and northern Puget Sound – see SEPA Checklist Figure 1). This reduces the risk of a single catastrophic event from releasing the entire population of fish from all the net pen facilities at the same time. Additionally, the WDFW calculation does not take into consideration that the farmed fish are contained within two layers of netting (the stock growing net and the predator exclusion net) on five sides of their pen (sides and bottom), and one layer of netting over the surface of the pen (the bird deterrent net). Even in a catastrophic event, the likelihood of fish escaping without incurring physical damage as they egress the pen structure is very low. It is more likely that a large percentage of the fish that would escape in a catastrophic event would become trapped in the netting structures and either be killed outright or suffer injuries to the point that their survival time outside of the pens would be reduced. This was observed during the Cypress Island Site 2 event when the pen structures where significantly distorted by tidal forces and a large number of the fish were found dead in the bottom of pens. Salmonids rely on a skin barrier system composed of mucus cells scattered in the epidermis that secretes products with antimicrobial activities (K. Shepard 1994). Removal of this outer layer of fish skin may increase susceptibility to infections (Y.S. Svendsen 1997). Abrasions to the skin mucus layer and especially the loss of scales can quickly result in bacterial infections in the skin and the formation of ulcerations, a general decline in the physical condition, and an increased mortality rate.

From experience with past escapes of farmed fish, the percentage of fish that survive in the wild only do so temporarily, as they do not thrive outside of the net pen environment. Escaped farmed Atlantic Salmon recovered in the wild typically are found to not have been actively feeding, becoming more emaciated over time. Hatchery enhancement stocks as well as commercially-domesticated stocks have been shown to rapidly lose foraging, predator avoidance, and reproductive traits in comparison to naturally-produced stocks. The WDFW statement that *"there will be over 6,000 fertile female fish"* is an over simplification that does not account for the other physical and biological factors that substantially change this hypothetical scenario. While a small percentage of the farmed population may be fertile, the risk of those fish all escaping, surviving, reaching sexual maturity, and then successfully reproducing would be reduced by the reality factors described in this response. Cooke understands agency concerns regarding both escaped fertile fish as well as sterile fish, which is why the company will continue to improve upon farming practices and containment measures, and invest in new technology and materials that help to minimize the risks of stock loss from their farms.

WDFW posed a hypothetical scenario of escapement similar to the 2017 Cypress Island Site 2 escape, postulating that had *"the last escape event (~250,000 fish) occurred with triploid steelhead, the number of fertile females (425 – 3,750, based on the rates discussed above)"* would have escaped. Although 3,750 may be a large number of fertile fish relative to spawning wild steelhead, the biological reality of this hypothetical scenario needs to be taken into consideration. Not all escaped fertile fish would mature and interbreed with wild steelhead due to a number of factors.

First, the fertile farmed fish would not all mature in captivity by the time of escape. Normal age of Rainbow Trout/steelhead maturation is 4 years, whereas the total culture duration for Cooke operations is 9 months in fresh water and 14 months in seawater. Thus, the age at harvest is approximately 2 years less than the normal maturation age. Even in a worst-case scenario if 50% of the fertile fish were to be mature at the time of escape, this would leave approximately 213 to 1,875 mature escapees.

Second, the fertile escaped fish would need to feed, survive, and mature in the wild. Studies of the transition of escaped farmed salmonids to feeding on native fauna show low rates from 2 to 13% (Morton and Volpe 2002, Abrantes et al. 2011). Feeding is critical for both survival and maturation. Survival of released farmed Rainbow Trout/steelhead in a protected lake was only 40% within one year, even with access to excess food near the net pens (Blanchfield et al. 2009). Thus, the likelihood of escaped fertile female Rainbow Trout/steelhead in Puget Sound transitioning to feeding and surviving to maturation is low (10%). In the hypothetical situation of 425 to 3,750 fertile escapees, the survival rate prediction (40%) would be that approximately 43 to 375 fertile fish might survive to sexual maturity.

Third, if escaped fertile female farmed fish survived and matured, they would need to migrate to a river during the spawning season in order to interbreed with native steelhead. This would require reproductive synchrony with native steelhead as well as precise navigation to a specific river, which is highly unlikely due to diverse genetic backgrounds and the lack of homing imprinting in the farmed Rainbow Trout/steelhead. Steelhead bred in captivity quickly (within one to two generations) lose reproductive fitness (Araki et al. 2008). The magnitude of this fitness loss has been estimated as 40% per generation in captivity (Araki et al. 2007). The Rainbow Trout/steelhead proposed for Cooke Puget Sound marine net pens have been reared in captivity for approximately 30 generations. Even within just two generations of captive rearing, applying the estimated 40% fitness loss per generation results in the fertile Rainbow Trout/steelhead with only approximately 20% of their original reproductive fitness. If this 20% reproductive fitness loss were applied to the likelihood of 43 to 375 fish surviving to successfully mate with wild steelhead, the number of reproductively viable females would be reduced to 9 to 75 fish as a most conservative estimate. If the actual 30 generations of captive rearing and reproductive fitness loss were applied to the likelihood of 43 to 375 fertile fish surviving a marine net pen escapement, the ability of those fish to achieve reproductive success would be extremely low.

C.5 It is important to note that we are also concerned with biological, ecological, and social effects of escaped fish on non-listed but valued species in the Puget Sound area.

Cooke Aquaculture Pacific Response: See the response to information requests in Sections A and B above.

Cooke believes that the application materials previously submitted to WDFW for re-approval of the Marine Finfish Aquaculture Permit; the revised SEPA Checklist information; and the additional information provided in this document and literature review (Attachment C) on the proposed species change to native Rainbow Trout/steelhead, in conjunction with the 1990 *Programmatic EIS on Fish Culture in Floating Net Pens*, previous NMFS risk assessments and literature reviews regarding Puget Sound net pen salmon farming, and the numerous prior Federal ESA reviews and biological opinions on net pen aquaculture projects in Puget Sound, have looked at the potential effects from many perspectives, over many years.

In addition, previous decisions by the Pollution Control Hearings Board on net pen aquaculture in Puget Sound, along with the continued development of new and improved regulations by local, State and Federal agencies all help to minimize and mitigate risks of significant impacts that might otherwise occur as a result of escaped farmed fish in the environment. Attachment C is an updated scientific literature review and bibliography with regard to salmon and Rainbow Trout/steelhead aquaculture, prepared by technical experts as part of this SEPA Additional Information document. Curriculum vitae for the technical experts are provided in Attachment E.

D. OPERATIONAL DIFFERENCES

D.1 Research and industry information regarding no substantive differences in feeding, water quality, benthic impacts, disease and disease control in net pen salmon farming.

Cooke Aquaculture Pacific Response: Commercial salmonid diets for marine-reared Rainbow Trout/steelhead and salmon do not substantially differ. Cooke anticipates utilizing the same or very similar salmonid diets to those currently used to grow Atlantic Salmon in Puget Sound marine net pens. Peak biomass levels will be the same or less than past peak biomass levels for the Atlantic Salmon operations. There will likely be a difference in the time span of peak biomass as the production cycle is expected to be shorter by several months. Current fresh Atlantic Salmon seafood markets are favorable to a certain percentage of larger fish sizes (14 to 16 pounds or larger); therefore, continuing to feed and grow the population to extend the harvesting period can be financially beneficial. On the other hand, the net pen-reared fresh Rainbow Trout/steelhead market is set up primarily around fish of smaller size categories (from approximately 3 to 9 pounds), and there is less market acceptance for farmed Rainbow Trout/steelhead in the 12 pound and up size category. In addition, farmed triploid Rainbow Trout/steelhead are known to have reduced fitness and are less tolerant of physical stressors in comparison to diploid trout. This is due to the tendency of triploid Rainbow Trout/steelhead to have smaller heads relative to their body size, and a subsequent reduction in gill surface area to body mass ratio. Physical exertion from strong currents and hypoxic conditions can cause reduced feeding, loss of growth, and possibly increased mortality. The company anticipates harvesting the fish beginning around the 7 pound size category. By comparison, the company has historically harvested their Atlantic Salmon stocks when the fish reached the 10 to 11 pound average weight category. It is anticipated that the harvesting volumes and frequency would be increased for the triploid Rainbow Trout/steelhead production in comparison to the Atlantic Salmon production based on the smaller targeted average weight and narrower market size acceptance. Based on the tentative growth projections for triploid Rainbow Trout/steelhead, it is likely that the production cycle will be shorter by several months in comparison to raising Atlantic Salmon resulting in the farm sites being fallowed more frequently than past Atlantic Salmon production cycles.

Because there will be no significant differences in feed components, biomass levels, feed volumes or metabolic characteristics between raising Atlantic Salmon or Rainbow Trout/steelhead at the net pen sites, there will likely be little, to no detectable difference in water quality or benthic effects from the change of species. There is industry knowledge about the differences in feeding behavior between the two species as Rainbow Trout/steelhead tend to be more surface-orientated feeders and also do not tend to present as strong of a “stop feeding” signal in general compared to net pen-reared Atlantic Salmon do. However, each group of fish and specific strains within a species can also exhibit different feeding behaviors. The farm feeding technicians need to “learn” the distinct feeding behavior of the Rainbow Trout/steelhead population just like they learned each new generation of Atlantic Salmon which they have raised. There are a numerous variables that can affect the daily feeding behavior of net pen-reared salmon especially when you consider that these farmed fish are being fed for an average of 450 days at the net pen facility before they reach harvest size. Cooke trains and relies on a select number of employees to perform the feeding operation which helps to control and maintain continuity of the process. Those technicians are responsible for detecting the differences in feeding behavior from day to day and for closely watching the feed delivery quantities to prevent over- or under-feeding the pen population. Underwater feed monitoring cameras will be used to observe the feeding behavior of the fish population along with using periodic sample weights, daily expected feed rates, tracking feed quantities, and comparing actual versus expected growth rates to actively monitor the feeding process.

The Hope Island, Clam Bay, Fort Ward and Orchard Rocks sites are all scheduled to receive updated NPDES permits from the Department of Ecology in 2019. The renewed permits call for an increase in benthic monitoring from the current level of once every 2 years to annually. This increased frequency of sediment sampling as well as other water quality monitoring and reporting requirements are all designed to monitor for negative impacts and allow active management to protect the environment.

Cooke does not anticipate any substantial differences in disease control practices or disease control chemicals to be used at the farm sites as a result of the species change proposal. The all-female triploid Rainbow Trout/steelhead stocks will be vaccinated at the hatchery for common bacterial pathogens and IHN virus. The same farm site biosecurity measures will be employed as those currently being used for Atlantic Salmon production.

D.2 Feed types, feed conversion ratios, growth rates, density, biomass, harvest cycles: literature on current net pen salmon and trout farming.

Cooke Aquaculture Pacific Response: Over the past 20 to 30 years, commercial salmon and trout diets have improved substantially. Research into the specific nutritional requirements of salmonids has led to the development of highly digestible nutrients that contribute to rapid and efficient growth and the health of the fish. The fish feed ingredients for marine-reared Rainbow Trout/steelhead diets are the same as that currently used for growing Atlantic Salmon in net pens, formulated for optimal growth and feed conversion rates. The feed conversions for commercially-reared marine net pen Rainbow Trout/steelhead and Atlantic Salmon are similar and dependent on the feed management strategies and techniques used by the individual grower. Significant developments have also been made over the past 10 years in underwater feed and biomass monitoring technologies, such as underwater camera systems, Doppler radar systems, and active sonar capable of real-time biomass growth measurements of the fish. These tools are used by aquatic farmers to monitor feed levels, the growth of the fish stocks, and to calculate the feed conversion rates of the population. As this food production system continues to grow, the aquaculture industry will drive research and development of new and more efficient commercial salmonid diets, feeding techniques, and feed monitoring technologies. Information provided to Cooke regarding marine net pen Rainbow Trout/steelhead production in Chile over the past three years (2016 to 2018) showed a biological feed conversion rate of 1.16, which is similar to some of the best industry standards for marine net pen Atlantic Salmon production (Ron Malnor, Skretting Feeds, personal communication). Cooke would begin by using a commercial salmon diet following the species change to Rainbow Trout/steelhead. Feed trials would likely be carried out using various energy-to-protein ratios to further identify the best site-specific formulations for growth, health, and flesh quality.

Commercial production of Rainbow Trout/steelhead has grown exponentially since the 1950s, especially in Europe and more recently in Chile. This is primarily due to increased inland production in countries such as France, Italy, Denmark, Germany and Spain to supply their domestic markets, and mariculture in cages in Norway and Chile for the export market. Chile is currently the largest producer. Other major producing countries include Norway, France, Italy, Spain, Denmark, USA, Germany, Iran and the UK.

(FAO 2019, Fisheries and Aquaculture). Feeds for Rainbow Trout/steelhead have been modified over the years, and cooking-extrusion processing of foods now provides compact nutritious pelleted diets for all life stages. Pellets made in this way absorb high amounts of added fish oil and permit the production of high-energy feeds, with more than 16% fat. Dietary protein levels in feeds have increased from 35 to 45%, and dietary fat levels now exceed 22% in high-energy feeds. Feed formulations for Rainbow Trout/steelhead use fish meal, fish oil, grains and other ingredients, but the amount of fish meal has been reduced to less than 50% in recent years by using alternative protein sources such as soybean meal. These high-energy diets are efficiently converted by Rainbow Trout/steelhead, often at food conversion ratios close to 1:1 (FAO 2018, Fisheries and Aquaculture).

The planting and production of Cooke all-female triploid Rainbow Trout/steelhead would follow a similar pattern used for the year-around production of Atlantic Salmon at Cooke existing Puget Sound marine net pen facilities. This production cycle utilized two temporally separated saltwater entry groups that originate from a single year-class of brood fish. The groups are typically referred to as spring-entry and fall-entry smolts. Egg production from a single-year class of brood stock can be extended by photo adjustment of the adult fish at the hatchery. Fertilized eggs that are produced from the spring spawn and the fall spawn cohort are transferred into the smolt production hatchery at different time periods. This produces a fall saltwater-ready entry group of fish originating from the spring eggs, and spring saltwater-entry group that comes from the fall-spawned eggs. The seawater-ready all-female triploid Rainbow Trout/steelhead smolts are produced in approximately 9 to 11 months post-hatching. The estimated size of the fish at saltwater entry would be from 100 to 180 gram average weights. The projected marine growth phase from the saltwater-entry smolt to harvestable-sized fish would be approximately 12 to 18 months. Projected average weight at harvest would be from 7 to 10 pounds. Harvesting the fish population typically takes from 2 to 5 months depending upon the number of fish at a site, market conditions, and production plans. By transferring the juvenile fish (smolt) from the hatchery to the marine environment at different times of the year (spring-entry and fall-entry groups), the production of harvestable-sized fish can be spread out during the entire year.

The Fort Ward, Orchard Rocks, Clam Bay and Hope Island net pen sites all have current leases with the Washington Department of Natural Resources. Cooke is requesting approval of a new Marine Finfish Aquaculture Permit for these facilities to raise an all-female triploid Rainbow Trout/steelhead stock, so that the company can begin raising a native stock of fish and continue to produce healthy seafood, local jobs, and economic activity in Washington.

The Cypress Island Site 1 and Site 3, and Port Angeles Harbor sites are also included in the request for approval of a change of species to the all-female triploid stock of Rainbow Trout/steelhead because Cooke is hopeful that the legal issues around the WDNR leases can be resolved. At this time however, there are no plans to plant fish at the Port Angeles or Cypress Island sites until there is further certainty in their legal status. Finally, the Cypress Island Site 2 facility has been completely removed from the water and Cooke has formally asked Ecology to withdraw the Cypress Site 2 NPDES permit. Cooke is therefore not requesting a new Marine Finfish Aquaculture Permit for Cypress Island Site 2 because this site will not be rebuilt.

The list below identifies the existing Cooke marine net pen sites that could eventually transition to raising all-female triploid Rainbow Trout/steelhead, pending agency approvals, and the estimated possible number of fish that could be planted at each site for each generation/production cycle. Planting and the subsequent harvesting of the fish populations would occur at different times of the year at the different sites. The anticipated growth cycle in the marine net pens is approximately 14 to 16 months between first fish in (planted) and last fish out (harvested). Because of this, the various individual sites and/or growing areas may alternate between growing a spring-entry group for one generation, and subsequently be planted with a fall/winter generation.

Site Name	Approximate Number of Fish	Notes
Hope Island	350,000	
Clam Bay	1,000,000	
Orchard Rocks	600,000	
Fort Ward	300,000	
Cypress Site 1	300,000	WDNR lease pending legal status
Cypress Site 3	400,000	WDNR lease pending legal status
Port Angeles	600,000	WDNR lease pending legal status
Cypress Site 2	0	Pens are removed and will not be replaced
Total:	3,550,000*	*Planting cohorts occurs at different sites and times of year. Total fish number would vary depending on the production cycle.

No differences in water quality or sediment quality are expected to result from this change in species or the accompanying Rainbow Trout/steelhead-specific feeds that would be used.

Marine net pen cultivation and production protocols for Rainbow Trout/steelhead are basically the same as those used for Atlantic Salmon. Maximum cage density levels are expected to be managed at the same levels (approximately 0.9 to 1.2 lb./ft^3 or 15 to 20 kg/m^3), resulting in comparable maximum biomass levels that have historically been attained at each of the existing Cooke Aquaculture sites. Depending upon fish size at harvest (targeted mean weights of approximately 7 to 9 lbs. or 3.5 to 4.2 kg), the fish population sizes at each marine net pen site are expected to be similar to stocking levels for Atlantic Salmon.

D.3 What are the differences in feed conversion between steelhead and Atlantic salmon?

Cooke Aquaculture Pacific Response: See the response to Questions D.1 and D.2, above.

D.4 Species differences in net pen aquaculture (comparison between cultured diploid Atlantic Salmon and triploid Rainbow Trout).

Cooke Aquaculture Pacific Response: Within the same or similar rearing environment (mid-latitude temperate marine waters), the commercial culturing of salmonid species such as Atlantic Salmon and Rainbow Trout/steelhead do not differ substantially. Both fish have similar biologic and metabolic requirements as well as temperature and oxygen demands. Atlantic Salmon and Rainbow Trout/steelhead also share very similar life histories. Commercially-domesticated stocks of Atlantic Salmon typically reach sexual maturity at 4 years of age or more; however, a certain percentage (typically males) will mature at 3 years of age. Rainbow Trout/steelhead typically take from 4 to 5 years to reach maturation. Unlike most other Pacific salmon species that die after spawning, both Rainbow Trout/steelhead and Atlantic Salmon are capable of reproducing more than once in their life time, although rates of survival after spawning are typically very low.

Cooke anticipates there will be some production planning differences between culturing Atlantic Salmon and the all-female triploid Rainbow Trout/steelhead. In general, the U.S. fresh farmed Atlantic Salmon seafood market is set up around the harvested fish product being in the 10 to 14 pound dressed “head on, gutted” (HOG) range. Atlantic Salmon farms set their production plans to achieve these targeted average weights within a projected time frame, knowing that the farm will need to be harvested out (e.g., emptied of one generation) within a certain time frame that will allow a fallow period before stocking the farm with the next generation of juvenile fish. In general, growth of Atlantic Salmon to these size ranges can be achieved in the marine net pens in a 14 to 20-month time period.

The commercial market for net pen-grown Rainbow Trout/steelhead in the United States is based on smaller average weights, typically a product in the 7 to 10-pound range. Industry knowledge of the growth rates for domesticated all-female triploid Rainbow Trout/steelhead in net pens indicate the target sizes can be achieved in approximately 14 to 16 months depending mainly upon original stocking size and site-specific growing conditions. This would result in a shorter production cycle and an increased frequency of fallow periods over a given amount of time compared to Atlantic Salmon culture.

Cooke does not anticipate major differences in stocking densities, daily feed requirements, or disease susceptibility compared to current planning for Atlantic Salmon production. Up to the time of this writing, all Atlantic Salmon stocked into Cooke Puget sound marine net pens received an intraperitoneal injection of an inactivated, multivalent vaccine against furunculosis, classical vibriosis, cold water vibriosis, and infectious hematopoietic necrosis. Cooke anticipates the continued use of this vaccine in the production of the all-female triploid Rainbow Trout/steelhead. Biosecurity methods and disease control practices would remain the same, because disease vectors and potential pathogens are similar if not identical for both types of salmonids.

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[4] These documents were cited in *Common Questions about Atlantic Salmon Net Pen Aquaculture* (January 2016) prepared by Cooke Aquaculture Pacific and submitted with the application for the Port Angeles-East Marine Net Pen Relocation project. Technical reports in this list of Additional Resource Material also partially informed the Cooke Aquaculture Pacific responses to WDFW questions and information requests related to the species conversion proposal to transition existing Puget Sound marine net pens from Atlantic Salmon farming to all-female triploid Rainbow Trout/steelhead.

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Attachment C

Annotated Bibliography

Attachment C to SEPA Checklist
Rainbow Trout Net-Pen Aquaculture: Annotated Bibliography

Prepared by

Walton Dickhoff, Ph.D.[1] and Don Weitkamp, Ph.D.[2]

Cooke Aquaculture Pacific Marine Aquaculture Permit Application to Transition from Raising Atlantic Salmon to Raising Sterile All-Female Triploid Rainbow Trout/Steelhead at the Cooke Existing Marine Net Pen Sites in Puget Sound, Washington

INTRODUCTION

This document provides summaries in abstract format for technical publications that address one or more basic issues of concern identified by the Washington Department of Fish & Wildlife for their review of the Cooke Aquaculture application for re-approval of their Marine Finfish Aquaculture Permit (WAC 220-370-100) to convert Atlantic Salmon farms to domesticated stocks of a mono-sex, sterile Rainbow Trout (*Oncorhynchus mykiss*). This review focuses on publications produced since 2000 due to the substantial changes that have occurred during the last two decades in net-pen aquaculture techniques and practices. Information derived from these technical publications was used to inform the Cooke Aquaculture responses to WDFW questions and information requests in SEPA Checklist Attachment B.

Each publication is presented in a reference/abstract format listed in alphabetical order according to the senior author's last name. The abstracts are grouped according to the basic net pen rearing issues that the papers address. Several abstracts are listed under more than one issue because of the multiple issues they address. The abstracts generally are those provided by the authors with minor alterations. A few are brief summaries of lengthy summaries of unpublished reports.

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BASIC NET-PEN ISSUES

Baskett, M.L., S.C. Burgess, and R.S. Waples. 2013. Assessing strategies to minimize unintended fitness consequences of aquaculture on wild populations. Evolutionary Applications 6(7):1090-1108.

Artificial propagation programs focused on production, such as commercial aquaculture or forestry, entail strong domestication selection. Spillover from such programs can cause unintended fitness and demographic consequences for wild conspecifics. The range of possible management practices to minimize such consequences vary in their control of genetic and demographic processes. The authors use a model of coupled genetic and demographic dynamics to evaluate alternative management approaches to minimizing unintended consequences of aquaculture escapees. They found if strong natural selection occurs between escape and reproduction, an extremely maladapted (i.e., nonlocal-origin, highly domesticated) stock could have fitness consequences analogous to a weakly diverged cultured stock; otherwise, wild population fitness declines with increasing maladaptation in the cultured stock. Reducing escapees through low-level leakage is more effective than reducing an analogous number of escapees from large, rare pulses. This result arises because low-level leakage leads to the continual lowering of wild population fitness and subsequent increased proportional contribution of maladapted cultured escapees to the total population. Increased sterilization efficacy can cause rapid, nonlinear reductions in unintended fitness consequences. Finally, sensitivity to the stage of escape indicates a need for improved monitoring data on how the number of escapees varies across life cycle stages.

Colt, J., S. Summerfelt, T. Pfeiffer, S. Fivelstad, and M. Rust. 2008. Energy and resource consumption of land-based Atlantic Salmon smolt hatcheries in the Pacific Northwest (USA). Aquaculture 280 (1-4):94-108.

The authors evaluate the resource and energy requirements of six different types of land-based, hatchery production systems located in the U.S. Pacific Northwest: flow-through with a gravity water supply, flow-through with a pumped water supply, flow-through with pure oxygen, partial reuse system, partial reuse with heating, and a reuse system for the production of Atlantic Salmon (*Salmo salar*) smolts. Key parameters used in the evaluation include direct energy, indirect energy, transportation energy, greenhouse gas emissions, and pollutant discharges. Power (electricity and natural gas) and feed energy accounted for the majority of the required energy for all the rearing option evaluated. The sum of the fixed capital and chemicals components accounted for less than 2-12% of the total energy budget for any rearing option. The energy efficiency (energy output/energy input) of the six options ranges from 0.97% for flow-through with pumped supply to 3.49% for the flow-through with gravity supply. The rearing options with the three highest energy efficiencies were flow-through with gravity supply (3.49%), partial reuse (2.75%), and reuse (2.64%). On a kg of smolt produced basis, the six rearing options showed a wide range in performance. The reuse system had the lowest water (2 m^3 /kg) and land (0.13 m^2/ kg) requirements and the third lowest total energy requirement (288 MJ kg^{-1}). The partial reuse system had the second lowest total power requirement (276 MJ /kg), a low land requirement (0.21 m^2/kg), and moderate water requirements (33 m^3/kg). The partial reuse with temperature control had the second highest total power requirement (657 MJ /kg) and land and water requirements similar to the partial reuse system without temperature control. The flow-through system with pumped water supply had the highest water (289 m^3 kg^{-1}), land (2.19 m^2/kg), and energy requirements (786 MJ /kg) of any of the rearing options. By comparison, the flow-through system with gravity water supply had the lowest energy requirement (218 MJ/kg), a moderate land requirement (0.78 m^2/ kg), and a high-water requirement (214 m^3/kg). The ranking of the six rearing options based capital and operating costs are likely to be quite different from those based on energy, water, and greenhouse gas emissions.

Costa-Pierce, B.A. and C.J. Bridger. 2002. The role of marine aquaculture facilities as habitats and ecosystems. Responsible Marine Aquaculture 2002:105-144.

Too often the public is provided with a bleak image of marine aquaculture facilities as industrial waste areas, depleting the natural environment and its biodiversity, and creating a desert from an ocean oasis. However, this image frequently has little rigorous scientific basis. Environmental problems have been found only in aquaculture settings with poor management plans, wasteful feeding strategies and where overproduction exceeds the carrying capacity of the natural environment causing the degradation of clean water – the very basis of a successful aquaculture venture. Cage aquaculture facilities provide habitats and nursery areas for juvenile and adult wild fish, and numerous invertebrate and algal species essential to sustaining healthy marine ecosystems and wild fish stocks. There is an unbalanced focus on marine animal husbandry causing a concomitant lack of appreciation for the positive environmental attributes of marine agronomy, a vital economic sector of global aquaculture. Indeed, tidal wetland, mangrove forest and seagrass restoration aquaculture – in addition to establishment and maintenance of oyster reefs - are important examples of aquaculture creating, enhancing and maintaining productive marine ecosystems, habitats and water quality in a long-term, sustainable manner. There is an urgent need for additional research to generate *primary* data on the positive and negative roles of marine aquaculture in the biogeochemical cycles, habitats and ecosystems of coastal oceans worldwide. The little research that has been done to date has documented numerous examples of marine aquaculture facilities that revitalize natural habitats, ecosystems and marine fisheries, as opposed to degrading the natural environment and competing with the wild fisheries sector. Without more comprehensive assessments and additional research, plans for the sustainable expansion of marine aquaculture will suffer from a lack of a scientific basis for rational planning and policy, and continue to be replaced by heresy, junk science and advocacy.

Felsing, M., B. Glencros, and T. Telfer. 2005. Preliminary studies on the effects of exclusion of wild fauna from aquaculture cages in a shallow marine environment. Aquaculture. 243:159–174.

Previous investigations into the environmental impacts at a shallow-water oligotrophic marine experimental cage aquaculture site in Western Australia have found no accumulation of organic material, and limited changes in macrofaunal communities. It was hypothesized that wild fish populations in the area consumed particulate wastes emanating from the cage, thus reducing the benthic impacts. An experiment was designed to quantify the accumulation of organic material on the seabed occurring in the presence and absence of wild fauna. Three treatments were arranged in duplicate, cages without exclusion nets (normal situation) (CAGE-FISH); cages surrounded by a 35-mm mesh exclusion net (preventing wild fish access to the sea bed and water column near the cage) (CAGE-FISH-EXCL); and empty cages surrounded by exclusion nets (to control for effects from the exclusion net) (CAGE-EXCL). In addition, four reference sites without cages (REF) were sampled. Following baseline sampling, Rainbow Trout (Oncorhynchus mykiss) were stocked into the CAGE-FISH and the CAGE-FISH-EXCL treatments at an initial stocking density of 2.4 kg/m. The experiment was terminated after 62 days, at a final stock density of 5.6 kg/m. Sampling found significantly greater accumulation of nutrients and fine sediments under the cages enclosed in the exclusion net than in other treatments and sites. Levels of organic carbon deposition at cages with exclusion nets was found to be 4.5±1.0 g C/m /day (mean ± S.E.) compared to 0.7 to 1.1 g C//m/day at control and reference sites. The accumulation of nutrients at the CAGE-FISH-EXCL sites was correlated to distinct changes in macrofaunal community composition, with a sharp increase in overall macrofaunal abundance and a growing dominance of capitellid polychaetes. Based on a comparison between sedimentation rates within and outside excluded areas, the proportions of the total sediment nutrients consumed by wild fish were calculated to be 40 to 60%. It was concluded that in the natural coastal system of Western Australia or comparable environments, wild fish are potential important consumers of cage aquaculture waste materials. The fact that sediment C, N and P did not increase below cages with fish and no exclusion nets suggests that the benthic fauna, including surface grazing fish, at these sites were able to assimilate much of the remaining total sedimentary nutrients.

Goddard, S. 2012. Feed management in intensive aquaculture. Springer Science & Business Media.

This book has been written as a guide to the management and use of formulated feeds in intensive fish and shrimp culture. While its focus is on the use of commercially produced feeds in intensive production systems, it is anticipated that many of the practical issues covered formulated feeds in less intensive systems. Feeds and feeding are the major variable operating costs in intensive aquaculture and the book is primarily intended to aid decision making by fish farm managers in areas of feeding policy.

The dramatic increases in aquaculture production seen over the past 15 years have been made possible, in large part, by gains in our understanding of the food and feeding requirements of key fish and shrimp species. A global aquaculture feeds industry has developed and a wide range of specialist feeds is now sold. The new options in feeds and feeding systems, which are becoming available, necessitate continual review by farmers of their feeding policies, where choices must be made as to appropriate feed types and feeding methods. While growth rates and feed conversion values are the prime factors of interest to farmers, other important issues, such as product quality and environmental impacts of farm effluents, are also directly related to feed management practices. While there is an extensive literature dealing with the known nutritional requirements of farmed fish and shrimp, less attention has been focused on feed management practices. However, as the various sectors of the industry have matured, and markets have become more competitive, it has become evident that farmers seeking to control their production costs, in order to remain competitive, must critically examine the selection, use, and performance of aquaculture feeds.

While this book emphasizes the practical issues of feed management, it also seeks to provide insight into the biological and environmental factors that underlie the feeding responses of fish and shrimp, and which must be taken into account when determining appropriate feeding policies for specific farming operations. No attempt has been made here to describe in detail the feeding regimens used for any particular species. The examples used throughout the text have been chosen from many different sectors of the industry in an attempt to illustrate the general principles of feed management.

Hardy, R.W. 2010. Utilization of plant proteins in fish diets: effects of global demand and supplies of fishmeal. Aquaculture Research. 41(5):770-6.

Aquafeed ingredients are global commodities used in livestock, poultry and companion animal feeds. Cost and availability are dictated less by demand from the aquafeed sector than by demand from other animal feed sectors and global production of grains and oilseeds. The exceptions are fishmeal and fish oil; use patterns have shifted over the past two decades resulting in nearly exclusive use of these products in aquafeeds. Supplies of fishmeal and oil are finite, making it necessary for the aquafeed sector to seek alternative ingredients from plant sources whose global production is sufficient to supply the needs of aquafeeds for the foreseeable future. Significant progress has been made over the past decade in reducing levels of fishmeal in commercial feeds for farmed fish. Despite these advances, the quantity of fishmeal used by the aquafeed sector has increased as aquaculture production has expanded. Thus, further reduction in percentages of fishmeal in aquafeeds will be necessary. For some species of farmed fish, continued reduction in fishmeal and fish oil levels is likely; complete replacement of fishmeal has been achieved in research studies. However, complete replacement of fishmeal in feeds for marine species is more difficult and will require further research efforts to attain.

Harvey, A.C. 2016. Investigating fitness consequences of hybridisation between farmed and wild Atlantic salmon. Dissertation, Prifysgol Bangor University.

Farmed fish display genetic differences from wild fish in a variety of morphological, behavioral and physiological traits as a result of the domestication process and selective breeding. Farmed salmon typically outgrow wild salmon by large ratios under hatchery conditions, although observed growth differences are much less in the wild. It is possible that farmed salmon have become adapted to regulated domestic environments, while concurrently they are unable to perform as well in more variable wild environments. Escaped farmed salmon interact with wild salmon through resource competition and disease transmission, and can interbreed with wild salmon. The introduction of mal-adapted domestic genotypes into wild populations can lower their productivity. Comparative studies that assess the effects of hybridization on life-history traits linked to fitness are important in understanding how interbreeding will affect the resilience of wild populations. This thesis investigated the freshwater growth and survival of multiple families derived from various farmed, wild and F1 hybrid salmon populations when reared at contrasting 1) temperatures, 2) densities and rearing conditions, 3) food availabilities, and 4) diets. In all experiments farmed salmon outgrew wild and hybrid salmon, and their hybrids displayed intermediate growth. Relative growth differences detected at contrasting temperatures were population-specific; indicating that the competitive balance between conspecifics may depend upon genetic background and river temperature. Findings highlight the merits of adopting a more spatially resolved approach to risk management of wild populations. In all other experiments the relative growth differences among groups did not differ across treatments, indicating that farmed fish have retained their plasticity in response to respective experimental treatments. Although experiments were conducted under controlled conditions, findings suggest that the investigated treatments are not individually responsible for elevated growth differences observed in hatchery conditions or the lower growth differences observed between farmed and wild salmon in the wild.

Liu, S., Y. Palti, K.E. Martin, J.E. Parsons, and C.E. Rexroad III. 2017. Assessment of genetic differentiation and genetic assignment of commercial Rainbow Trout strains using a SNP panel. Aquaculture 468:120-125.

Rainbow Trout (*Oncorhynchus mykiss*) is the most widely cultured cold freshwater fish in the world. Troutlodge, Inc., one of the largest commercial rainbow trout egg producers in the world, has eight breeding populations of rainbow trout. Assessment of population genetic differentiation is critical for selective breeding, and genetic assignment is often required to address production issues arising on fish farms. Previously, they developed a SNP (single nucleotide polymorphism) panel for parentage assignment in rainbow trout. The objectives of this study were: 1) to characterize the genetic differentiation of the eight Troutlodge breeding populations; and 2) to evaluate the accuracy of genetic assignment to identify Troutlodge fish using the same SNP panel previously developed for parentage assignment. A total of 1,732 breeders of the eight Troutlodge breeding populations were genotyped with the SNP panel. The global F_{ST} over all SNPs was 0.13 and the pairwise F_{ST} between any two breeding populations ranged from 0.056 to 0.195. Both phylogenetic tree and structure analyses revealed that the odd year and even year populations for the same strain were closely related to each other. Also, the eight populations were clustered into two groups. Based on the results of self-assignment, 97.1% of the Troutlodge breeders were correctly assigned to the population of origin. To further evaluate the accuracy of genetic assignment, we also genotyped 280 egg production fish from three Troutlodge populations of known origin, 49 fish from a Canadian farm, 70 fish from a farm in Idaho and 188 fish from four known non-Troutlodge strains. Among the 280 production fish, 98.2% fish were correctly assigned to the Troutlodge strain of origin. Consistent with the purchasing records, the fish from the two farms were also correctly assigned to two Troutlodge strains. Based on the SNP genotypes, 185 out of 188 fish (98.4%) from the four non-Troutlodge strains could be excluded as Troutlodge fish. In conclusion, our previously developed SNP panel for parentage assignment is also useful to characterize genetic differentiation and has sufficient power for genetic assignment of commercial rainbow trout strains used in this study.

Mobrand, L.E., J. Barr, L. Blankenship, D.E. Campton, T.T.P. Evelyn, T.A. Flagg, C.V.W. Mahnken, L.W. Seeb, P.R. Seidel, and W.W. Smoker. 2005. Hatchery reform in Washington State: principles and emerging issues. Fisheries 30(6):11-23.

Hatcheries support nearly all major fisheries for Pacific salmon (*Oncorhynchus* spp.) and steel-head (anadromous *O. mykiss*) in the Pacific Northwest. However, hatcheries have been a major source of controversy for over 30 years. The Hatchery Scientific Review Group (HSRG) was tasked by Congress to identify solutions to well-known problems so hatcheries could better meet their goals of supporting sustainable fisheries and assisting with the conservation of natural populations. The authors reviewed more than 100 facilities and 200 programs and identified three principles of hatchery reform: 1) goals for each program must be explicitly stated in terms of desired benefits and purposes; 2) programs must be scientifically defensible; and 3) hatchery programs must respond adaptively to new information. They also identified several emerging issues critical to the success of hatcheries. They concluded that hatcheries must operate in new modes with increased scientific oversight and that they cannot meet their goals without healthy habitats and self-sustaining, naturally-spawning populations.

Nash, C.E., P.R. Burbridge, and J.K. Volkman. 2005. Guidelines for ecological risk assessment of marine fish aquaculture. National Oceanic and Atmospheric Administration, Technical Memorandum NMFS-NWFSC-71, Silver Spring, MD.

Significant sea lice infestation of farmed salmon has never been an issue because net-pens are located in areas where the salinity is too low for lice proliferation in Washington.

Noakes, D.J. 2014. Environmental impacts of salmon net pen farming. Salmon: biology, environmental impact and economic importance. Nova Science Inc., New York, NY, p. 239-256.

The absolute and relative abundance of farmed and wild is a factor in determining potential impacts. Genetic and ecological impacts are likely to be greatest in Norway and Scotland where farmed Atlantic Salmon interact with depressed populations of wild Atlantic Salmon. Similar impacts are also possible in the United States but these are likely to be insignificant when compared to similar interactions involving cultured salmon produced by the large-scale enhancement programs in the Pacific. Waste discharge and pollution from salmon farming appear to be primarily near-field with reversable impacts within 100 m of farm sites.

Price, C.S. and J.A. Morris, Jr. 2013. Marine cage culture and the environment: Twenty-first century science informing a sustainable industry. National Oceanic and Atmospheric Administration. Technical Memorandum NOS-NCCOS-164. Silver Spring, MD. Available online: http://aquaticcommons.org/14685/1/NOS%20NCCOS%20164.pdf

This report provides a comprehensive review of some predominant environmental risks that marine fish cage culture aquaculture, as it is currently conducted, poses in the marine environment and designs and practices now in use to address these environmental risks in the U.S. and elsewhere. Today's finfish aquaculture industry has learned, adapted and improved to lessen or eliminate impacts to the marine habitats in which it operates. The authors conducted a critical review of the large body of scientific work published since 2000 on the technological innovation that has made it possible to grow marine finfish in the coastal and open ocean. Along with this opportunity comes environmental risk. As a federal agency charged with stewardship of the nation's marine resources, NOAA requires tools to evaluate the benefits and risks that aquaculture poses in the marine environment, to implement policies and regulations which safeguard our marine and coastal ecosystems, and to inform production designs and operational procedures compatible with marine stewardship.

There is an opportunity to apply the best available science and globally proven best management practices to regulate and guide a sustainable United States marine finfish farming aquaculture industry. There are strong economic incentives to develop this industry, and doing so in an environmentally responsible way is possible if stakeholders, the public and regulatory agencies have a clear understanding of the relative risks to the environment and the feasible solutions to minimize, manage or eliminate those risks. This report spans many of the environmental challenges that marine finfish aquaculture faces to serve as a useful tool to those interested in and responsible for the industry and safeguarding the health, productivity and resilience of our marine ecosystems.

Water Quality: The primary potential effects to water quality associated with marine cage culture include dissolved nitrogen and phosphorus, turbidity, lipids and dissolved oxygen fluxes. Usually there are no measurable effects 30 m beyond the cages when farms are sited in well-flushed waters. The trend of many studies over the last 20 y indicates that improvements in feed formulation and feeding efficiency are the major reasons for decreased nutrient loading and acceptable water quality in and near farms, and explains why significant enrichment to the water column at offshore farms is generally not detected.

Benthic Effects: Well-managed farms may exhibit little perturbation and, where chemical changes are measured, impacts are typically confined to within 100 m of the cages. Benthic chemical recovery is often rapid following harvest. Within an adaptive management framework, a good monitoring program can be used to adjust farm management to avert serious and persistent impacts to the benthos.

Marine Life: If farm nutrients accumulate and persist in the water column or sediment, marine organisms can be impacted. At appropriately-sited and well-managed farms, natural processes can be sufficient to assimilate nutrients. At some farm sites, a phytoplankton response to nutrient loading was reported, but generally this is a low risk and causal linkages to algal blooms are not evident. Changes in the benthic community are evident when sediments become enriched with organic farm waste nutrients. At well flushed sites in deep water and with efficient feed management, ecological impacts tend to be minimal and confined to the area just beneath the cages. The excess food and waste released from fish cages may be food for wild fish, especially benthic feeders. Cages may also provide shelter and foraging habitat for wild fish. These characteristics may be beneficial to the local and regional environment.

Chemicals: The use of antibiotics, therapeutants and antifoulants at marine fish farms has declined greatly (up to 95%) in the last 20 y, resulting in decreased potential for secondary harmful effects of these chemicals on the marine environment. Heavy metals from feed and antifoulants are known to accumulate beneath cages, but are often in low concentrations and sequestered in the sediment.

Quinn, P., J.R. Winton, D. Huppert, and R. Hilborn. 2008. An evaluation of the effects of conservation and fishery enhancement hatcheries on wild populations of salmon. Advances in Marine Biology 53:61-194.

The historical, political and scientific aspects of salmon hatchery programs designed to enhance fishery production, or to recover endangered populations, are reviewed. The authors point out that the establishment of hatcheries has been a political response to societal demands for harvest and conservation; given this social context, they then critically examined the levels of activity, the biological risks, and the economic analysis associated with salmon hatchery programs. A rigorous analysis of the impacts of hatchery progress was hindered by the lack of standardized data on release sizes and survival rates at all ecological scales, and since hatchery program objectives are rarely defined, it was also difficult to measure their effectiveness at meeting release objectives. Debates on the genetic effects of hatchery programs on wild fish have been dominated by whether correct management practices can reduce negative outcomes, but they note that there has been an absence of programmatic research approaches addressing this important issue. Competitive interactions between hatchery and wild fish were observed to be complex, but studies researching approaches to reduce these interactions at all ecological scales during the entire salmon life history have been rare, and thus are not typically considered in hatchery management. Harvesting of salmon released from fishery enhancement hatcheries likely impacts vulnerable wild populations; managers have responded to this problem by mass marking hatchery fish, so that fishing effort can be directed towards hatchery populations. However, the effectiveness of this approach is dependent on accurate marking and production of hatchery fish with high survival rates, and it is not yet clear whether selective fishing will prevent overharvest of wild populations. Research demonstrating disease transmission from hatchery fish to wild populations was observed to be equivocal; evidence in this area has been constrained by the lack of effective approaches to studying the fate of pathogens in the wild. They reviewed several approaches to studying the economic consequences of hatchery activities intended to inform the social decisions surrounding programs, but recognized that placing monetary value on conservation efforts or on hatcheries that mitigate cultural groups' loss of historical harvest opportunities may complicate these analyses. Economic issues have rarely been included in decision making on hatchery programs.

Refstie, S., S.J. Helland, and T. Storebakken. 1997. Adaptation to soybean meal in diets for Rainbow Trout, *Oncorhynchus mykiss*. Aquaculture 15*3*(3-4):263-272.

Three experiments were conducted with rainbow trout maintained in fresh water. In Experiment 1, growth was measured in groups of 34-g trout fed either a fish meal control diet (Diet 1) or a diet 60% soybean meal (Diet 2) for two periods of 28 days each. In Experiment 2, Diets 1 and 2 were fed to groups of 100-g trout for 10 days prior to stripping of feces and digestibility determination. In Experiment 3, groups of 200-g trout were fed either a fish meal control diet containing 100 mg Y_2O_3/kg diet as an inert marker (Diet 3), or a diet containing 40% soybean meal and 100 mg Yb_2O_3/kg diet as an inert marker (Diet 4) for an adaptation period of 7 days. Thereafter, both diets were offered in excess in a 1:1 mixture for 2 days. Qualitative preference for the two diets was estimated as the proportions of Y_2O_3 and Yb_2O_3 in feces. Subsequently, digestibility was determined for all diets.

Refstie, S., Ø. J. Korsøen, T. Storebakken, G. Baeverfjord, I. Lein, and A.J. Roem. 2000. Differing nutritional responses to dietary soybean meal in Rainbow Trout (*Oncorhynchus mykiss*) and Atlantic Salmon (*Salmo salar*). Aquaculture 190(1-2):49-63.

This study was carried out to compare the responses of Rainbow Trout and Atlantic Salmon to temperature (LT)-fish meal as the sole protein ingredient or 32% fish meal and 30% soybean meal, were each fed to triplicate groups of 0.1 kg trout or 0.2 kg salmon maintained in 7 ºC freshwater. The experiment lasted 84 days, divided into three periods. Weight gain of the trout was similar with both diets, whereas the salmon gained 44% more weight with the fish meal diet than with the soybean meal diet. The apparent digestibilities were 6% higher for nitrogen, 8% higher for fat, and 11% higher for energy in the trout than in the salmon. The feed conversion ratio (FCR) was 24% lower, the nitrogen retention 34% higher, and the energy-retention 28% higher in the salmon than in the trout. Within each species, the digestibility of nitrogen was similar for both diets. In trout, the digestibilities of fat and energy were 4% higher, the nitrogen retention 8% higher, and the energy retention 9% higher with the fish meal than with the soybean meal diet. Similarly, but more severe within the salmon, the respective digestibilities of fat and energy were 16 and 9% higher, the nitrogen retention 19% higher, and the energy retention 23% higher with the fish meal than with the soybean meal diet. Both species developed enteritis in the distal intestine when fed the soybean meal diet.

Reinbold, D., G.H. Thorgaard, and P.A. Carter. 2009. Reduced swimming performance and increased growth in domesticated Rainbow Trout, *Oncorhynchus mykiss*. Canadian Journal of Fisheries and Aquatic Science 66:1025–1032.

Domesticated populations of Rainbow Trout probably have been selected for high growth rates and large body size, which may have resulted in reduced sprint swimming performance. The authors hypothesized that more domesticated populations of Rainbow Trout would have higher growth rates and larger body size, but slower swim speed, relative to semi-wild populations. They tested this hypothesis by measuring body mass, body length, and sprint swim speed multiple times over 92 d in progeny from crosses between males from three clonal lines and an outbred female. They found significantly higher body masses and significantly slower swim speeds in the highly domesticated Arlee and Hot Creek progeny groups compared with the semi-wild Swanson hybrid progeny group, supporting our hypothesis. Growth rates also differed significantly among groups, but at the ages measured, the Swanson hybrid progeny had an intermediate growth rate. However, given the differences in body mass, either growth rates were higher in the more domesticated progeny groups at young unmeasured ages and (or) they hatched at a significantly larger body mass than the semi-wild Swanson hybrid.

Rooney, R.C., and C.L. Podemski. 2009. Effects of an experimental Rainbow Trout (*Oncorhynchus mykiss*) farm on invertebrate community composition. Canadian Journal of Fisheries and Aquatic Sciences 66:1949–1964.

The authors examined the development of changes in the zoobenthos along a transect from an experimental Rainbow Trout farm in Lake 375, Experimental Lakes Area, northwestern Ontario, Canada. After 2 months, invertebrate abundance was reduced under the fish cage (2542 ± 569 individualsm–2) compared with samples collected 45 m away (16137 ± 2624 individualsm–2). Taxa richness was also depressed, but changes in biomass were variable. Reductions in abundance and richness at high organic loading levels are consistent with earlier models developed for the marine environment of responses to organic loading in marine systems. After two production cycles, the significant principal components axis explaining 76% of total variance in abundance was correlated with distance from the cage (Spearman rank correlation, r = –0.775, p = 0.014) and with chemical variables recommended for freshwater aquaculture monitoring (Pearson's correlation coefficient, r = 0.78, 0.76, and 0.75 with p = 0.013, 0.018, and 0.020 for pore-water ammonia and sediment Cu and Zn, respectively). The effects of farming were localized, dissipating within 15 m of the cage edge. Invertebrate abundance demonstrated the most potential for incorporation into monitoring schemes at new farms. At established farms, richness may be a valuable monitoring metric.

Rust, M.B., K.H. Amos, A.L. Bagwill, W.W. Dickhoff, L.M. Juarez, C.S. Price, J.A. Morris Jr, and M.C. Rubino. 2014. Environmental performance of marine net-pen aquaculture in the United States. Fisheries 39(11):508-524.

The United States has a small net-pen salmon industry dating back over 40 y and a nascent net-pen industry for other marine fish. The United States net-pen aquaculture sector has improved its resource efficiency in terms of the amount of fish meal and fish oil used in feeds and reduced its environmental impacts in terms of the mass loading and impact of nutrient discharge on the receiving ecosystem, the incidence and treatment of fish diseases, the use of antibiotics, and the number and impact of fish escapes, while increasing production. These changes can be attributed to a combination of advances in science and technology, rising cost of fish meal/oil, improved management, and informed regulatory practices. Net-pen aquaculture has become an efficient food production system. Existing laws and regulations in the United States effectively address most of the potential adverse environmental effects of net-pen aquaculture.

In Washington, significant sea lice infestation of farmed salmon has never been an issue because net-pens are located in areas where the salinity is too low for lice proliferation (Nash et al. 2005); therefore, treatment has not been necessary.

U.S. fish farms must monitor discharges to the benthos and water column to meet the standards of the Clean Water Act, which established the National Pollutant Discharge Elimination System (NPDES). In 2004, the U.S. Environmental Protection Agency (USEPA) developed a national effluent rule for net-pen aquaculture (USEPA 2004), establishing effluent limitations for aquaculture facilities into waters of the United States.

Environmental impact models now allow regulators to assess the suitability of sites, understand the potential risks and benefits of proposed net-pen operations, and estimate the limits of acceptable farm biomass before they are permitted.

Troyer, R.M., S.E. LaPatra and G. Kurath. 2000. Genetic analyses reveal unusually high diversity of infectious haematopoietic necrosis virus in rainbow trout aquaculture. Journal of General Virology 81(12):2823-2832.

Infectious haematopoietic necrosis virus (IHNV) is the most significant virus pathogen of salmon and trout in North America. Previous studies have shown relatively low genetic diversity of IHNV within large geographical regions. In this study, the genetic heterogeneity of 84 IHNV isolates sampled from rainbow trout (*Oncorhynchus mykiss*) over a 20-y period at four aquaculture facilities within a 12 mile stretch of the Snake River in Idaho, USA was investigated. The virus isolates were characterized using an RNase protection assay (RPA) and nucleotide sequence analyses. Among the 84 isolates analyzed, 46 RPA haplotypes were found and analyses revealed a high level of genetic heterogeneity relative to that detected in other regions. Sequence analyses revealed up to 7±6% nucleotide divergence, which is the highest level of diversity reported for HNV to date. Phylogenetic analyses identified four distinct monophyletic clades representing four virus lineages. These lineages were distributed across facilities, and individual facilities contained multiple line ages. These results suggest that cocirculating IHNV lineages of relatively high genetic diversity are present in the IHNV populations in this rainbow trout culture study site. Three of the four lineages temporal trends consistent with rapid evolution.

U.S. Environmental Protection Agency (USEPA). 2004. Aquaculture operations—laws, regulations, policies and guidance. Available online: www.epa.gov/agriculture/anaqulaw. html#Effluent%20Guidelines.

In 2004, the U.S. Environmental Protection Agency (USEPA) developed a national effluent rule for net-pen aquaculture (USEPA 2004), establishing effluent limitations for aquaculture facilities into waters of the United States. Environmental impact models now allow regulators to assess the suitability of sites, understand the potential risks and benefits of proposed net-pen operations, and estimate the limits of acceptable farm biomass before they are permitted.

Utter, F. and J. Epifanio. 2002. Marine aquaculture: genetic potentialities and pitfalls. Reviews in Fish Biology and Fisheries, 12(1):59-77.

The authors made several recommendations for future aquaculture efforts based on their extensive review of available information.

1. Articulate simple and clearly-defined goals;
2. Use indigenous populations, or;
3. Use fully contained non-native populations;
4. Commit to monitoring and evaluation; and
5. Take prompt and effective remedial actions when required.

Containment through sterility provides a valuable model an aquaculture operation where releases increase genetic risk to indigenous populations.

ESCAPEMENT ISSUES

Abrantes, K.G., J.M. Lyle, P.D. Nichols, and J.M. Semmens. 2011. Do exotic salmonids feed on native fauna after escaping from aquaculture cages in Tasmania, Australia? Canadian Journal of Fisheries and Aquatic Sciences 68(9):1539-1551.

Atlantic Salmon, and Rainbow Trout, are farmed in Tasmania, Australia, where fish sometimes escape into the natural environment. If escapees are able to survive and feed on native fauna, it is likely that they will have ecosystem impacts. Stomach content, body condition (muscle lipid content and Fulton' K), stable isotope, and fatty acid analysis were used to determine if escaped salmonids feed on native fauna. Results indicate that, in general, escaped salmonids do not feed on native fauna. Salmonids loose condition after escaping, and escapee stomachs were mostly empty or contained non-nutritious material or feed pellets. Nevertheless, almost a quarter of rainbow trout stomachs contained native fauna. The majority of escapees had biochemical composition similar to caged animals, indicating that these fish had not switched to feed on local food sources. However, a small fraction of escapees conclusively showed changes in biochemical parameters indicative of a shift to feeding on native fauna. Given the numbers and frequency of escapes, this can have an important impact on native species and on the ecology of Macquarie Harbor.

Amos, K.H., J. Thomas, and B. Stewart. 2001. Pathogen transmission between wild and cultured salmonids: risk avoidance in Washington State, United States of America. Pages 83–89 in C.J. Rodgers, editor. Proceedings of an international conference on risk analysis in aquatic animal health. World Organization for Animal Health, Paris.

Most escaped farmed fish have low fitness for the wild and quickly become easy victims of predators such as marine mammals, other fish, and birds. Escapees are unlikely to generate an infectious dose (or infective pressure) sufficient to result in disease in a healthy, wild population. The mere presence of a pathogen alone will not cause disease without environmental factors that play a large role in triggering disease events.

Baskett, M.L., S.C. Burgess, and R.S. Waples. 2013. Assessing strategies to minimize unintended fitness consequences of aquaculture on wild populations. Evolutionary Applications 6(7):1090-1108.

Spatiotemporal patterns of distribution of Sea Bass, *Dicentrarchus labrax,* and Sea Bream, *Sparus aurata*, and their influence on artisanal fisheries are explored before and after an escape event that released 1.5 million fish into the wild off La Palma (Canary Islands). Data were collected by *in situ* visual census and first sale data as a proxy of artisanal fisheries landings. Permutational ANOVA of escapee abundances in shallow coastal habitats revealed consistent spatial patterns that linked densities of these fish to distance from escape point, whereas temporal patterns were related to a higher biomass released during winter. A nearby marine protected area did not show different densities of escaped fish. Local artisanal fleet catches accurately reflected the massive escape event and offer the main contingency force to mitigate the potential negative effects of massive escape events over shallow coastal habitats.

Blanchfield, P.J., L.S. Tate, and C.L. Podemski. 2009. Survival and behaviour of Rainbow Trout (*Oncorhynchus mykiss*) released from an experimental aquaculture operation. Canadian Journal of Fisheries and Aquatic Sciences 66(11):1976-1988.

The potential for farmed fish that have escaped from open-cage aquaculture operations to affect native populations will depend on their survival and behavior in the wild. Used standard commercial practices to rear 10 tons of Rainbow Trout in a 23 ha lake at the Experimental Lakes Area (Ontario, Canada). Each fall (2003–2005), they released farmed Rainbow Trout (escapees) into the study lake and monitored their movements using automated positioning telemetry. Rainbow Trout experienced high annual mortality

(~50%), with none surviving beyond 3 years. Farmed fish had narrowly defined pelagic distributions that comprised the upper few meters of the water column, even when at the cage site. Although released Rainbow Trout dispersed throughout the study lake, most spent significant portions of time at the cage site, especially during normal operation when commercial feed was available. Core use areas (50% Kernel) included the farm for half of the released fish. Surviving Rainbow Trout showed continued reliance upon the cage site in their second year. However, wide dispersal, high growth rate, and lack of reliance on the cage site by some escaped fish warrant further research to assess potential effects of open-cage aquaculture in the water bodies where the industry occurs.

Bridger, C.J., R.K. Booth, R.S. McKinley, and D.A. Scruton. 2001. Site fidelity and dispersal patterns of domestic triploid steelhead trout (*Oncorhynchus mykiss* Walbaum) released to the wild. ICES Journal of Marine Science 58:510–516.

A combined acoustic and radio telemetry system was deployed within Bay d'Espoir, Newfoundland, to determine whether cultured steelhead trout (*Oncorhynchus mykiss*) released in the vicinity of a commercial aquaculture site remain at the site (site fidelity) or disperse from it. Two sets of fish releases (summer and winter 1998) were performed to determine seasonal effects on movements in the wild. Simulated escapes in summer involved 68 fish released from the cage system and 66 fish released from a cage towed approximately 1 km away from the grow-out site. The winter releases involved three batches of 30 fish each, one from the cage system and two off-site over the side of a boat (at 200 and 1000 m distance) after transport on board, with no cage towing involved. The results suggest site fidelity among steelhead released during the growing season. Fidelity was only slightly larger for on-site releases than off-site releases. Off-site released steelhead make a rapid return to their rearing sites, suggesting homing behavior. During the winter, the movement to the overwintering release site was less directed with a higher degree of dispersal. Released steelhead eventually dispersed from the release site, and in both seasons displayed a directed movement to the hydroelectric spillway, which is also the location of the local salmonid hatchery. Implications of the results are discussed in light of the development of recapture methodologies for aquaculture salmonids.

Brown, R., B. Pflugrath, D. Trott, and G. McMichael. 2012. Movement of net-pen–released Rainbow Trout in Lake Rufus Woods and Lake Pateros. Report to The Confederated Tribes of the Colville Reservation. 42 p. Available online: https://www.researchgate.net/profile/Geoffrey_Mcmichael2/publication/267268810_Movement_of_Net-Pen-Released_Rainbow_Trout_in_Lake_Rufus_Woods_and_Lake_Pateros/links/544930890cf2f63880810153.pdf

Large Rainbow Trout *Oncorhynchus mykiss* (>10 lb) have been present in the reservoir since net-pen aquaculture began in 1989. Many of these fish are escapees from aquaculture facilities or have been purchased by the Colville Tribes for release into the reservoir to augment the fishery. To maintain this fishery, the Fish and Wildlife Department of the Colville Tribes began research in 2008 to examine angling pressure, catch rates, and trout harvest. Understanding the extent of entrainment of rainbow trout at Chief Joseph Dam was also an important goal. To determine appropriate management strategies for the fishery, an acoustic telemetry study was initiated in fall 2010. Net-pen–reared Rainbow Trout were surgically implanted with acoustic transmitters and released from the net-pen location at river kilometer (rkm) 915.5. Releases of 99, 100, and 94 fish were made in November 2010, and January and March 2011. Autonomous receivers (called nodes) were used to detect the tagged fish at several locations upstream and downstream of the release area, including the forebay of Chief Joseph Dam and two sites downstream of the dam. Fish were monitored until the end of July 2011. Entrainment of net-pen–released rainbow trout appeared to be associated with the rates of spill past Chief Joseph Dam because a large number of fish were entrained at the onset of spill. There was no clear relationship between the size of fish and their likelihood of entrainment. Because the fish appeared to be removed from Lake Rufus Woods fairly quickly, due to either harvest or entrainment, it would be unlikely that release of smaller fish (~1 lb) from net-pens instead of larger fish would yield a high number of larger, trophy-size fish. However, the probability that released fish will remain in the reservoir would likely be higher if the releases are made when there is little if any spill. It was clear from several sources of data (residence time, last detection location, location of recovered tags, and travel times) that there are two main areas where implanted fish spent a large amount of time and were likely to be harvested. These locations were the forebay of Chief Joseph Dam and the area adjacent to the net-pens where the fish were released. Another area used to a moderate extent was near rkm 933, close to another set of fish farms.

Charles, C., P.J. Blanchfield, and D.M. Gillis. 2017. Site fidelity of escaped rainbow trout to an experimental freshwater aquaculture facility and habitat overlap with native fish fauna." Aquaculture Environment Interactions 9: 415-428.

The distribution and habitat use of escaped farmed fish is often difficult to assess after their dispersal from commercial open-pen aquaculture facilities. The authors examined site fidelity of rainbow trout *Oncorhynchus mykiss* after release from an experimental farm (escaped fish) in a small lake, as well as habitat overlap (3D kernel utilization distributions) with a native salmonid, lake trout *Salvelinus namaycush*. Fish were implanted with telemetry transmitters, and their movements were monitored by acoustic arrays during (2006−2007) and after (2008−2009) commercial production. This latter period allowed for assessment of habitat use by escapees in the absence of any influence of cage production, simulating conditions similar to fallowing or long-distance dispersal. Escaped rainbow trout were regularly present at the cage site (23% of total positions) during production, typically at times of the day which coincided with feeding, but were rarely near the cage site (2% of total positions) during the post-production period, and instead greatly increased their occupancy of the near-shore region. Lake trout did not display an affinity to the cage site in either the production or post-production periods (~1% of total positions), and volumetric overlap with rainbow trout was relatively low throughout the entire study. Their results indicate that in the absence of ongoing production at commercial aquaculture operations. Rainbow Trout escapees can readily switch to foraging on native fauna in the near-shore regions of the lake and did not directly compete with native lake trout.

Clark, D., K.L. K. Murphy, and A. Windrope. 2017. Cypress Island Atlantic Salmon net pen failure: an investigation and review. Washington Department of Natural Resources, Olympia, WA. 120 p.

On August 19, 2017, a ten-cage net pen rearing 305,000 Atlantic salmon off Cypress Island in northern Puget Sound collapsed. Previously on July 24-25 in which moorings twice failed and the net pen moved hundreds of feet. The probable cause of both the incidents was failure to adequately clean the nets containing the fish.

- Insufficient cleaning, led to excessive biofouling by mussels and other marine organisms.
- Breakdowns in net cleaning machines contributed to this condition.
- The excessive biofouling significantly increased the drag (force) on the net pen array from tidal currents.
- Increased drag exceeded the holding power of the mooring system in both incidents.
- On July 24-25, the mooring system experienced both anchor dragging and breaking of attachment points between the moorings and the net pen.
- On August 19, some combination of anchor dragging, failure of mooring attachment points, and failure of structural members of the net pen framing resulted in the collapse of the net pen.
- Failure to address the biofouling effectively after the July incident directly contributed to the August failure.

Tidal currents were the mechanism of the July incident and August failure. The tidal currents on July 24 were the strongest during the summer of 2017 but were not unprecedented. The tidal currents on August 19 were less than those of July 24-25. While tidal currents continued to increase on August 20 and 21, the August 21 solar eclipse did not alter the normal pattern of seasonal tidal strength.

Properly designed, sited, and maintained, salmon net pens should be able to withstand combinations of tidal currents, wind, and wave forces that reasonably could be expected to occur at a site. This net pen had been operated without incident at this site for seven years (and for nine years previously at a location several hundred feet away).

State agencies concluded that:

- There were 305,000 fish in the net pen prior to failure.
- In August and September, Cooke reported harvesting/extracting 145,000 fish from the collapsed net pen.
- The Panel concluded that Cooke could only have extracted 42,000 to 62,000 fish, 43% of what Cooke reported.
- The Panel estimates that between 243,000 and 263,000 fish actually escaped.
- Of the escaped fish, 57,000 have been recovered.
- Between 186,000 and 206,000 Atlantic salmon remain unaccounted for.

Cooke Aquaculture Pacific, Northwest Resources Law letter dated January 29, 2018. Cooke Comments to Incident Review Panel (IRP) regarding IRP draft report, *Cypress Island Atlantic Salmon net pen failure: an investigation and review*.

The comment letter from Cooke Aquaculture Pacific reviews the *Draft Report on the Cypress Island Atlantic Salmon net pen failure: an investigation and review*, which was produced by the IRP on January 30, 2018. Cooke was given a brief amount of time by the IRP (from 9:00 a.m. on Friday, January 26, 2018 to 9:00 a.m. on Monday, January 29, 2018) to review the report and to make only factual corrections. Cooke produced this letter, dated January 29, 2018, summarizing their concerns and factual corrections to the draft report. While Cooke Aquaculture Pacific disagreed with a number of the conclusions in the report (as set forth in the January 29, 2018 letter), it concurs with the conclusions regarding a lack of environmental impacts associated with escaped fish. The letter also states the following:

Several factually correct findings found in the draft report that are worth enumerating:

- Cooke agrees that its fish were healthy at the time of release, with no endemic bacterial, viral, or parasitic (such as sea lice) pathogens detected in the group sampled immediately after release.
- Cooke agrees that the fish stock used do not eat in the wild and can confirm that all gastrointestinal tracts sampled by DFW, Cooke, and tribes were empty.
- Cooke agrees that the declining condition and weight of released fish were an important factor post-release in decreasing survival of fish in Puget Sound.
- Cooke agrees that the stress of an unfamiliar environment and lack of regular feedings resulting in a decreasing nutritional profile likely played a significant role in reducing survival of escaped fish.
- Cooke agrees with the assessment that the escaped fish in saltwater were all likely dead by November/December 2017.
- Cooke agrees with the assessment of freshwater survival and agrees with the need to monitor the Skagit and other rivers where Atlantic salmon have been detected. In fact, Cooke has already offered to fund that monitoring.

Consuegra, S., N. Phillips, G. Gajardo, and C.G. de Leaniz. 2011. Winning the invasion roulette: Escapes from fish farms increase admixture and facilitate establishment of non-native rainbow trout. Evolutionary Applications 4(5):660-671.

Aquaculture is a major source of invasive aquatic species, despite the fact that cultured organisms often have low genetic diversity and tend to be maladapted to survive in the wild. Yet, to what extent aquaculture escapees become established by means of high propagule pressure and multiple origins is not clear. The authors analyzed the genetic diversity of 15 established populations and four farmed stocks of non-native rainbow trout in Chile, a species first introduced for recreational fishing around 1900, but which has in recent decades escaped in large numbers from fish farms and become widespread. Hybrids between farm escapes and established trout were present in all rivers at frequencies ranging between 7% and 69%, and population admixture was positively correlated with genetic diversity. They suggest that non-native salmonids introduced into the Southern Hemisphere could benefit from admixture because local adaptations may not have yet developed, and there may be initially little fitness loss resulting from outbreeding depression.

Dempster, T., P. Arechavala-Lopez, L.T. Barrett, I.A. Fleming, P. Sanchez-Jerez, and I. Uglem. 2018. Recapturing escaped fish from marine aquaculture is largely unsuccessful: alternatives to reduce the number of escapees in the wild. Reviews in Aquaculture 10(1):153-167.

Farmed fish that escape and mix with wild fish populations can have significant ecological and genetic consequences. To reduce the number of escaped fish in the wild, recapture is often attempted. The authors review the behaviors of escapees post-escape, and how recapture success varies with escaped fish size, the size of the initial escape event and recapture methods. Success rates of fishing gears varied among species, with gill-nets and coastal barrier nets most effective for recapture of salmonids. Recapture success was strongly negatively correlated with both fish size and the number of fish escaped, regardless of species. Recapture success was universally low across all studied species (8%). Numerous tracking studies of escaped fish indicate that recapture efforts should be initiated within 24 h of an escape incident for highest recapture success. However, most large escape events are due to storms, which mean recapture efforts rarely start within this timeframe. Recapture of escaped fish is broadly ineffective in marine habitats, with rare exception. High bycatch rates during ineffective recapture attempts imply that large-scale recapture efforts should be weighed against the possibility of affecting wild fish populations negatively. They suggest three alternative approaches to reduce escapee numbers in wild habitats: (i) protect populations of predatory fish around sea-cage farms from fishing, as they prey upon smaller escapees; (ii) construct impact offset programs to target recapture in habitats where escapees can be efficiently caught; and (iii) ensure technical standards are legislated so that fish farmers invest in preventative technologies to minimize escapes.

Kendall, N.W., G.W. Marston, and M. M. Klungle. 2017. Declining patterns of Pacific Northwest steelhead trout (*Oncorhynchus mykiss*) adult abundance and smolt survival in the ocean. Canadian Journal of Fisheries and Aquatic Sciences 74(8):1275-1290.

Examination of population abundance and survival trends over space and time can guide management and conservation actions with information about the spatial and temporal scale of factors affecting them. The authors analyzed steelhead trout (anadromous *Oncorhynchus mykiss*) adult abundance time series from 35 coastal British Columbia and Washington populations along with smolt-to-adult return (smolt survival) time series from 48 populations from Washington, Oregon, and the Keogh River in British Columbia. More than 80% of the populations have declined in abundance since 1980. A multivariate autoregressive state-space model revealed smolt survival four groupings: Washington and Oregon coast, lower Columbia River, Strait of Juan de Fuca, and Puget Sound – Keogh River populations. Declines in smolt survival rates were seen for three of the four groupings. Puget Sound and Keogh River populations have experienced low rates since the early 1990s. Correlations between population pairs' time series and distance apart illustrated that smolt survival rates were more positively correlated for proximate populations, suggesting that important processes, including those related to ocean survival, occur early in the marine life of steelhead.

Kostow, K.E., A.R. Marshall, and S.R. Phelps. 2003. Naturally spawning hatchery steelhead contribute to smolt production but experience low reproductive success. Transactions of the American Fisheries Society 132(4):780-790.

The authors used genetic mixture analyses to show that hatchery summer-run steelhead *Oncorhynchus mykiss*, an introduced life history in the Clackamas basin of Oregon, where only winter-run steelhead are native, contributed to the naturally produced smolts out-migrating from the basin. Hatchery-produced summer steelhead smolts were released starting in 1971, and returning adults were passed above a dam into the upper Clackamas River until 1999. In the 2 y of study, summer steelhead adults, mostly hatchery fish, made up 60 to 82% of the natural spawners in the river. Genetic results provided evidence that interbreeding between hatchery summer and wild winter steelhead was likely minor. Hatchery summer steelhead reproductive success was relatively poor. They estimated that they produced only about one-third the number of smolts per parent that wild winter steelhead produced. However, the proportions of summer natural smolts were large (36–53% of the total naturally produced smolts in the basin) because hatchery adults predominated on the spawning grounds during our study. Very few natural-origin summer adults were observed, suggesting high mortality of the naturally produced smolts following emigration. Counts at the dam demonstrated that hatchery summer steelhead predominated on natural spawning grounds throughout the 24-y hatchery program. Their data support a conclusion that hatchery summer steelhead adults and their offspring contribute to wild winter steelhead population declines through competition for spawning and rearing habitats.

Martens, M.T., A.J. Wall, G.G. Pyle, B.A. Wasylenko, W.A. Dew, R.H. Devlin, and P.J. Blanchfield. 2014. Growth and feeding efficiency of wild and aquaculture genotypes of Rainbow Trout (*Oncorhynchus mykiss*) common to Lake Huron, Canada. Journal of Great Lakes Research 2014.

Selective breeding of salmonid fishes for the purpose of commercial aquaculture has resulted in domesticated strains possessing a divergent physiological and behavioral phenotype from that of wild conspecifics. Freshwater production of Rainbow Trout has been occurring in regions of Lake Huron, Canada, for decades yet the growth and performance of domestic (aquaculture) versus wild (naturalized) strains are poorly understood. The authors conducted two trials to examine growth differences between size-matched wild and domestic strains of juvenile Rainbow Trout: 1) reared separately and fed to satiation; and 2) reared together and fed a reduced ration to induce competition. They used bioenergetics models to assess strain-specific growth rates across a range of water temperatures (5, 10, 15 and 18°C) as well as Lake Huron temperatures during the open-water season. Domestic rainbow trout showed a growth advantage throughout the 102 d trials, and by the end of the study had achieved a mass two-fold that of the wild strain and had greater fork length, condition, and thermal growth coefficient (TGC) under both treatments. Rapid growth of domestic strain fish was achieved through the combination of enhanced feed consumption (by ~40%) and feeding efficiency (up to 60% lower feed conversion ratio) relative to wild fish. Divergence in growth rates between strains was most pronounced (N3×) when modeled with Lake Huron open-water temperatures. They demonstrate that the growth and feed- conversion efficiency differ significantly between these two strains of Rainbow Trout under laboratory conditions, suggesting that differences could be even greater in nature.

Jacobsen, J.A., and L.P. Hansen. 2001. Feeding habits of wild and escaped farmed Atlantic Salmon, *Salmo salar* L., in the Northeast Atlantic. ICES Journal of Marine Science 58:916– 933.

The stomach contents of 2,992 wild and 863 putative escaped farmed Atlantic Salmon caught on floating long-lines in a Faroese research fishery in the late autumn (November–December) and winter (February–March) in the Northeast Atlantic (63–66°N and 1–10°W) during three consecutive fishing periods 1992/1993–1994/1995 were analyzed. Hyperiid amphipods of the genus *Themisto*, euphausiids and mesopelagic shrimps are important sources of food for salmon in the autumn period and various mesopelagic fish as lantern fishes, pearlsides and barracudinas become equally important during the late winter period. The occasional presence in the stomachs of larger fish such as herring, blue whiting and mackerel is not considered to be evidence that these fish are a main source of food for salmon in the sea north of the Faroes. The proportion of stomachs containing food was significantly lower during autumn (53%) than during winter (78%). However, temperature-dependent evacuation rates could partly explain the apparent lower stomach content during the autumn, since the average ambient sea-surface temperature is 7°C in autumn compared to 3°C in winter. There was evidence of selective foraging. Fish were preferred over crustaceans, and amphipods were chosen over euphausiids. Large salmon (3+SW) tended to be more piscivorous than smaller fish. There was no difference in condition factor, number and weight proportions of prey, or in diet between wild and escaped farmed salmon, which suggests that escaped farmed salmon adapt well to the "wild" life in the ocean.

Jensen, Ø., T. Dempster, E.B. Thorstad, I. Uglem, and A. Fredheim. 2010. Escapes of fishes from Norwegian sea-cage aquaculture: causes, consequences and prevention. Aquaculture Environment Interactions 1(1):71-83.

The escape of fish from aquaculture is perceived as a threat to wild fish populations. The escapes problem is largely caused by technical and operational failures of fish farming equipment. In Norway, 3.93 million Atlantic Salmon *Salmo salar*, 0.98 million Rainbow Trout *Oncorhynchus mykiss* and 1.05 million Atlantic Cod *Gadus morhua* escaped from 2001 to 2009. Salmonids primarily escape after structural failures of containment equipment, while a far greater proportion of cod than salmon escape through holes in the nets. Correlative evidence suggests that after the Norwegian technical standard (NS 9415) for sea-cage farms took effect in 2004, the total number of escaped Atlantic Salmon declined from >600,000 (2001 to 2006) to <200,000 fish/y (2007 to 2009), despite the total number of salmon held in sea-cages increasing by 44% during this period. No similar decrease in escaped cod has occurred, suggesting that other measures, such as improved netting materials for sea-cages, are required. In addition to escaping as juveniles or adults, cod may reproduce in sea-cages, and thus fertilized eggs escape to the environment. The ecological effects of 'escape through spawning' are unclear, and methods to inhibit escape by this mechanism are being explored. To prevent escapes of juvenile and adult fish as sea-cage aquaculture industries develop, the authors recommend that policy-makers implement a five-component strategy: 1) establish mandatory reporting of all escape incidents; 2) establish a mechanism to analyze and learn from the mandatory reporting; 3) conduct mandatory, rapid, technical assessments to determine the causes of escape incidents involving more than 10,000 fish; 4) introduce a technical standard for sea-cage aquaculture equipment coupled with an independent mechanism to enforce the standard; and 5) conduct mandatory training of fish farm staff in escape-critical operations and techniques.

Johnsson, J.I., and M.V. Abrahams. 1991. Interbreeding with domestic strain increases foraging threat of predation in juvenile steelhead trout (*Oncorhynchus mykiss*): an experimental study. Canadian Journal of Fisheries and Aquatic Sciences 48:243–247.

The foraging behavior of laboratory-reared juvenile steelhead trout and steelhead/domesticated rainbow trout hybrids were compared. In 10 replicate experiments, 10 fish from each strain were allowed to choose between foraging in a safe area or an area containing a predator. The hybrid trout were significantly more willing to risk exposure to the predator than were the steelhead. It was possible that differences in the relative willingness to risk exposure may have reflected differences in their susceptibility to predation. A second experiment measured the susceptibility of these two strains to the predator by simulating standardized encounters between predator and prey. Both strains suffered identical mortality rates and therefore were considered to be equally susceptible to the predator. This experiment confirmed that the hybrid trout were significantly more willing to take risks than the wild steelhead. These results indicate that interbreeding between escaped hatchery and wild fish may have a potentially damaging effect on the wild population.

Klinger, D. and R. Naylor. 2012. Searching for solutions in aquaculture: charting a sustainable course. Annual Review of Environment and Resources, 37, pp. 247-276.

Aquaculture is currently the fastest growing animal food production sector and will soon supply more than half of the world's seafood for human consumption. Continued growth in aquaculture production is likely to come from intensification of fish, shellfish, and algae production. Intensification is often accompanied by a range of resource and environmental problems. The authors review several potential solutions to these problems, including novel culture systems, alternative feed strategies, and species choices. They examine the problems addressed; the stage of adoption; and the benefits, costs, and constraints of each solution. Policies that provide incentives for innovation and environmental improvement are also explored. They end the review by identifying easily adoptable solutions and promising technologies worth further investment.

Knudsen, R., P.A. Amundsen, and A.H. Rikardsen. 2011. Individual feeding specialisation of a naïve vs. veteran predators. Ecology of Freshwater Fish 20(4):522-528.

The authors explored the incidence of individual feeding specialization among a naïve predator (non-native Rainbow Trout post-smolts) and two native experienced predators (sea-run Arctic char and sea-trout) in a subarctic Norwegian fjord. Interindividual foraging niche stability was obtained by combining information on stomach contents (recent dietary niche) with trophically transmitted parasite infestation (time-integrated historical dietary niche) of individual predators. Individual fish showed a high degree of resource specialization as prey items such as gammarids and small fish (both potential intermediate host of parasites) rarely co-occurred in stomachs. In both naïve and veteran predators, positive associations between the intensity of a specific parasite species and the occurrence of their respective intermediate host (gammarids or fish) in the stomachs of individual predators demonstrated temporally interindividual feeding specializations. Several behavioral phenotypes clearly co-existed in both naïve and veteran predator populations, including gammaridivore (benthic feeders), piscivore (pelagic feeders) or insectivore (pleuston feeders) individuals. The likely mechanism of this observed interindividual resource specialization in the non-native naïve predators involves a behavioral component of which rapid learning seems to be a key factor.

Leggatt, R.A., P.T. O'Reilly, P.J. Blanchfield, C.W. McKindsey, and R.H. Devlin. 2010. Pathway of effects of escaped aquaculture organisms or their reproductive material on natural ecosystems in Canada. Fisheries and Oceans Canada. Scientific Advisory Secretariat Research Document 2010:70.

This document provides an overview of the pathways of effects escaped aquaculture fish (specifically finfish and bivalves) may have on natural ecosystems in Canada. Escape, survival, dispersal and reproduction of aquaculture organisms have been noted in many areas in Canada, although the scale of escapes in Canada is not known. In general, escaped fish have poor survival, foraging, and reproductive capacity relative to wild conspecifics. However, substantial evidence indicates escaped Atlantic Salmon can affect wild conspecifics through juvenile competition resulting in decreased productivity of wild juveniles, and through hybridization resulting in partial transfer of culture phenotypes to wild populations. However, the potential for escaped fish to affect wild populations through predation, marine competition, reproductive interference, and disease transfer pathways has been poorly studied. As well, a high degree of uncertainty exists for other escape species (e.g., marine finfish, other salmonids) due to insufficient evidence and uncertainty regarding extrapolation of existing information from other species and ecosystems. For shellfish, information from outside of Canada suggests that release of farmed bivalves can cause ecological disruptions where they are non-native. Effects are expected to be very context-specific and can be influenced by health of the receiving environment, geography, species and strain types, climate, life-stages released, among others. Overall, there is significant potential for escaped aquaculture organisms to impact natural ecosystems in Canada and this potential can be influenced by numerous environmental and genetic factors. However, the effects escaped fish may have on overall community dynamics or ecosystem function are not yet known.

Lindberg, M., P. Rivinoja, L.O. Eriksson, and A. Alanärä. 2009. Post-release and pre-spawning behaviour of simulated escaped adult Rainbow Trout *Oncorhynchus mykiss* in Lake Övre Fryken, Sweden. Journal of Fish Biology 74(3):691-698.

Using radio telemetry, the present study simulated the escape of 48 adult Rainbow Trout *Oncorhynchus mykiss* from a net-cage fish farm in the Lake Övre Fryken, Sweden. The post-release dispersal of *O. mykiss* was fast, showed long-range dispersal behavior, low winter survival and lacked the ability to find suitable spawning habitats. Thus, the present study suggested that reproducing for the first time may be an obstacle to the establishment of escaped farmed *O. mykiss*.

Naylor, R., K. Hindar, I.A. Fleming, R. Goldburg, S. Williams, J. Volpe, F. Whoriskey, J. Eagle, D. Kelso, and M. Mangel. 2005. Fugitive salmon: assessing the risks of escaped fish from net-pen aquaculture. Bioscience 55(5):427-437.

The farming of salmon and other marine finfish in open net pens continues to increase along the world's coastlines as the aquaculture industry expands to meet human demand. Farm fish are known to escape from pens in all salmon aquaculture areas. Their escape into the wild can result in interbreeding and competition with wild salmon and can facilitate the spread of pathogens, thereby placing more pressure on already dwindling wild populations. The authors assess the ecological, genetic, and socioeconomic impacts of farm salmon escapes, using a risk-assessment framework. They show that risks of damage to wild salmon populations, ecosystems, and society are large when salmon are farmed in their native range, when large numbers of salmon are farmed relative to the size of wild populations, and when exotic pathogens are introduced. They then evaluate the policy and management options for reducing risks and discuss the implications for farming other types of marine finfish.

Naylor, R.L., J. Eagle, and W.L. Smith. 2003. Salmon aquaculture in the Pacific Northwest a global industry with local impacts. Environment: Science and Policy for Sustainable Development 45 (8):18-39.

From the docks of declining coastal villages to the desks of corporate and government offices, salmon farming has been hailed as a new hope for the world's ailing offshore fishing industry and a way to reduce pressure on severely depleted fish stocks. However, the aquaculture industry has grown so quickly that, in many areas-including the Pacific Northwest-it has outstripped the wherewithal to address its adverse ecological impacts. Will fish farms, in the end, do more harm than good? Or can they deliver on their sustainable promise?

Patterson, K. 2010. The fate of farmed Rainbow Trout (*Oncorhynchus mykiss*) released from commercial aquaculture operations in Lake Huron. Thesis, University of Manitoba, Winnipeg, 193 p.

The fate of farmed fish after escape is poorly understood. The extent to which these fish might impact freshwater ecosystems is dependent upon their survival and distribution in the wild. She simulated small- and large-scale escape events from two commercial aquaculture operations in Lake Huron over 2 years. She combined the use of telemetry (120) and Floy (1000) tags to determine the fate of escaped farmed Rainbow Trout (*Oncorhynchus mykiss*). Once released, escapees dispersed rapidly, showed low site fidelity (~15% after 3 months) and were capable of long-distance movements (up to 360 km). Rainbow Trout experienced low survival (~50%) but maintained high growth rates both at and away from the farms. The results of this study provide a strong basis for understanding the potential risks that farmed fish may pose to the Lake Huron fish community and ecosystem in an escape event.

Rikardsen, A.H., and S. Sandring. 2006. Diet and size-selective feeding by escaped hatchery Rainbow Trout *Oncorhynchus mykiss* (Walbaum). ICES Journal of Marine Science 63:460– 465.

Escaped hatchery rainbow trout at post-smolt (120–340 g) and adult stages (800–3400 g) adapted differently to natural marine prey after escaping from two fish farms in northern Norway. About 1 month after escape (July), more than 57% of the post-smolt fed actively on fish larvae, which contributed 63–75% of the diet by weight. Surface insects were consumed by more than half the post-smolts and represented 24–48% of the diet during the 3-month period of sampling (June–August). One month after escaping, forage ratios (weight stomach/weight fish × 100) exceeded 1, similar to ratios recorded for other wild anadromous salmonid species in the area. Post-smolt weight increased during the sampling period and the condition factor was stable. In contrast, the condition factor of escaped adult fish reduced significantly and the forage ratios were consistently low (0.05–0.77) during the 15 months of sampling (March–August) following their escapement. These fish fed primarily on a variety of different indigestible items (especially particles of seaweed and small pieces of wood) that contributed about 70% of the stomach content weight. The authors took fish larvae only in July. Although generally contributing little to their overall diet, marine prey of great variety was consumed by the adult fish. The results indicate that young domestic rainbow trout more easily adjust to natural feeding after escape than the older, larger fish, which often fed on indigestible items similar in shape to the commercial pellets to which they were accustomed.

Rust, M.B., K.H. Amos, A.L. Bagwill, W.W. Dickhoff, L.M. Juarez, C.S. Price, J.A. Morris Jr, and M.C. Rubino. 2014. Environmental performance of marine net-pen aquaculture in the United States. Fisheries 39(11):508-524.

Efforts to reduce escapes in salmon farming in Washington State and British Columbia, Canada, have been successful. From 1987 to 1996, the average annual escape rate was 3.7% of annual harvest, whereas more recently (2000–2009) escape rate averaged 0.3%. The primary concern of escaped fish is the potential for them to interbreed with wild conspecifics and reduce the long-term fitness of the wild population.

Shallenberger, E., C.C. Tribes, D.C. Richards, J.E.J. Rensel, Z. Siegrist, F.J. O'Brien, and D. Kiefer. 2011. Rufus Woods Lake–Columbia River Reservoir Morphometrics, Initial Food Web and Rainbow Trout Fishery Studies. Available online: http://citeseerx.ist.psu.edu/viewdoc/download?doi=10.1.1.470.8385&rep=rep1&type=pdf

Commercial net pens are used in Rufus Woods Lake (RWL) within Colville Confederated Tribal jurisdiction to rear and intentionally release some sterile Rainbow Trout (RBT). Most of the fish caught in the RWL fishery are from net pen origin. Relatively large size RBT released by the CCT into RWL are purchased from the net pen growers. RBT are released at different intervals throughout the year for fishing by CCT members and non-treaty anglers. Their study incorporates information from companion studies conducted by the CCT Fish and Wildlife Department that involved collection of fish stomachs for analyses of contents by their team.

A total of 409 fish stomach samples were collected during creel surveys and from a gillnet study. Organisms in stomachs were identified to lowest practical taxon along with documentation of remnants of fish pellets and other contents. We calculated several summary statistics and graphically analyzed the stomach content data, again focusing primarily on RBT stomach contents. A large proportion (61%) of the total collected were less than 1.6 kg, the mean size of releases. Most of the acoustically tagged net pen RBT that were later recovered lost weight (about 5% loss) but this was for an average period of only 17 days. Given that most acoustically tagged fish were tracked and present for a short time (a few days to several months) and the mean residence time of acoustic tagged fish recovered was only a few weeks, as well as the fact that the mean weight loss of the acoustically tagged fish recovered averaged 5%, we believe that some of these smaller fish may have been RWL or Lake Roosevelt net pen escaped fish.

Diets of the 409 fish examined varied significantly among fish within and between time periods. At least 96 separate prey taxa were found in the stomach samples. Twenty-five percent of the RBT stomachs (N = 73) were empty but there was significant variation of mean percent empty among sampling periods. Aquatic based food items made up more of the RBT diet than did terrestrial food items. The vast majority of individual organisms in RBT stomachs were very small pelagic crustaceans (e.g. daphnia, copepods, ostracods, etc.) followed by diptera (midges and flies), snails, and terrestrial arthropods (insects and spiders).

From 14 to 17% of the RBT stomachs examined contained crayfish (9%) or fish (5-8%). However, most of the crayfish and fish occurred in only a few trout stomachs. This could indicate that few RBT had acquired the skill or ability to feed on this often abundant food source in RWL, particularly in light of the fact that most of the RBT were large fish (> 30 to 40 cm).

Skilbrei, O.T. 2012. The importance of escaped farmed Rainbow Trout (*Oncorhynchus mykiss*) as a vector for the salmon louse (*Lepeophtheirus salmonis*) depends on the hydrological conditions in the fjord. Hydrobiologia, 686(1):287-297.

The objectives of the study were to see if escaped Rainbow Trout spread rapidly or not from fish farms, and to test whether the hydrological conditions in a fjord influence their vertical distribution and importance as vector for the salmon lice. Fifty farmed rainbow trout were tagged with acoustic transmitters including depth sensors and released from two of 11 fish farms in the fjord system. In addition, unintentionally escaped rainbow trout were recaptured for analysis of salmon lice and stomach content. Dispersal out of the fjord system was limited. Most fish stayed in the vicinity of and moved between the fish farms but fed primarily on a variety of indigestible items. They moved in the warm relatively fresh surface layer from late spring until early autumn where the risk of being infested with salmon lice was low. They swam gradually deeper and became much more infested with salmon lice as the surface layers cooled and salinity and temperature gradients became less distinct over the course of the winter. The observed post-escapement behavior may challenge the control of the spread of diseases and parasites between neighboring farms and to wild fish, but also increases opportunities for recapture.

Skilbrei, O.T., and T. Jørgensen. 2010. Recapture cultured salmon following a largescale escape experiment. Aquaculture Environ. Inter. 1:107–151.

A large-scale escape experiment using 1,031 adult Atlantic Salmon *Salmo salar* was performed in the Hardangerfjord in western Norway to study the dispersal of escaped salmon, evaluate the effect of a gill-net fishery targeting escaped salmonids and test whether surface trawling is an effective way of recapturing escaped salmon in a large fjord system. The salmon of mean weights 1.56 and 5.5 kg were released from 2 commercial fish farms in late September 2006. All fish were tagged with external tags, and 48 were also equipped with acoustic transmitters. A surface pair-trawl (50 m wide and 8 m deep) was constructed to optimize catchability and maneuverability in the fjord environment. Trawling was unsuccessful, and caught only 6 simulated escapees. Telemetry data confirmed that the fish were available along the towing tracks, and they assume that towing speed and/or trawl size may have been suboptimal with regard to avoidance by fish in the fjord environment. Gill-netting proved to be an efficient method of recapture. The total reported recapture rate (of 114 fishers) was 40%, but a significantly higher recapture rate (67%) of the more highly rewarded acoustic transmitters, and the distribution of the fish in time and space, suggest that the actual catch may have been substantially higher. Approximately 90% of the catches were taken within 40 km of the release sites over the course of 4 wk. The authors conclude that a significant proportion of escaped adult salmon can be recaptured if the catch effort within the fjord basin is widespread and lasts for at least 4 wk.

Thorstad, E. B., I. A. Fleming, P. McGinnity, D. Soto, V. Wennevik, and F. Whoriskey. 2008. Incidence and impacts of escaped farmed Atlantic Salmon *Salmo salar* in nature. Report from Technical Working Group on Escapes of the Salmon Aquaculture Dialogue, 112 p.

Since the mid-1960s, Atlantic salmon Salmo salar farming has grown into a large industry beyond the native range of the species. This report examines and evaluates i) the incidence and impacts of escaped farmed salmon in nature, and ii) the technologies and efforts to prevent escapes and to reduce their impacts upon wild salmon and the environment.

Detailed information on salmon production, reported escapes from fish farms and monitoring of escaped farmed salmon in nature is given for each of the salmon producing countries. Escapes from fish farms occur from marine net pens in all salmon producing countries, as both repeated "trickle" losses of relatively small numbers of fish, and through large-scale episodic events. Numbers of farmed salmon escaping to the wild are large relative to the abundance of their wild conspecifics. Negative effects by escaped farmed salmon on wild Atlantic salmon populations have been scientifically documented, including both ecological interactions and genetic impacts of inter-breeding. It has been shown that inter-breeding of farm with wild salmon can result in reduced lifetime success, lowered individual fitness, and decreases in production over at least two generations.

The Atlantic salmon is a poor colonizer outside its native range. The probability that escaped Atlantic salmon will establish populations where the species is exotic seems low, but cannot be ruled out. It is difficult to predict if or how Atlantic salmon will adapt to the regions where they are exotic. The most important management issue at present is the need to reduce the numbers of escaped farmed salmon in nature. Among technologies and efforts to reduce impacts of escapes, sterilization and farm exclusion zones look to be among the most promising.

Tlusty, M. F., J. Andrew, K. Baldwin, and T. M. Bradley. 2008. Acoustic conditioning for recall/recapture of escaped Atlantic salmon and rainbow trout. Aquaculture 274:57–64.

Escape of salmon from sea cages is a problem that continues to plague the aquaculture industry. Data collected during the past 15 years from Norway, Scotland, Ireland, Canada and U.S. suggest significant impacts on natural runs of fish and economic losses to producers. The present report investigated the feasibility of using acoustic conditioning as a means of recalling/recapturing escaped fish. Atlantic Salmon (*Salmo salar*) and Rainbow Trout (*Oncorhynchus mykiss*) were found to respond to frequencies in the range of 50 – 400 Hz equally well. Subsequently, both species were conditioned to a 250 Hz acoustic tone during feeding. Juvenile and sub-adult fish readily conditioned to the acoustic signal within four days, with the maximum number of fish responding (85% salmon, 96% trout) by day seven. To assess retention of conditioning, fish were exposed to a single tone without feed reinforcement every one, two or four weeks.

Salmon and trout continued to respond for a seven-month period with no significant decrease (88% salmon, 97% trout) in response. No significant differences were observed in the response of *either* species to tones differing in frequency by up to 200 Hz (89% salmon, 96 trout) and intensity by 20 dB (91% salmon, 96% trout). Both species were reproducibly recalled to a cage or feeding ring in a 3.7 m tank, but were reluctant to re-enter the cage. The findings indicate that salmon and trout are readily conditioned to acoustic signals and retain that conditioning for an extended period of time without reinforcement. These characteristics suggest that acoustic conditioning has potential as a means to recall escaped salmon and when coupled with recapture, can reduce interactions with wild stocks and losses to the producer.

Veinott, G., and R. Porter. 2013. Discriminating Rainbow Trout sources using freshwater and marine otolith growth chemistry. North American Journal of Aquaculture 75(1):7-17.

Rainbow Trout *Oncorhynchus mykiss* are nonindigenous to Newfoundland. Subsequent to the development of marine cage rearing of Rainbow Trout in the Atlantic provinces in the early 1970s. Rainbow Trout have been captured in 33 rivers on the west and south coast of Newfoundland. These escapees may have negative impacts on wild populations, particularly Atlantic Salmon *Salmo salar* and Brook Trout *Salvelinus fontinalis*. In this study, the chemical fingerprints in the freshwater and marine growth sections of otoliths were used to distinguish three groups of Rainbow Trout of known origins: two hatcheries and one wild population. The results were then used to assign fish of unknown origin to the three known-origin groups and thus estimate the proportion of escapees. The three known sources produced distinct chemical fingerprints in the freshwater growth of the otoliths (cross validation test, average accuracy of over 93%); whereas, the marine growth in the otoliths produced a single chemical fingerprint for the two hatchery-origin groups distinct from the wild population. Results indicated that at least 60% of the unknown-origin fish were aquaculture escapees. Vaterite was encountered in 70–80% of the known hatchery-origin fish, 0% in the wild population, and 50% in the escapees. It appears that escapees with vaterite had a lower survival rate. The presence–absence of vaterite did not appear to be useful in distinguishing escapees from a wild population.

Volpe, J.P., E.B. Taylor, D.W. Rimmer, and B.W. Glickman. 2000. Evidence of natural reproduction of aquaculture-escaped Atlantic salmon in a coastal British Columbia river. Conservation Biology 14 (3):899-903.

The authors present evidence of the first successful natural spawning of Atlantic Salmon (*Salmo salar*) documented on the Pacific coast of North America. Twelve juvenile Atlantic salmon composed of two, year classes were captured in the Tsitika River, British Columbia. They analyzed restriction-length polymorphisms of PCR-amplified 5S rDNA and mtDNA to confirm that these individuals were Atlantic salmon. Scale analysis strongly suggested they were the products of natural spawning by feral adults. The gut contents, size, and condition of these individuals suggest that Atlantic salmon are successfully maturing in the Tsitika River, BC. This event has raised concerns that the presence and possible establishment of feral Atlantic Salmon may further jeopardize the continued persistence of already fragile native Pacific salmonids through competition for resources and occupation of niches that are currently underutilized.

Benthic Impacts

Belle, S.M., and C.E. Nash. 2008. Better management practices for net pen aquaculture. Pages 261–330 in C.S. Tucker and J. Hargreaves, editors. Environmental best management practices for aquaculture. Blackwell Publishing, Ames, Iowa.

The BMPs for current net-pen technology have been published by national salmon producer associations as codes of practice to avoid or minimize environmental effects. North American programs have been less prescriptive regarding equipment specifications and more focused on equipment testing, preventive maintenance, and monitoring and improvement of standard operating protocols. Site characteristics—especially current patterns—will affect the efficiency of feed utilization (and therefore waste production) and the accumulation or dispersal of wastes. Adequate water velocities will disperse solid wastes, ensure good water quality, and reduce the probability that the local carrying capacity of the site is exceeded. Whether a site is depositional or erosional depends on current velocity, storm frequency and magnitude, hydrography, and local circulation patterns, among other factors. Bottom type and granulometry can be used to determine whether a site is depositional or erosional.

Fallowing on net-pen farms is a relatively new technique. Management practices that address the linkage between benthic and water-column environments are relatively new. Gear placement can significantly affect water circulation patterns on a site.

Chamberlain, J., and D. Stucchi. 2007. Simulating the effects of parameter uncertainty on waste model predictions of marine finfish aquaculture. Aquaculture 179:127–140.

Models that simulate the input and fate of waste materials from marine cage finfish farms are considered valuable tools within management strategies for predicting environmental impacts. However, the overall utility of these models may be limited because of uncertainty regarding values used to parameterize and configure simulations, and variability in coupling predicted flux and benthic impacts. This study applies the aquaculture waste model DEPOMOD (Cromey, C.J., Nickell, T.D., Black, K.D. 2002a. DEPOMOD—modelling the deposition and biological effects of waste solids from marine cage farms. Aquaculture 214, 211–239.) at a marine finfish farm in British Columbia and examines i) the effect of uncertainty in three model parameters (percent waste feed, carbon concentration of feed and fecal material) and one process (resuspension) on model outputs; ii) the relative contribution of waste feed and fecal material to the predicted carbon flux distribution; and iii) the relationships between model outputs and high resolution field survey data collected at the site. Simulation of resuspension processes resulted in predictions that were considered unrealistic as 98% of the applied material was transported out with the model domain. When resuspension processes were not simulated, the applied waste feed value was the most significant contributing factor to the predicted range in model outputs up to $\sim$ 100 m from the farm site. The waste feed component accounted for between 50% and 75% of the overall carbon deposition, dependent upon the applied carbon concentration of the particles, and potentially in excess of 80% of the predicted carbon flux at the cage edge. The effect of uncertainty in the applied carbon concentrations of fecal material to the overall range in predicted flux was minimal. Coupling of model outputs with field measurements indicated that, within the predicted envelope of uncertainty, significant alterations to the benthic community structure (H`, ITI) and sediment geochemistry ($S^{=}$) indicative of the transition between oxic and anoxic benthic zonation status occurred at predicted flux values of between $\sim$ 1 and 5 gC/m^2/d.

Hargrave, B.T. 2003. A scientific review of the potential environmental effects of aquaculture in aquatic ecosystems, Volume 1. Fisheries and Oceans Canada, Canadian Technical Report of Fisheries and Aquatic Sciences 2450, Ottawa. Available online at: www.dfo-mpo. gc.ca/science/enviro/aquaculture/sok-edc/volume1/hargrave-eng.htm.

Sediment profile images (SPI) of cores collected by SCUBA diver were obtained using a modified Hargrave corer from fish farm sites in the Bay of Fundy, Canada and southeastern Tasmania, Australia. Shipboard and land-based photography were used to obtain the SPI with a tripod mounted digital camera and image analysis by commercially available software. Computer images were analyzed to determine the variables used by Nilsson and Rosenberg [Mar. Ecol., Prog. Ser. 197 (2000) 139], modified to account for non-equilibrium conditions, to assess successional stages of organic enrichment. To validate the method, they concurrently sampled macrofaunal species composition and abundance and measured profiles of redox potentials and total sulphides by ion analysis. In each case, the null hypothesis that sediments collected directly under an active salmon net-pen were indistinguishable from a nearby reference site was rejected. The SPI method can successfully detect organic enrichment where impacts occur in soft sediments in geographically diverse locations.

Hargrave, B., M. Holmer, and C. Newcombe. 2008. Towards a classification of organic enrichment in marine sediments based on biogeochemical indicators. Marine Pollution Bulletin 56:810–824.

A nomogram is developed to show that pH, redox potentials (Eh_{NHE}) and measures of dissolved sulfides ($H_2S + HS^- + S^{2-}$)(total free S^{2-}) can be used to classify organic enrichment impacts in marine sediments. The biogeochemical cycle of sulfur in marine sediments is described to show that changes in macrobenthic infauna community structure associated with high levels of organic matter supply result from stress due to oxygen deficiency (hypoxia and anoxia) and toxic effects of S^{2-}. The changes reflect enhancement of microbial sulfate reduction under conditions of high organic matter sedimentation and the progressive formation of hypoxic–anoxic conditions measured by decreased Eh_{NHE} and increased concentrations of S^{2-}. The nomogram provides a basis for classification of the oxic status of marine sediments based on changes in inter-related biological and biogeochemical variables along an organic enrichment gradient.

Holmer, M., D. Wildish, and B. Hargrave. 2005. Organic enrichment from marine finfish aquaculture and effects on sediment biogeochemical processes. Pages 181–206 in B.T. Hargrave, editor. Handbook of environmental chemistry, Volume 5M, Springer Verlag, Berlin.

Organic enrichment of sediments underlying fish farms in temperate and tropical coastal zones is reviewed to identify similarities and important biogeochemical differences. Improvements in technology have allowed farms to move from depositional sites to more erosional offshore locations. However, low cost farms are still being located in sheltered areas, in particular in the tropics. Important differences in the response of sediment geochemical variables to organic enrichment are associated with finfish aquaculture located under highly diverse hydrographic and sedimentological conditions in different coastal areas. In temperate latitudes where farms are often located over soft bottom, organic enrichment increases sediment microbial activity and may alter benthic community structure. Enhanced anaerobic activity may lead to accumulation of sulfides with adverse effects on aerobic bacteria, plants and fauna due to progressive oxygen depletion. In warm temperate waters, such as the Mediterranean and tropical latitudes, many farms are located in more advective areas with coarse-grained carbonate-rich sediments. Effects of organic enrichment in these areas are less well described, but studies have also shown sulfide accumulation in sediments indicative of deteriorated benthic habitats.

Kalantzi, L., and L. Karakassis. 2006. Benthic impacts of fish farming: meta-analysis of community and geochemical data. Marine Pollution Bulletin 52:484–493

A number of 41 papers dealing with the benthic effects of fish farming were reviewed and the values of the variables studied were extracted to be used in a meta-analysis of effects. The papers used covered a wide range of farmed species, geographic regions, management practices and specific site characteristics (e.g., depth, exposure, and sediment type). Therefore, the total data-set may not be considered as biased towards a particular set of conditions as is often the case with data collected in a single study. More than 120 biological and geochemical variables were monitored, occasionally using different sampling and analytical protocols for the same variables. The rank correlation analysis between all possible pairs of variables in the data set showed a large number of significant positive or negative correlations, reflecting the response of these variables to benthic organic enrichment. The use of stepwise regression showed that most biological and geochemical variables are determined by a combination of distance from the farm with bottom depth and/or latitude. Results of stepwise regression, repeated separately for each type of sediment, showed that although the general pattern was similar among different types of sediments, the coefficients varied considerably indicating changes of the distance affected by settling particulate organic material for different sediment types. The overall conclusion is that the complicated interactions between variables and the lack of data, such as current speed, induce difficulties in setting common or uniform environmental quality standards for benthic effects of fish farming and these should take into account the existing considerable differences between geographic regions, depth zones and sediment types.

Rooney, R.C. and C.L. Podemski. 2009. Effects of an experimental Rainbow Trout (*Oncorhynchus mykiss*) farm on invertebrate community composition. Canadian Journal of Fisheries and Aquatic Sciences, 66(11):1949-1964.

The authors examined the development of changes in the zoobenthos along a transect from an experimental Rainbow Trout farm in Lake 375, Experimental Lakes Area, northwestern Ontario, Canada. After 2 months, invertebrate abundance was reduced under the fish cage (2542 ± 569 individuals/m^2) compared with samples collected 45 m away (16 137 ± 2624 individuals/m^2). Taxa richness was also depressed, but changes in biomass were variable. Reductions in abundance and richness at high organic loading levels are consistent with earlier models developed for the marine environment of responses to organic loading in marine systems. After two production cycles, the significant principal components axis explaining 76% of total variance in abundance was correlated with distance from the cage (Spearman rank correlation, $r = -0.775$, $p = 0.014$) and with chemical variables recommended for freshwater aquaculture monitoring (Pearson's correlation coefficient, $r = 0.78$, 0.76, and 0.75 with $p = 0.013$, 0.018, and 0.020 for pore-water ammonia and sediment Cu and Zn, respectively). The effects of farming were localized, dissipating within 15 m of the cage edge. Invertebrate abundance demonstrated the most potential for incorporation into monitoring schemes at new farms. At established farms, richness may be a valuable monitoring metric.

USEPA (United States Environmental Protection Agency). 2010. Update to the biological evaluation submitted April 17 and August 6, 2008, regarding EPA action on Washington's marine finfish rearing facility provision contained in the Sediment Management Standards. Prepared for National Marine Fisheries Service, Seattle, WA. 80 p.

This Endangered Species Act analysis concluded that listed fish species that occur in Washington State waters are not likely to be affected by Washington's marine finfish rearing facility provision, WAC 173-204-412. The EPA also determined that there is No Effect for ESA-listed species (fish, mammals, turtles), under NOAA jurisdiction, analyzed for EPA's approval of Washington's marine finfish rearing facility provision, WAC 173-204-412.

Wellman, S., K.A. Kidd, C.L. Podemski, P.J. Blanchfield, and M.J. Paterson. 2017. Incorporation of wastes by native species during and after an experimental aquaculture operation. Freshwater Science 36(2): 387-401.

Freshwater aquaculture increases dissolved and particulate nutrients near fish cages, but the degree to which they are incorporated into tissues of native animals is uncertain. At the Experimental Lakes Area in northwestern Ontario, Canada, a cage culture of Rainbow Trout was operated seasonally for 5 y, and invertebrates and fishes were collected before, during, and after culturing from the experimental lake and a reference lake to assess changes in stable isotopes of C and N in their tissues. The feed contained marine fishmeal and was higher in C and N isotope values ($\delta^{13}C$ and $\delta^{15}N$) than lake biota by ≥4‰ (all taxa) and 3‰ (all invertebrates), respectively. During the aquaculture operation, $\delta^{15}N$ of littoral and pelagic invertebrates, profundal chironomids, minnows, and Lake Trout (*Salvelinus namaycush*) increased by 2 to 5 ‰ relative to before aquaculture values. In the 1st and 2nd years after aquaculture, $\delta^{15}N$ of several invertebrate taxa and all fishes continued to increase 1 to 2 ‰/y. In contrast, during aquaculture, only minnows and trout had significant increases (up to 3 ‰) in $\delta\ ^{13}C$. In the period after aquaculture, the $\delta^{13}C$ of fishes, plankton, and profundal chironomids declined to below values measured before or during aquaculture. Isotopic analysis of native biota can be used to monitor assimilation of cage culture wastes in freshwater ecosystems.

Woodcock, S.H., T. Strohmeier, Ø. Strand, S.A. Olsen, and R.J. Bannister. 2018. Mobile epibenthic fauna consume organic waste from coastal fin-fish aquaculture. Marine environmental research 137:16-23.

Organic waste released from fin-fish aquaculture is being dispersed further as industry growth has led to the expansion of open net cages in dynamic coastal locations. Here the authors investigate the response of three mobile epibenthic invertebrates (brittle stars, urchins and brown crabs), whose natural habitats overlap with large scale coastal salmon farming. Using fatty acids and stable isotopes, they found these organisms displayed decreases in δ13C and δ15N and elevated levels of C18 fatty acids reflective of terrestrial components of fin-fish feeds. Furthermore, they found these three species consume aquaculture organic waste not only directly adjacent to the farm vicinity (0-20 m from cage edge) but up to 1 km away in the case of brittle stars and brown crabs. As aquaculture feeds shift to contain more terrestrial ingredients, the biochemistry of fauna feeding on organic waste is also being shifted, the result of these changes is currently unclear.

ESCAPED STEELHEAD IMPRINTING/HOMING

Steelhead, like other anadromous salmonids have a strong tendency to home to their natal stream as they become sexually mature. Only a very small fraction (~2%) of triploid steelhead have the potential to become sexually mature. However, even these few pen-reared steelhead are likely to have a weak tendency to select a specific stream as they will have imprinted on a freshwater source far from their rearing location with no migratory experience between the two locations.

Home stream imprinting is the physiological process by which anadromous salmonids develop the capacity to recognize the odor of their natal stream. Imprinting commonly occurs at the time the juvenile salmonids undergo smoltification, the physiological development process during which they develop the capacity to reside in salt water. In the case of the triploid steelhead smoltification will occur at the initial freshwater rearing facility prior to transport to the salt water rearing site. Any escaped steelhead will not have experience a migratory path between their natal rearing location and their saltwater rearing site. Absent this experience during their smolting phase they are likely to have a weak attraction to any specific fresh water stream, further reducing the survival of the few sexually mature individuals to potentially mate with wild or hatchery steelhead.

Abadía-Cardoso, A., E.C. Anderson, D.E. Pearse, and J. Carlos Garza. 2013. Large-scale parentage analysis reveals reproductive patterns and heritability of spawn timing in a hatchery population of steelhead (*Oncorhynchus mykiss*). Molecular Ecology 22(18):4733-4746.

Understanding life history traits is an important first step in formulating effective conservation and management strategies. The use of artificial propagation and supplementation as such a strategy can have numerous effects on the supplemented natural populations and minimizing life history divergence is crucial in minimizing these effects. The authors used single nucleotide polymorphism (SNP) genotypes for large-scale parentage analysis and pedigree reconstruction in a hatchery population of steelhead, the anadromous form of rainbow trout. Nearly complete sampling of the brood stock for several consecutive years in two hatchery programs allowed inference about multiple aspects of life history. Reconstruction of cohort age distribution revealed a strong component of fish that spawn at two years of age, in contrast to program goals and distinct from naturally spawning steelhead in the region, which raises a significant conservation concern. The first estimates of variance in family size for steelhead in this region can be used to calculate effective population size and probabilities of inbreeding, and estimation of iteroparity rate indicates that it is reduced by hatchery production. Finally, correlations between family members in the day of spawning revealed for the first time a strongly heritable component to this important life history trait in steelhead and demonstrated the potential for selection to alter life history traits rapidly in response to changes in environmental conditions. Taken together, these results demonstrate the extraordinary promise of SNP-based pedigree reconstruction for providing biological inference in high-fecundity organisms that is not easily achievable with traditional physical tags.

Bett, N.N. and S.G. Hinch. 2016. Olfactory navigation during spawning migrations: a review and introduction of the Hierarchical Navigation Hypothesis. Biological Reviews 91(3):728-759.

Migrations are characterized by periods of movement that typically rely on orientation towards directional cues. Anadromous fish undergo several different forms of oriented movement during their spawning migration and provide some of the most well-studied examples of migratory behavior. During the freshwater phase of the migration, fish locate their spawning grounds *via* olfactory cues. The authors synthesize research that explores the role of olfaction during the spawning migration of anadromous fish, most of which focuses on two families: Salmonidae (salmonids) and Petromyzontidae (lampreys). They draw attention to limitations in this research, and highlight potential areas of investigation that will help fill in current knowledge gaps. They use the information assembled from this review to formulate a new hypothesis for natal homing in salmonids. Their hypothesis posits that migrating adults rely on three types of cues in a hierarchical fashion: imprinted cues (primary), conspecific cues (secondary), and non-olfactory environmental cues (tertiary). They provide evidence from previous studies that support this hypothesis.

Bley, P.W. and J.R. Moring. 1988. Freshwater and ocean survival of Atlantic salmon and steelhead: a synopsis (No. FWS-88 (9)). Maine University at Orono.

Accurate values for survival of Atlantic Salmon (*Salmo salar*) are necessary for effective management of the species, particularly in areas with active restoration efforts. The steelhead (*Oncorhynchus mykiss*) is a species close to the Atlantic salmon, in both life history and taxonomy. Comparison of survival estimates at different life stages can be informative. The data available on survival in freshwater and saltwater is scattered among technical reports, scientific papers, and unpublished records. This report summarizes much of this material in a comparative synopsis by life stages. Though not intended to be a complete life history compendium, it presents the available information in a single report.

Bridger, C.J., R.K. Booth, R.S. McKinley, and D.A. Scruton. 2001. Site fidelity and dispersal patterns of domestic triploid steelhead trout (*Oncorhynchus mykiss* Walbaum) released to the wild. ICES Journal of Marine Science 58(2):510-516.

A combined acoustic and radio telemetry system was deployed within Bay d'Espoir. Newfoundland to determine whether cultured steelhead trout (*Oncorhynchus mykiss*) released in the vicinity of a commercial aquaculture site remain at the site (site fidelity) or disperse from it. Two sets of fish releases (summer and winter 1998) were performed to determine seasonal effects on movements in the wild. Simulated escapes in summer involved 68 fish released from the cage system and 66 fish released from a cage towed approximately 1 km away from the grow-out site. The winter releases involved three batches of 30 fish each one from the cage system and two off-site over the side of a boat (at 200 and 1,000 m distance) after transport on board with no cage towing involved. The results suggest site fidelity among steelhead released during the growing season. Fidelity was only slightly larger for on-sits releases than off-site releases. Off-site released steelhead make a rapid return to their rearing sites, suggesting homing behavior. During the winter, the movement to the overwintering release site was less directed with a higher degree of dispersal. Released steelhead eventually dispersed from the release site. and in both seasons displayed a directed movement to the hydroelectric spillway, which is also the location of the local salmonid hatchery. Implications of the results are discussed in light of the development of recapture methodologies for aquaculture salmonids.

Bridger, C.J. 2002. Movement and mitigation of domestic triploid steelhead trout (*Oncorhynchus mykiss*) escaped from aquaculture grow-out cages. Thesis, Memorial University of Newfoundland. 113 p.

Cultured fish may occur in the wild from intentional release for restocking and sea ranching purposes or aquaculture escapees from ocean grow-out facilities. Aquaculture facilities lose some individuals during the production cycle, especially when sea cages are used. In addition to economic loss incurred to the fish farmer from escapement, potential disease, ecological and genetic interactions between escapees and wild conspecifics are of concern. The author monitored escapee movement, by tracking transmitter-implanted domestic female triploid steelhead trout in the wild using sophisticated biotelemetry fixed data-logging and manual tracking techniques, in Bay d'Espoir, Newfoundland, Canada. Of the 68 triploid steelhead released on-site, in July 1998, 51 (75%) remained within a 500 m radius of the summer grow-out site 32 days after release. Similar to on-site released triploid steelhead, 17 of 66 (26%) triploid steelhead released approximately 1000 m outside of the summer grow-out site returned to the site within 4 h of release. Subsequent tracking found that an additional 26 triploid steelhead had returned to the summer grow-out two days after release, bringing the total number of off-site released triploid steelhead return to 65%.

Triploid steelhead trout released during the winter displayed lower fidelity than those released in summer. Dispersed triploid steelhead during summer were detected in the vicinity of other salmonid aquaculture sites throughout the bay. Summer and winter released triploid steelhead both displayed a directed movement upstream towards the hydroelectric spillway, also the location of the local salmonid hatchery. Results suggest escaped triploid steelhead trout may survive in the wild − moving between summer grow-out sites and the hydroelectric spillway, while feeding on excess farm feed during the summer season. A biotelemetry methodology was developed, and also described herein, to monitor and optimize potential recapture traps for the salmonid aquaculture industry. Recapturing escapees aggregating near aquaculture sites may help mitigate negative implications through removal of escapees from the wild.

Bridger, C.J., R.K. Booth, R.S. McKinley, and D.A. Scruton. 2001. Site fidelity and dispersal patterns of domestic triploid steelhead trout (*Oncorhynchus mykiss* Walbaum) released to the wild. ICES Journal of Marine Science 58(2):510-516.

A combined acoustic and radio telemetry system was deployed within Bay d'Espoir, Newfoundland, to determine whether cultured steelhead trout released in the vicinity of a commercial aquaculture site remain at the site (site fidelity) or disperse from it. Two sets of fish releases (summer and winter 1998) were performed to determine seasonal effects on movements in the wild. Simulated escapes in summer involved 68 fish released from the cage system and 66 fish released from a cage towed approximately 1 km away from the grow-out site. The winter releases involved three batches of 30 fish each, one from the cage system and two off-site over the side of a boat (at 200 and 1000 m distance) after transport on board, with no cage towing involved. The results suggest site fidelity among steelhead released during the growing season. Fidelity was only slightly greater for on-site releases than off-site releases. Off-site released steelhead make a rapid return to their rearing sites, suggesting homing behavior. During the winter, the movement to the overwintering release site was less directed with a higher degree of dispersal. Released steelhead eventually dispersed from the release site, and in both seasons displayed a directed movement to the hydroelectric spillway, which is also the location of the local salmonid hatchery. Implications of the results are discussed in light of the development of recapture methodologies for aquaculture salmonids.

Cooper, J.C. and A.T Scholz. 1976. Homing of artificially imprinted steelhead (Rainbow) trout, *Salmo Gardner*. Journal of the Fisheries Board of Canada 33(4):826-829.

Greater numbers of rainbow trout (*Oncorhynchus mykiss*) exposed to morpholine as fingerlings as compared to trout left unexposed to morpholine as fingerlings homed to a stream scented with this odor (174 vs. 16). In order to locate this stream, the imprinted fish are able to search a distance of at least 13 km. These data support the olfactory hypothesis of homing.

Goetz, F.A., E. Jeanes, M.E. Moore, and T.P. Quinn. 2015. Comparative migratory behavior and survival of wild and hatchery steelhead (*Oncorhynchus mykiss*) smolts in riverine, estuarine, and marine habitats of Puget Sound, Washington. Environmental Biology of Fishes 98(1):357-375.

Declines in the survival of steelhead (*Oncorhynchus mykiss*) populations in protected waters of Washington and British Columbia have drawn attention to the need for more information on migratory patterns and losses in river, estuary, and nearshore habitats. Accordingly, acoustic telemetry was used to quantify movements by wild and hatchery steelhead smolts released from 2006 to 2009 in the Green River, and tracked through Puget Sound, Washington. Survival varied by release group and migration segment but overall survival rates from release to the Strait of Juan de Fuca were 9.7% for wild and 3.6% for hatchery fish. These rates are low relative to similar studies on steelhead. Survival was higher for wild fish along all migration segments than hatchery-origin fish; the greatest loss for both groups coincided with the slowest travel rates as fish first entered the estuary and as they exited Puget Sound. Wild fish travelled faster than hatchery fish in the river (15.1 vs. 4.4 km/d) with the fastest travel in the lower river (41 vs. 20.2 km/d) and slowest immediately after release (3.7 vs. 2.4 km/d). The travel rates of wild and hatchery fish became progressively more similar over time: 15.4 vs. 10.6 km/d in the estuary, and 10.3 vs. 9.3 km/d in nearshore areas. Movement was primarily nocturnal in the river, nearly equal between day and night in the upper estuary, and predominately diurnal in the lower estuary and nearshore waters, with no difference between wild and hatchery fish. The migration in marine water showed an early offshore movement and a strong northward and westward orientation, and all fish exited the Strait of Juan de Fuca rather than the Strait of Georgia. The findings support research suggesting that declines in wild and hatchery steelhead populations may be caused primarily by factors in the early marine period.

Hara, T.J., S. Macdonald, R.E. Evans, T. Marui, and S. Arai. 1984. Morpholine, bile acids and skin mucus as possible chemical cues in salmonid homing: electrophysiological re-evaluation. Pages 363-378 in Mechanisms of migration in fishes, Springer, Boston, MA.

The olfactory-imprinting and pheromone hypotheses of salmon homing recognize the involvement of olfaction in the recognition of the home stream. However, physiological basis for olfactory recognition and the nature of home stream odors have not yet been established. In this paper the state of knowledge and advances in the study of chemical cues relevant to salmonid homing are reviewed, with special emphasis on 1) imprinting to morpholine, 2) skin mucus as a chemical signal, and 3) chemoreceptor responses to bile acids. Although homing of salmonids artificially imprinted to morpholine appears evident from behavioral studies, the olfactory detection of morpholine has not been adequately demonstrated. The skin mucus has been shown to be a potent olfactory stimulus for salmonids. Chemical characterization revealed that free amino acids present in the mucus were primarily responsible for olfactory stimulation; a synthetic mucus, a mixture of amino acids based on the analysis data, induced olfactory response indistinguishable from that induced by the original mucus. Electrophysiological studies showed that bile acids, especially taurine conjugates, were not only potent olfactory stimulants, but also highly-specific taste stimuli for rainbow trout. The threshold concentration for taurolithocholic acid, the most potent bile acid tested, was estimated at 10-12 M, nearly 4 log units lower than that for L-proline, the most potent taste stimulant reported for this species. In the olfactory system bile acids were as stimulatory as amino acids, with the threshold being almost 1,000 times higher than those for trout taste receptors. Because high sensitivity of the salmonid gustatory system to certain chemicals has now been demonstrated, it is no longer appropriate to consider olfaction to be the sensory modality for chemical detection only on the basis of its high sensitivity. In the light of these findings some important issues for future study are discussed.

Hasler, A.D., A.T. Scholz, and R.M. Horrall. 1978. Olfactory imprinting and homing in Salmon: recent experiments in which salmon have been artificially imprinted to a synthetic chemical verify the olfactory hypothesis for salmon homing. American Scientist 66(3):347-355.

Describes nearly 30 y of laboratory and field experiments primarily concerned with the homing migration of the Coho Salmon (*Oncorhynchus kisutch*) and the Brown Trout (*Salmo trutta*). The use of olfaction for homing would require that each stream have a characteristic and persistent odor the fish can perceive, that the fish can discriminate between odors of different streams, and that they can retain an "odor memory" between downstream migration and homing migration. The results of the studies provide conclusive evidence for olfactory imprinting.

Hasler, A.D. and A.T. Scholz. 2012. Olfactory imprinting and homing in salmon: Investigations into the mechanism of the imprinting process (Vol. 14). Springer Science & Business Media.

A review of publications and research leading to our current understanding of salmonid imprinting and homing.

Keefer, M.L., and C.C. Caudill. 2012. A review of adult salmon and steelhead straying with an emphasis on Columbia River populations. Technical Report 2012-6, College of Natural Resources, University of Idaho. 86 p.

This literature review provides an overview of available information on the many inter-related mechanisms associated with juvenile imprinting and emigration and subsequent homing and straying behaviors by returning adults. The review includes a synthesis of published straying data from the Columbia River basin, with additional comparison data from representative studies outside of the Columbia system. Topics covered in the review and data synthesis were developed in consultation with U.S. Army Corps of Engineers (USACE) biologists as part of a coordinated effort to identify critical knowledge gaps and to provide a context for prioritizing research and management needs. In the review, we identified potentially important demographic and genetic factors affecting both donor populations (populations strayed from) and recipient populations (populations receiving strays).

Snake River steelhead straying model: This review also includes results from a Snake River modeling exercise that was developed in parallel with the literature review. The model estimates the number of adult steelhead strays for donor and recipient populations across a range of adult straying rates, smolt abundance at Lower Granite Dam, transportation rate from the Snake River, and smolt-to-adult returns (SARs) for hatchery, wild, in-river, and barged populations. Model outputs indicate that transported hatchery steelhead contribute the largest number of strays in most simulations. The absolute number of strays also tended to increase with smolt abundance, as SARs increased, and as transport proportion increased. As part of the modeling exercise, the authors developed a simple numerical model to show the proportion of strays in a wild recipient population (i.e., relative abundance) in relation to donor population size, recipient population size, and donor stray rate. This model shows that strays from large donor populations can numerically overwhelm native fish in small recipient populations, even at low (~1%) stray rates.

Key findings:

- Juvenile transportation can increase adult straying.
- Hatchery rearing contributes to adult straying.
- Adult straying is often associated with juvenile olfactory imprinting.
- Transportation of juvenile salmon and steelhead is commonly associated with adult straying.
- Most strays enter sites that are geographically close to natal streams.
- Strays can have positive, negative, or neutral effects on recipient populations.

Kenaston, K.R., R.B. Lindsay, and R.K. Schroeder. 2001. Effect of acclimation on the homing and survival of hatchery winter steelhead. North American Journal of Fisheries Management, 21(4):765-773.

The authors evaluated prerelease acclimation of hatchery winter steelhead *Oncorhynchus mykiss* in Whittaker Creek, a tributary of the Siuslaw River, Oregon, as a management strategy to attract returning adults to a release site where they could be removed. The objective was to reduce the number of hatchery fish in wild steelhead spawning areas while providing hatchery steelhead for recreational fisheries. They found no significant difference in homing rate or survival between hatchery steelhead acclimated for 30 d and those trucked from the hatchery and directly released. For the 1991–1993 broods, a mean of 92% of directly released fish and 97% of acclimated fish were accounted for in Whittaker Creek. In contrast, 15% of adults from hatchery smolts released at four traditional sites in the main-stem Siuslaw River were accounted for in Whittaker Creek. The spatial distribution of the catch in recreational fisheries was similar for the direct and acclimated groups; that catch, however, was nearer Whittaker Creek than the catch from traditional releases. The study shows that acclimation of juveniles is not necessary to achieve a high rate of homing of adult hatchery steelhead to a release site. Direct tributary releases combined with an adult collection facility can be used as a management strategy to minimize effects of hatchery fish on wild stocks, yet still provide recreational harvest.

Lindberg, M., P. Rivinoja, L.O. Eriksson, and A. Alanärä. 2009. Post-release and pre-spawning behaviour of simulated escaped adult Rainbow Trout *Oncorhynchus mykiss* in Lake Övre Fryken, Sweden. Journal of Fish Biology, 74(3), pp. 691-698.

Using radio telemetry, they present study simulated the escape of 48 adult Rainbow Trout *Oncorhynchus mykiss* from a net-cage fish farm in the Lake Övre Fryken, Sweden. The post-release dispersal of *O. mykiss* was fast, showed long-range dispersal behavior, low winter survival and lacked the ability to find suitable spawning habitats. These results suggested that reproducing for the first time may be an obstacle to the establishment of escaped farmed *O. mykiss*.

Scholz, A.T., C.K. Gosse, J.C. Cooper, R.M. Horrall, A D. Hasler, R.I. Daly, and R.J. Poff. 1978. Homing of Rainbow Trout transplanted in Lake Michigan: a comparison of three procedures used for imprinting and stocking. Transactions of the American Fisheries Society 107(3):439-443.

The authors compared the homing ability of three groups of Rainbow Trout (*Oncorhynchus mykiss*) stocked in Lake Michigan by different procedures. One group of juvenile rainbow trout was imprinted to a synthetic chemical, morpholine, during the pre-smolt and smolt stages, and a second group was not imprinted. Both groups were stocked directly into Lake Michigan, 1 km north of the Little Manitowoc River. A third group of trout was retained in a pond on the Little Manitowoc River during the pre-smolt and smolt stages and then released into Lake Michigan at the same location as the other two groups. During the adult spawning migration, morpholine was metered into the Little Manitowoc River. This river and 16 other locations were monitored for returning fish. The morpholine-imprinted fish returned to the Little Manitowoc River in greater numbers and strayed less than did fish from the other two treatment groups. This result is a consequence of exposure to a unique odor cue at the critical period for imprinting.

Scholz, A.T., R.J. White, M. Muzi, and T. Smith. 1985. Uptake of radio-labelled triiodothyronine in the brain of steelhead trout (*Salmo gairdneri*) during parr-smolt transformation: implications for the mechanism of thyroid activation of olfactory imprinting. Aquaculture, 45(1-4):199-214.

Two groups of 13-month-old trout were treated with either TSH (induced smolts) or saline (parr) to examine for in vivo uptake of radio-labelled triiodothyronine ($^{125}I\text{-}T_3$) into brain nuclei. This was accomplished by injecting a tracer dose of $^{125}I\text{-}T_3$ either alone or with a 1,000-fold excess of nonradioactive T_3 and measuring the amount of radioactivity that accumulated in isolated brain nuclei in each case. Uptake of $^{125}I\text{-}T_3$ was reduced by 80–90% for both TSH and saline groups receiving nonradioactive T_3 when compared with their counterparts receiving radio-labelled T_3 only. This result is consistent with the suggestion that trout brain contains saturable nuclear receptors for T_3. Similar experiments indicate gill, integument and olfactory epithelium may also be target cells that contain T_3 receptors. In gut and muscle no difference in uptake of $^{125}I\text{-}T_3$ was seen between fish with or without nonradioactive T_3, suggesting that these tissues do not contain T_3 receptors. All of the putative target tissues contained consistently more radioactivity and all nontarget tissues consistently less radioactivity in the TSH induced smolts than in the saline pre-smolts. This result is consistent with the conjecture that an ontogenetic change occurs in receptor binding kinetics during smolt transformation; i.e., T_3 receptor concentration and/or binding affinity may have increased in the target tissues of the TSH group so that less T_3 would be available for uptake by nontarget tissues.

Skilbrei, O.T. 2012. The importance of escaped farmed Rainbow Trout (*Oncorhynchus mykiss*) as a vector for the salmon louse (*Lepeophtheirus salmonis*) depends on the hydrological conditions in the fjord. Hydrobiologia *686*(1):287-297.

The objectives of the study were to see if escaped Rainbow Trout (*Oncorhynchus mykiss*) spread rapidly or not from fish farms, and to test whether the hydrological conditions in a fjord influence their vertical distribution and importance as vector for the salmon lice (*Lepeophtheirus salmonis*). Fifty farmed rainbow trout were tagged with acoustic transmitters including depth sensors and released from two of 11 fish farms in the fjord system. In addition, unintentionally escaped rainbow trout were recaptured for analysis of salmon lice and stomach content. Dispersal out of the fjord system was limited. Most fish stayed in the vicinity of and moved between the fish farms but fed primarily on a variety of indigestible items. They moved in the warm relatively fresh surface layer from late spring until early autumn where the risk of being infested with salmon lice was low. They swam gradually deeper and became much more infested with salmon lice as the surface layers cooled and salinity and temperature gradients became less distinct over the course of the winter. The observed post-escapement behavior may challenge the control of the spread of diseases and parasites between neighboring farms and to wild fish, but also increases opportunities for recapture.

Westley, P.A., T.P. Quinn, and A.H. Dittman. 2013. Rates of straying by hatchery-produced Pacific salmon (*Oncorhynchus* spp.) and steelhead (*Oncorhynchus mykiss*) differ among species, life history types, and populations. Canadian Journal of Fisheries and Aquatic Sciences 70(5):735-746.

The authors ask whether straying differs among species, life history types, and populations of adult hatchery-produced Pacific salmon and steelhead in the Columbia River basin. Previous estimates of straying have been confounded by various factors influencing the probability of individuals returning to non-natal sites (e.g., off-station releases), whereas analyses undertaken here of nearly a quarter million coded-wire tag recoveries control for these factors. Their results revealed large and generally consistent differences in the propensity to stray among species, life history types within species, and populations. Paired releases indicated that (i) Chinook Salmon (*Oncorhynchus tshawytscha*) strayed more (mean population range 0.11–34.6%) than Coho Salmon (*Oncorhynchus kisutch*) (0.08–0.94%); (ii) ocean-type Chinook (5.2–18.6%) strayed more than stream-type Chinook (0.11–10%); and Chinook salmon (0.90–54.9%) strayed more than steelhead (0.30–2.3%). They conclude these patterns are largely the result of species-specific behavioral and endocrine factors during the juvenile life stages, but analyses also suggest that environmental factors can influence straying during the adult upstream migration.

PARASITE ISSUES

Beamish, R., J. Wade, W. Pennell, E. Gordon, S. Jones, C. Neville, K. Lange, and R. Sweeting. 2009. A large, natural infection of sea lice on Pacific salmon in the Gulf island area of British Columbia, Canada. Aquaculture 297:31-37.

High levels of sea lice generally exceeding a prevalence of 60% were found on all species of juvenile Pacific salmon and on juvenile Pacific Herring in the Gulf Islands area within the Strait of Georgia, British Columbia. Virtually all sea lice were *Caligus clemensi* and most stages were maturing or mature. There are no active fish farms in this area, indicating that this is a naturally occurring epizootic of sea lice. It is possible that the infection was associated with Pacific Herring that spawned in the area in the spring, although the linkage between the spawning Pacific Herring and the infection on Pacific salmon was not determined.

Brauner, C.J., M. Sackville, Z. Gallagher, S. Tang, L. Nendick, and A.P. Farrell. 2012. Physiological consequences of the salmon louse (*Lepeophtheirus salmonis*) on juvenile Pink Salmon. Philosophical Transactions of the Royal Society B Biological Sciences 367(1596). https://doi.org/10.1098/rstb.2011.0423

Pink Salmon, *Oncorhynchus gorbuscha*, are the most abundant wild salmon species and are thought of as an indicator of ecosystem health. The salmon louse, *Lepeophtheirus salmonis*, is endemic to pink salmon habitat but these ectoparasites have been implicated in reducing local pink salmon populations in the Broughton Archipelago, British Columbia. This allegation arose largely because juvenile pink salmon migrate past commercial open net salmon farms, which are known to incubate the salmon louse. Juvenile pink salmon are thought to be especially sensitive to this ectoparasite because they enter the sea at such a small size (approx. 0.2 g). The authors describe how 'no effect' thresholds for salmon louse sublethal impacts on juvenile pink salmon were determined using physiological principles. These data were accepted by environmental managers and are being used to minimize the impact of salmon aquaculture on wild Pink Salmon populations.

Bricknell, I.R., Dalesman, S.J., O'Shea, B., Pert, C.C., and Luntz, A.J. 2006. Effect of environmental salinity on sea lice *(Lepeophtheirus salmonis)* settlement success. Diseases of Aquatic Organisms. 71(3):201-12.

This study looked at the infectivity and survival rates of sea lice copepodids to different salinities. Results showed lower salinities resulted in a reduced survival and infectivity of *Lepeophtheirus salmonis* copepodids on potential host species. The sea louse *Lepeophtheirus salmonis* (Krøyer, 1837) (Copepoda: Caligidae) is an ectoparasite of salmonid fish. It has earlier been proposed that the free-swimming infectious copepodid stage of L. salmonis gather at river mouths to infect wild Atlantic salmon *Salmo salar* and sea trout *S. trutta* smolts during their seaward migration. This study used aquarium-based methods to investigate the survival, infective ability and behavior of *L. salmonis* copepodids exposed to short periods of low salinity levels, such as those encountered at river mouths. Survival of free-swimming copepodids was found to be severely compromised at salinity levels below 29 parts per thousand (ppt). Attachment to an *S. salar* host did not aid copepodid survival during post-infection exposure to low salinity environment, and a reduction in salinity appears to reduce the ability of copepodids to remain attached to *S. salar* smolts. Pre-infection exposure of copepodids to reduced salinity levels reduced infection of *S. salar*. Infection levels at reduced salinity were lower than predicted from the free-swimming survival experiment, suggesting that low salinity compromises the copepodids' ability to sense or respond to the presence of a host. In salinity gradients, copepodids demonstrated avoidance of salinities below 27 ppt, by both altering their swimming behavior and changing the orientation of passive sinking. Avoidance of low salinity levels may be due to their adverse effects on copepodid physiology, as suggested by the reduction in survival. Sinking rates were also faster in reduced salinity, suggesting that remaining in the water column would be more energetically demanding for the copepodids at reduced salinity. These results show that both survival and host infectivity of *L. salmonis* are severely compromised by short-term exposure to reduced salinity levels.

Godwin, S.C., L.M. Dill, M. Krkošek, M.H.H. Price, and J.D. Reynolds. 2017. Reduced growth in wild juvenile sockeye salmon *Oncorhynchus nerka* infected with sea lice. Journal of Fish Biology 91(1):41-57.

Daily growth rings were examined in the otoliths of wild juvenile Sockeye Salmon *Oncorhynchus nerka* to determine whether infection by ectoparasitic sea lice *Caligusc lemensi* and *Lepeophtheirus salmonis* was associated with reduced host body growth, an important determinant of survival. More than 98% of the sea lice proved to be *C. clemensi* and the fish that were highly infected grew more slowly than uninfected individuals. Larger fish also grew faster than smaller fish. Finally, there was evidence of an interaction between body size and infection status, indicating the potential for parasite-mediated growth divergence.

Krkošek, M., M. Lewis, and J.P. Volpe. 2005. Transmission dynamics of parasitic sea lice from farm to wild salmon. Proceedings of the Royal Society B. 272:689–693.

Marine salmon farming has been correlated with parasitic sea lice infestations and concurrent declines of wild salmonids. The authors report a quantitative analysis of how a single salmon farm altered the natural transmission dynamics of sea lice to juvenile Pacific salmon. They studied infections of sea lice (*Lepeophtheirus salmonis* and *Caligus clemensi*) on juvenile Pink Salmon (*Oncorhynchus gorbuscha*) and Chum Salmon (*Oncorhynchus keta*) as they passed an isolated salmon farm during their seaward migration down two long and narrow corridors. Their calculations suggest the infection pressure imposed by the farm was four orders of magnitude greater than ambient levels, resulting in a maximum infection pressure near the farm that was 73 times greater than ambient levels and exceeded ambient levels for 30km along the two wild salmon migration corridors. The farm-produced cohort of lice parasitizing the wild juvenile hosts reached reproductive maturity and produced a second generation of lice that re-infected the juvenile salmon. This raises the infection pressure from the farm by an additional order of magnitude, with a composite infection pressure that exceeds ambient levels for 75km of the two migration routes. Amplified sea lice infestations due to salmon farms are a potential limiting factor to wild salmonid conservation.

Krkošek, M., J.S. Ford, A. Morton, S. Lele, R. A. Myers, and M.A. Lewis. 2007. Declining wild salmon populations in relation to parasites from farm salmon. Science 318:1772–1775.

Rather than benefiting wild fish, industrial aquaculture may contribute to declines in ocean fisheries and ecosystems. Farm salmon are commonly infected with salmon lice (*Lepeophtheirus salmonis*), which are native ectoparasitic copepods. We show that recurrent louse infestations of wild juvenile Pink Salmon (*Oncorhynchus gorbuscha*), all associated with salmon farms, have depressed wild pink salmon populations and placed them on a trajectory toward rapid local extinction. The louse-induced mortality of pink salmon is commonly over 80% and exceeds previous fishing mortality. If outbreaks continue, then local extinction is certain, and a 99% collapse in pink salmon population abundance is expected in four salmon generations. These results suggest that salmon farms can cause parasite outbreaks that erode the capacity of a coastal ecosystem to support wild salmon populations.

Morton, A.B., and R. Williams. 2003. First report of a sea louse, *Lepeophtheirus salmonis*, infestation on juvenile Pink Salmon, *Oncorhynchus gorbuscha*, in nearshore habitat." The Canadian Field-Naturalist 117(4):634-641.

High infestation rates of the Sea Louse (*Lepeophtheirus salmonis*) have been reported on juvenile salmonids in Europe since 1989; however, this species has not been reported on juvenile Pacific salmonids until now. Magnitude of Sea Lice infestation was examined in 2001 on juvenile Pink Salmon (*Oncorhynchus gorbuscha*) migrating through a British Columbia archipelago. On average, the 751 juvenile Pink Salmon sampled weighed 2.25 g (± 0.039 SE), were infected with 11.3 (± 0.41 SE) Sea Lice per fish and 6.1 (± 0.24SE) Sea Lice per gram host weight. Fully 75.0% of fish were infected at loads to or higher than the lethal limit reported for much larger Sea Trout (*Salmo trutta*) post-smolts. Abundance (Kruskal-Wallis statistic = 100.95, $p<0.0001$) and intensity ($KW=70.05, p<0.0001$) of lice, and mean number of lice/ghost weight($K\text{-}W=112.23, p<0.0001$) were significantly higher in juvenile Pink Salmon in close proximity to salmon farms, than in Pink Salmon distant from salmon farms.

Morton, A., R.D. Routledge, and R. Williams. 2005. Temporal patterns of sea louse infestation on wild Pacific salmon in relation to the fallowing of Atlantic Salmon farms. North American Journal of Fisheries Management 25 (3):811-821.

The authors report on a 3-year study of the infestation rates of the sea louse, *Lepeophtheirus salmonis*, on wild juvenile Pink Salmon *Oncorhynchus gorbuscha* and Chum Salmon *O. keta* in the Broughton Archipelago, British Columbia. In 2002, the British Columbia Ministry of Agriculture, Fisheries, and Food ordered farm fallowing (i.e., the removal of farmed Atlantic Salmon *Salmo salar* from net-cages) along the presumed migration route of wild juvenile Pacific salmon in this area. The goal was to protect wild juvenile fish from sea louse infestation. We assessed the effectiveness of this decision by comparing sea louse infestation rates on wild juvenile salmon near three Atlantic Salmon farm sites prior to, during, and after fallowing. Overall, *l. salmonis* levels were significantly reduced ($P < 0.0001$) at the study sites during fallowing but returned to the original level after fallowing. The decline was age-specific. While the abundance of the earliest attached sea louse phase (the copepodid stage) declined by a factor of 42, the mean abundance of adult *L. salmonis* did not decline significantly. Changes in salinity and temperature could not account for the decline. This study provides evidence that the fallowing of Atlantic Salmon farms during spring juvenile salmon migrations can be an effective conservation and management tool for protecting wild salmon. While this correlation adds to the increasing weight of evidence linking Atlantic Salmon farms to increased parasite loads on wild salmon, greater cooperation between researchers and farmers will be necessary to isolate the causal mechanisms and provide safe seaward passage to wild juvenile salmon.

Morton, A., and R. Williams. 2006. Response of the sea louse *Lepeophtheirus salmonis* infestation levels on juvenile wild Pink, *Oncorhynchus gorbuscha*, and Chum, *O. keta*, Salmon to arrival of parasitized wild adult Pink Salmon. The Canadian Field-Naturalist 120(2):199-204.

Recent recurring infestations of Sea Lice, *Lepeophtheirus salmonis*, on juvenile Pacific salmon (*Oncorhynchus* spp.) and subsequent annual declines of these stocks have made it imperative to identify the source of Sea Lice. While several studies now identify farm salmon populations as sources of Sea Louse larvae, it is unclear to what extent wild salmonid hosts also contribute Sea Lice. The authors measured Sea Louse numbers on adult Pink Salmon (*Oncorhynchus gorbuscha*) migrating inshore. They also measured Sea Louse numbers on wild juvenile Pink and Chum Salmon (*Oncorhynchus keta*) migrating to sea before the adults returned, and as the two age cohorts mingled. Adult Pink Salmon carried an average of 9.89 (SE 0.90) gravid lice per fish and thus were capable of infecting the adjacent juveniles. Salinity and temperature remained favorable to Sea Louse reproduction throughout the study. However, all accepted measures of Sea Louse infestation failed to show significant increase on the juvenile salmon, either in overall abundance of Sea Lice or of the initial infective-stage juvenile lice, while the adult wild salmon were present in the study area. This study suggests that even during periods of peak interaction, wild adult salmon are not the primary source of the recent and unprecedented infestations of Sea Lice on juvenile Pacific Pink and Chum Salmon in the inshore waters of British Columbia.

Novales F.I., C. Gulbransen, M. Galbraith, and D. Stucchi. 2009. Monitoring and potential control of sea lice using an LED-based light trap. Canadian Journal of Fisheries and Aquatic Sciences 66(8):1371-1382.

Sea lice are ectoparasitic copepods that threaten salmon farming aquaculture and the viability of wild salmon populations. To control infestations on farmed salmon, several chemotherapeutants have been developed, but these are invasive (often causing fish stress and loss in production), costly, may induce parasite resistance over time, and their impact on the environment is a major social concern. The authors show that a light-emitting diode (LED)-based light trap can be used to monitor sea lice presence on fish and in the water. The performance of the light trap was tested in experimental tanks and in the ocean. Plankton net tows were also performed to compare catches with those from light traps. The light trap caught ~70% of salmon lice larval stages loaded onto a tank and ~24% of the adults. It also acted as a delousing agent by removing ~8% of adult salmon lice infective on Chinook Salmon (*Oncorhynchus tshawytscha*) smolts in tank experiments. In the ocean, the light trap caught 21 sea lice (10 *Lepeophtheirus salmonis* and 11 *Caligus clemensi*), comprising free-swimming and attached stages, while plankton net tows failed to capture any. They conclude that light traps constitute an effective, noninvasive, environmentally friendly method to monitor sea lice.

Orr, C. 2007. Estimated sea louse egg production from Marine Harvest Canada farmed Atlantic Salmon in the Broughton Archipelago, British Columbia, 2003–2004. North American Journal of Fisheries Management 27(1):187-97.

Recent infestations of sea lice *Lepeophtheirus salmonis* on wild juvenile Pink Salmon *Oncorhynchus gorbuscha* and subsequent declines in the number of returning adult Pink Salmon have raised concern for the health of wild fish relative to salmon farming activities in the Broughton Archipelago, British Columbia. He used available (but limited) industry data to estimate sea louse egg production from Atlantic Salmon *Salmo salar* farmed by Stolt Sea Farm (now Marine Harvest Canada, Inc., Campbell River, BC) in 2003 and 2004. The 12 active farms contained between 1 and 5 million Atlantic salmon during the 2 years and about 800,000 fewer mature salmon at the start of 2003 than in 2004. Sea louse egg production peaked during winter–spring in both years prior to the seaward migration period of the area's small and vulnerable juvenile pink salmon and chum salmon *O. keta*. Marine Harvest Canada salmon hosted over 6 million gravid sea lice that produced 1.6×10^9 eggs during 2 weeks in the winter of 2003–2004. Only half as many eggs were produced from the fewer hosts present during this period in 2003. Sea lice on farmed fish were further reduced to near zero each year through multiple uses of emamectin benzoate (Slice). Fewer farmed Atlantic Salmon and sea lice in 2003 coincided with lower abundance and prevalence of *L. salmonis* on juvenile Pink Salmon and Chum Salmon near farms. A recent agreement between industry and conservationists may help improve data quality, our understanding of the dynamics sea louse–salmon interactions, and our chances of conserving.

Poley, J.D., L.M. Braden, A.M. Messmer, O.O. Igboeli, S.K. Whyte, A. Macdonald, J. Rodriguez et al. 2018. High level efficacy of lufenuron against sea lice (*Lepeophtheirus salmonis*) linked to rapid impact on moulting processes. International Journal for Parasitology: Drugs and Drug Resistance 8(2):174-188.

Drug resistance in the salmon louse *Lepeophtheirus salmonis* is a global issue for Atlantic salmon aquaculture. Multiple resistance has been described across most available compound classes with the exception of the benzoylureas. To target this gap in effective management of *L. salmonis* and other species of sea lice (e.g. *Caligus* spp.), Elanco Animal Health is developing an in-feed treatment containing lufenuron (a benzoylurea) to be administered prior to seawater transfer of salmon smolts and to provide long-term protection of salmon against sea lice infestations. Benzoylureas disrupt chitin synthesis, formation, and deposition during all molting events. However, the mechanism(s) of action are not yet fully understood and most research completed to date has focused on insects. The exposed the first parasitic stage of *L. salmonis* to 700 ppb lufenuron for three hours and observed over 90% reduction in survival to the chalimus II life stage on the host, as compared to vehicle controls. This agrees with a follow up in vivo administration study on the host, which showed >95% reduction by the chalimus I stage. Transcriptomic responses of salmon lice exposed to lufenuron included genes related to molting, epithelial differentiation, solute transport, and general developmental processes. Global metabolite profiles also suggest that membrane stability and fluidity is impacted in treated lice. These molecular signals are likely the underpinnings of an abnormal molting process and cuticle formation observed ultra-structurally using transmission electron microscopy. Treated nauplii-staged lice exhibited multiple abnormalities in the integument, suggesting that the coordinated assembly of the epi- and procuticle is impaired. In all cases, treatment with lufenuron had rapid impacts on *L. salmonis* development. They describe multiple experiments to characterize the efficacy of lufenuron on eggs, larvae, and parasitic stages of *L. salmonis*, and provide the most comprehensive assessment of the physiological responses of a marine arthropod to a benzoylurea chemical.

Price, M.H.H. 2003. Early marine ecology of Pacific salmon: interactions with sea lice. Thesis, University of Victoria, BC. 121 p.

Pacific salmon (*Oncorhynchus* spp.) are key elements of ecological systems, and play an important role in the cultural foundation of human societies. All species of wild salmon face multiple, simultaneous threats, with habitat degradation likely playing a key role in survival. Open net-pen salmon farms can degrade important nursery marine habitat for wild juvenile salmon by disrupting natural salmonid host-parasite dynamics. The first two chapters in this thesis examine louse parasitism of wild juvenile Chum (*Oncorhynchus keta*), Pink (*O. gorbuscha*), and Sockeye Salmon (*O. nerka*) in relation to their marine migration past salmon farms.

He compares sites of low and high exposure to salmon farms, and include two areas without farms on British Columbia's central and north coasts to assess baseline infection levels. Louse prevalence and abundance were lowest and most similar to natural baseline levels at low exposure sites, and highest at high exposure sites in all farm regions. A significantly greater proportion of the lice infecting juvenile chum and pink salmon were *Lepeophtheirus salmonis* at high exposure sites. *Caligus clemensi* was the principal louse species infecting all juveniles in areas without salmon farms, and at low exposure sites within salmon farm regions; *C. clemensi* was also the dominant louse to infect juvenile sockeye that migrated past farms. Mixed-effects modelling results showed that exposure to salmon farms was the most consistent factor to explain the variation in louse infection levels, and support my hypothesis that salmon farms are a major source of sea lice on juvenile wild salmon in regions with salmon farms.

He discovered that juvenile sockeye at one particular location within the Georgia Strait hosted unusually high lice levels; this location was situated at a distance from salmon farms, but near a farm salmon processing facility. He found live sea lice, *Lepeophtheirus salmonis*, mucus, and fish tissue in effluent discharged from the processing facility. Sea lice transmitted from this source may pose a threat to wild salmon populations, and the release of potentially untreated offal, including blood water, is of considerable concern.

Roberts, L.J., J. Taylor, and C. Garcia de Leaniz. 2011. Environmental enrichment reduces maladaptive risk-taking behavior in salmon reared for conservation. Biological Conservation 144(7):1972-1979.

Hatcheries often produce bold fish that are maladapted to survive in the wild, as absence of predators and selection for fast growth tend to favor risk-taking behaviors. Not surprisingly, losses of hatchery fish through predation can be high immediately after release and this may account for the failure of many *ex-*

situ fish conservation programs. For supportive-breeding to be useful, it is essential that released fish are able to display natural behaviors. They compared the performance of juvenile Atlantic Salmon reared in environmentally-enriched tanks receiving natural prey and subjected to simulated predator attacks with fish reared under standard hatchery conditions while keeping densities constant. No differences were detected between controls and environmentally enriched fish in survival, final size or nutritional status. Yet, changes in rearing conditions had rapid and marked effects on risk-taking behavior. Environmentally enriched fish were 2.1 times less willing, and took significantly longer to leave shelter, than controls within two weeks of enrichment. Their study indicates that it is possible through environmental enrichment to modify at least one component of fishes' behavior known to have clear adaptive implications, i.e., the propensity of hatchery-reared fish to take excessive risks. *Ex-situ* conservation could therefore benefit from rearing fish in naturalized, structurally complex environments with natural prey to promote the development of more natural behaviors.

Rust, M.B., K. H. Amos, A.L. Bagwill, W.W. Dickhoff, L.M. Juarez, C.S. Price, J.A. Morris Jr, and M.C. Rubino. 2014. Environmental performance of marine net-pen aquaculture in the United States. Fisheries 39(11):508-524.

Efforts to reduce escapes in salmon farming in Washington State and British Columbia, Canada, have been successful. From 1987 to 1996, the average annual escape rate was 3.7% of annual harvest, whereas more recently (2000–2009) escape rate averaged 0.3%. The primary concern of escaped fish is the potential for them to interbreed with wild conspecifics and reduce the long-term fitness of the wild population.

Saksida, S.M., D. Morrison, and C.W. Revie. 2010. The efficacy of emamectin benzoate against infestations of sea lice, *Lepeophtheirus salmonis*, on farmed Atlantic Salmon, *Salmo salar* L., in British Columbia. Journal of Fish Diseases 33:913–917. https://doi.org/10.1111/ j.1365-2761.2010.01192.x

Sea lice are a naturally-occurring ectoparasite of wild salmon (Nagasawa 2001; Beamish, Neville, Sweeting & Ambers 2005). There is also clear evidence that these parasites are seldom a production or fish health concern on farms in British Columbia (Saksida, Constantine, Karreman and Donald 2007), in direct contrast to most other salmon-producing regions. There are significant numbers of wild salmon in the Pacific Ocean, and the presence of these large untreated populations may reduce the selection pressures that appear to be at work in regions where there are fewer wild hosts. In addition, there is evidence of a genetic difference between the Atlantic and Pacific *L. salmonis*. Health implications associated with infection by the Pacific species appear to be more benign.

Skilbrei, O.T. 2012. The importance of escaped farmed rainbow trout (*Oncorhynchus mykiss*) as a vector for the salmon louse (*Lepeophtheirus salmonis*) depends on the hydrological conditions in the fjord. Hydrobiologia, 686(1):287-297.

The objectives of the study were to see if escaped Rainbow Trout spread rapidly or not from fish farms, and to test whether the hydrological conditions in a fjord influence their vertical distribution and importance as vector for the salmon lice. Fifty farmed rainbow trout were tagged with acoustic transmitters including depth sensors and released from two of 11 fish farms in the fjord system. In addition, unintentionally escaped Rainbow Trout were recaptured for analysis of salmon lice and stomach content. Dispersal out of the fjord system was limited. Most fish stayed in the vicinity of and moved between the fish farms but fed primarily on a variety of indigestible items. They moved in the warm relatively fresh surface layer from late spring until early autumn where the risk of being infested with salmon lice was low. They swam gradually deeper and became much more infested with salmon lice as the surface layers cooled and salinity and temperature gradients became less distinct over the course of the winter. The observed post-escapement behavior may challenge the control of the spread of diseases and parasites between neighboring farms and to wild fish, but also increases opportunities for recapture.

Sutherland, B.J.G., J.M. Covello, S.E. Friend, J.D. Poley, K.W. Koczka, S.L. Purcell, T.L. MacLeod et al. 2017. Host–parasite transcriptomics during immune stimulant-enhanced rejection of salmon lice (*Lepeophtheirus salmonis*) by Atlantic Salmon (*Salmo salar*). FACETS 2(1): 477-495.

Salmon lice (*Lepeophtheirus salmonis*) are important ectoparasites of wild and farmed salmonids and cause major losses to the salmon farming industry throughout the Northern Hemisphere. With the emergence of resistance to several commonly used parasiticides, novel control strategies and integration of multiple treatment options are needed, including host immune stimulation. The authors investigate the effects of a functional feed containing a peptidoglycan and nucleotide formulation on *L salmonis* infection of Atlantic Salmon (*Salmo salar*) by characterizing lice infection levels, the expression of several host immune genes, and the parasite transcriptomic response to the immune stimulated host. Although initial infection intensities were low, the low dose (LD) immunostimulant diet reduced the total lice burden by 50% relative to controls. Immunostimulant fed hosts upregulated interleukin-1β in the skin and spleen. This gene has been implicated in successful responses of several salmonid species to salmon lice but is typically not observed in Atlantic Salmon, suggesting a favorable influence on the immune response. Lice infecting LD immune stimulated salmon overexpressed genes putatively involved in parasite immunity, including carboxylesterases, and expressed genes putatively involved in feeding (e.g., proteases). These lice response genes further improve the characterization of the transcriptome of the non-model parasite by identifying genes potentially involved in evading host immunity.

Sutherland, B.J., S.G. Jantzen, D.S. Sanderson, B F. Koop, and S.R. Jones. 2011. Differentiating size-dependent responses of juvenile Pink Salmon (*Oncorhynchus gorbuscha*) to sea lice (*Lepeophtheirus salmonis*) infections. Comparative Biochemistry and Physiology D 6:213–223. https://doi.org/10.1016/j.cbd.2011.04.001

Salmon infected with an ectoparasitic marine copepod, the salmon louse L *Epeophtheiru ssalmonis*, incur a wide variety of consequences depending upon host sensitivity. Juvenile Pink Salmon (*Oncorhynchus gorbuscha*) migrate from natal freshwater systems to the ocean at a young age relative to other Pacific salmon, and require rapid development of appropriate defenses against marine pathogens. The authors analyzed the early transcriptomic responses of naïve juvenile pink salmon of sizes 0.3 g (no scales), 0.7 g (mid-scale development) and 2.4 g (scales fully developed) six days after a low-level laboratory exposure to *l. salmonis* copepodids. All infected size groups exhibited unique transcriptional profiles. Inflammation and inhibition of cell proliferation was identified in the smallest size class (0.3 g), while increased glucose absorption and retention was identified in the middle size class (0.7 g). Tissue-remodeling genes were also up-regulated in both the 0.3 g and 0.7 g size groups. Profiles of the 2.4 g size class indicated cell-mediated immunity and possibly parasite-induced growth augmentation. Understanding a size-based threshold of resistance to *L. salmonis* is important for fisheries management. This work characterizes molecular responses reflecting the gradual development of innate immunity to *L. salmonis* between the susceptible (0.3 g) and refractory (2.4 g) Pink Salmon size classes.

Whyte, S.K., J.D. Poley, A. Mueller, C. Van Iderstine, K.E. Fitzpatrick, S.L. Purcell, B.F. Koop, S.C. Johnson, S. Wadsworth, and M.D. Fast. 2019. Avermectin treatment for *Lepeophtheirus salmonis*: Impacts on host (*Salmo salar*) and parasite immunophysiology. Aquaculture 501:488-501.

The avermectins, emamectin benzoate (EMB) and ivermectin (IVM) have been commonly used in North America over the last two decades to control the salmon louse, Lepeophtheirus salmonis, infections in farmed Atlantic Salmon. Emamectin benzoate, trade name SLICE ™, was used heavily in the Eastern Canadian industry between the years 2000–2008, due to its long-lasting protection and efficacy against all parasitic life stages. However, over reliance on this drug soon resulted in reduced sensitivity in many *L. salmonis* populations, resulting more recently in uses of higher treatment dosages and switching to the use of IVM. For these reasons, we investigated the effects of different dosages of EMB and multiple IVM treatments on baseline immunophysiological indicators, anti-viral responses and protection against subsequent salmon lice exposure in salmon smolts. Different doses of EMB or repeated treatment with IVM did not affect feeding behavior in salmon, however by the end of the second IVM treatment, some neurotoxicity was observed. A single (1×) EMB dose (50 μg/Kg) administered for 7 consecutive days had no significant effect on I abundance and development, whereas triple the dosage (150 μg/Kg) significantly reduced lice development, thereby eliminating subsequent stress responses in salmon associated with lice development to pre-adult stages. Emamectin benzoate and IVM treatment did not significantly impact expression of resting antigen presentation molecules in salmon (MH class I or II), however they did inhibit short-term (6 h) induced responses to the ISA virus. The impact of gender, as previously shown, had the greatest effect on louse transcriptomic regulation, but avermectin treatment also caused perturbations in gene expression. Transcriptome differences between lice on control and 1× EMB treated fish were larger than those observed for IVM or 3× EMB. Nearly half of the transcripts differentially expressed by IVM were also affected by one of the EMB treatments. Transcriptomic results from the louse suggest a high degree of similarity and concordance within and across studies in avermectin treatment, with gender of louse and dosage of drug significantly impacting the outcomes.

Weir, L.K. and J.W. Grant. 2005. Effects of aquaculture on wild fish populations: a synthesis of data. Environmental Reviews 13(4):145-68.

The potential adverse environmental effects of aquaculture have been the subject of considerable attention in both the media and the scientific literature. The authors synthesize the published scientific literature, primarily concerning Atlantic Salmon (*Salmo salar*), to assess the current data available regarding these potential effects. No data are available to test for the direct effects of aquaculture organisms on the demographics of wild fish populations. However, seven studies show that escaped salmon in the wild have lower fitness, as measured by survival and reproductive success, than native salmon. Thirteen other studies, encompassing 91 different traits, provide strong evidence of phenotypic differences between farmed and wild salmon, presumably because of artificial selection in the aquaculture environment. An additional 10 studies have documented significant genetic differences between farmed salmon and the wild fish with which they will interact, or potentially interact.

GENETICS, INTERBREEDING ISSUES

Benfey, T.J. 2016. Effectiveness of triploidy as a management tool for reproductive containment of farmed fish: Atlantic Salmon (*Salmo salar*) as a case study. Reviews in Aquaculture 8(3):264-82.

Atlantic Salmon dominates aquaculture production in its native North Atlantic range, raising concerns about the impacts of escaped farmed fish on wild populations. While physical confinement and operational management practices have improved steadily with the development of this industry, some escapes are inevitable. In the absence of effective measures for the rapid recapture of escaped fish, the only practical method currently available to minimize their impacts on wild populations is to ensure that they are female triploids and therefore reproductively sterile. The technology for producing all-female triploid populations of Atlantic salmon is simple and easily applied on a commercial scale, and routinely results in populations that are entirely female and >98% triploid. Aside from sterility, there are no population-wide phenotypic effects of triploidy, although triploids do tend to perform less well than diploids with respect to commercial culture characteristics and are also less likely than escaped diploids to outcompete or displace native salmon. Some uncertainties exist with respect to their disease resistance and their potential to become reservoirs for the spread of pathogens to wild populations. If the spawning potential of escaped farmed Atlantic Salmon is deemed to pose an unacceptable risk to native populations, then all-female triploid populations could be used as an alternative to reduce risk. Research should continue to focus on improving triploid performance through breeding programmes and optimization of husbandry conditions (including nutrition, environmental conditions and fish health), with the goal of making triploids an attractive option for fish farmers.

Blanchfield, P.J., L.S. Tate, and C.L. Podemski. 2009. Survival and behaviour of Rainbow Trout (*Oncorhynchus mykiss*) released from an experimental aquaculture operation." Canadian Journal of Fisheries and Aquatic Sciences 66(11):1976-1988.

The potential for farmed fish that have escaped from open-cage aquaculture operations to affect native populations will depend on their survival and behavior in the wild. The authors used standard commercial practices to rear 10 tons of Rainbow Trout (*Oncorhynchus mykiss*) in a 23 ha lake at the Experimental Lakes Area (Ontario, Canada). Each fall (2003–2005) they released farmed Rainbow Trout (escapees) into the study lake and monitored their movements using automated positioning telemetry. Rainbow Trout experienced high annual mortality (~50%), with none surviving beyond 3 years. Farmed fish had narrowly defined pelagic distributions that comprised the upper few meters of the water column, even when at the cage site. Although released rainbow trout dispersed throughout the study lake, most spent significant portions of time at the cage site, especially during normal operation when commercial feed was available. Core use areas (50% Kernel) included the farm for half of the released fish. Surviving rainbow trout showed continued reliance upon the cage site in their second year. However, wide dispersal, high growth rate, and lack of reliance on the cage site by some escaped fish warrant further research to assess potential effects of open-cage aquaculture in the water bodies where the industry occurs.

Waples, R.S., K. Hindar, and J.J. Hard. 2012 Genetic risks associated with marine aquaculture. U.S. Dept. of Commerce. NOAA Tech. Memo. NMFS-NWFSC-119, 149 p.

This technical memorandum is intended to provide managers with a better understanding of the genetic effects of marine aquaculture on natural populations, so that these factors can be more effectively incorporated into informed decisions pertaining to federal marine aquaculture policy and regulatory decisions. The paper summarizes what has been learned over the past several decades from experience in three major arenas: salmon (Salmonidae) aquaculture and hatcheries, propagation of marine species, and agriculture. The paper characterizes risks and benefits of marine aquaculture and artificial propagation programs and strategies to minimize genetic risks. Specifically for marine aquaculture those strategies involve the following: Reduce opportunities for escapes and reduce opportunities for reproduction of escapes in the natural environment. Among those strategies discussed in detail are containment, recapture of escapes, the use of sterile fish stocks, and the use of highly domesticated populations in culture. Sterility of fish stocks raised in culture has the potential to significantly reduce risks from escaped individuals along with domestication of stocks that reduces probabilities of survival to maturity and successfully reproducing.

Fish Disease Issues

Amos, K.H., and J. Thomas. 2002. Disease interactions between wild and cultured fish: Observations and lessons learned in the Pacific Northwest. Bulletin of the European Association of Fish Pathologists 22:95-102.

The authors examine the interactions of selected endemic pathogens in fish stocks in the Pacific Northwest. In particular, case histories involving infectious hematopoietic necrosis virus (IHNV), and viral hemorrhagic septicemia virus (VHSV) is discussed. Field observations and epidemiological studies indicate the natural hosts and reservoir of infection are wild fish populations, both salmonid and non-salmonid marine species. Salmon recovery and restoration initiatives which re-introduce or significantly increase the number of returning adult salmon into streams and rivers supplying water for hatcheries increase the threat of exposure to the natural pathogen reservoirs. Additionally, spawned-out salmon carcasses obtained at hatcheries are being utilized for nutrient enhancement of aquatic ecosystems. For hatcheries to continue their success as a tool for salmon enhancement and restoration, strategies to maintain/improve the pathogen status of hatchery water supplies need to be pursued.

Amos, K.H., L. Gustafson, J. Warg, J. Whaley, M. Purcell, J. Rolland, J. Winton, K. Snekvik, T. Meyers, B. Stewart, J. Kerwin, M. Blair, J. Bader, and J. Evered. 2014. U.S. response to a report of infectious salmon anemia virus in western North America. Fisheries 39:501-506.

Federal, State, and tribal fishery managers, as well as the general public and their elected representatives in the United States, were concerned when infectious salmon anemia virus (ISAV) was suspected for the first time in free-ranging Pacific Salmon collected from the coastal areas of British Columbia, Canada. This article documents how national and regional fishery managers and fish health specialists of the U.S. worked together and planned and implemented actions in response to the reported finding of ISAV in British Columbia. To date, the reports by Simon Fraser University remain unconfirmed and preliminary results from collaborative U.S. surveillance indicate that there is no evidence of ISAV in U.S. populations of free-ranging or marine-farmed salmonids on the west coast of North America.

Chen, M.F., S.M. O'Neill, A.J. Carey, R.H. Conrad, B.A. Stewart, K.R. Snekvik, G.M. Ylitalo, and P.K. Hershberger. 2018. Infection by *Nanophyetus salmincola* and toxic contaminant exposure in out-migrating steelhead from Puget Sound, Washington: Implications for Early Marine Survival. Journal of Aquatic Animal Health 30(2):103-118.

Out-migrating steelhead *Oncorhynchus mykiss* from four Puget Sound rivers and associated marine basins of Puget Sound in Washington State were examined for the parasite, *Nanophyetus salmincola* in 2014 to determine whether recent trends in reduced marine survival are associated with the presence of this pathogen. A subset of steelhead from three of these river–marine basin combinations was analyzed for the presence of persistent organic pollutants (POPs) to assess whether exposure to these contaminants is a contributing factor to their reduced marine survival. The prevalence and parasite load of *N. salmincola* were significantly higher in fish from central and southern Puget Sound than in fish from river systems in northern Puget Sound. The proportion of steelhead samples with concentrations of POPs higher than adverse effects thresholds (AETs) or concentrations known to cause adverse effects was also greater in fish from the central and southern regions of Puget Sound than in those from the northern region. Polybrominated diphenyl ether concentrations associated with increased disease susceptibility were observed in 10 and 40% of the steelhead sampled from central and southern Puget Sound regions, respectively, but in none of the fish sampled from the northern region. The AET for polychlorinated biphenyls was exceeded in steelhead collected from marine habitats: 25% of the samples from the marine basins in the central and southern regions of Puget Sound and 17% of samples from northern Puget Sound region. Both *N. salmincola* and POP levels suggest there are adverse health effects on out-migrating steelhead from one southern and one central Puget Sound river that have lower early marine survival than those from a river system in northern Puget Sound.

Nash, C.E., P.R. Burbridge, and J.K. Volkman. 2005. Guidelines for the ecological risk assessment of marine fish aquaculture. National Oceanic and Atmospheric Administration, Technical Memorandum NMFS-NWFSC-71. Silver Spring, MD.

This report provides guidelines for assessing the ecological risks of marine fish aquaculture in a variety of marine ecosystems. Ten areas of substantive risk are identified in the interaction between marine fish aquaculture. The risks include: increased organic loading, increased inorganic loading, residual heavy metals, transmission of disease organisms, residual therapeutants, biological interaction of escapes with wild populations, physical interaction with marine wildlife, physical impact on marine habitat, using wild juveniles for grow-out, and harvesting industrial fisheries for aqua-feeds.

The chances of each risk occurring can differ greatly in accordance with the natural characteristics of the local ecosystem and its geographic location. Therefore, each assessment template contains a biological overview of its respective risk and briefly discusses factors that may enhance or mitigate the risk's occurrence.

Rust, M.B., K.H. Amos, A.L. Bagwill, W.W. Dickhoff, L.M. Juarez, C.S. Price, J.A. Morris Jr, and M.C. Rubino. 2014. Environmental performance of marine net-pen aquaculture in the United States. Fisheries 39(11):508-524.

Efforts to reduce escapes in salmon farming in Washington State and British Columbia, Canada, have been successful. From 1987 to 1996, the average annual escape rate was 3.7% of annual harvest, whereas more recently (2000–2009) escape rate averaged 0.3%. The primary concern of escaped fish is the potential for them to interbreed with wild conspecifics and reduce the long-term fitness of the wild population.

WILD FISH ATTRACTION

Callier, M.D., C.J. Byron, D.A. Bengtson, P.J. Cranford, S.F. Cross, U. Focken, H.M. Jansen et al. 2018. Attraction and repulsion of mobile wild organisms to finfish and shellfish aquaculture: a review. Reviews in Aquaculture 10(4):924-949.

Knowledge of aquaculture–environment interactions is essential for the development of a sustainable aquaculture industry and efficient marine spatial planning. The effects of fish and shellfish farming on sessile wild populations, particularly infauna, have been studied intensively. Mobile fauna, including crustaceans, fish, birds and marine mammals, also interact with aquaculture operations, but the interactions are more complex and these animals may be attracted to (attraction) or show an aversion to (repulsion) farm operations with various degrees of effects. This review outlines the main mechanisms and effects of attraction and repulsion of wild animals to/from marine finfish cage and bivalve aquaculture, with a focus on effects on fisheries-related species. Effects considered in this review include those related to the provision of physical structure (farm infrastructure acting as fish aggregating devices (FADs) or artificial reefs (ARs), the provision of food (e.g. farmed animals, waste feed and feces, fouling organisms associated with farm structures) and some farm activities (e.g. boating, cleaning). The reviews show that the distribution of mobile organisms associated with farming structures varies over various spatial (vertical and horizontal) and temporal scales (season, feeding time, day/night period). Attraction/repulsion mechanisms have a variety of direct and indirect effects on wild organisms at the level of individuals and populations and may have implication for the management of fisheries species and the ecosystem in the context of marine spatial planning. This review revealed considerable uncertainties regarding the long-term and ecosystem-wide consequences of these interactions. The use of modelling may help better understand consequences, but long-term studies are necessary to better elucidate effects.

Dempster, T., I. Uglem, P. Sanchez-Jerez, D. Fernandez-Jover, J.T. Bayle-Sempere, R. Nilsen, and P.A. Bjørn. 2009. Coastal salmon farms attract large and persistent aggregations of wild fish: an ecosystem effect. Marine Ecology Progress Series 385:1–14.

Coastal aquaculture is a globally expanding enterprise. Currently, 1,200 salmon farms operate in coastal Norway, yet their capacity to aggregate and subsequently modify wild fish distributions is poorly known. Aggregations of wild fish at 9 farms and 9 control locations were counted on 3 separate days in June to August 2007. On each sampling occasion, 6 counts were made at 5 distinct depth-strata at each farm and control location. Wild fish were 1 to 3 orders of magnitude more abundant at farms than at control sites, depending on the location. Gadoid fish (*Pollachius virens, Gadus morhua* and *Melanogrammus aeglefinus*) dominated farm-associated assemblages and were present across a wide range of sizes, from juveniles to large adults. Estimated total farm aggregated wild fish biomass averaged 10.2 metric tonnes (t) per farm across the 9 farms (range: 600 kg to 41.6 t). Applied across the geographical range of Norway's 1200 salmon farms, their estimates indicate that salmon farms attract and aggregate over 12000 t of wild fish into a total of just 750 ha of coastal waters on any given day in summer. Possible consequences of these persistent, substantial aggregations of wild fishes at farms include a heightened potential for the transfer of pathogens from salmon farms to wild fish and among adjacent salmon farms, and altered availability of wild fish to fisheries. Restrictions on fishing in the immediate surrounds of salmon farms may avoid farms acting as ecological traps, particularly for species with depressed populations such as *G. morhua*, which are highly attracted to farms.

Fernandez-J.D., R.I. Martinez, P. Sanchez-Jerez, J.T. Bayle-Sempere, J.A.I. Jimenez, F.J.M. Lopez, P.A. Bjørn, I. Uglem, and T. Dempster. 2011. Waste feed from coastal fish farms: a trophic subsidy with compositional side-effects for wild gadoids. Estuarine, Coastal and Shelf Science, 91(4):559-568.

Aquaculture of carnivorous fish species in sea-cages typically uses artificial feeds, with a proportion of these feeds lost to the surrounding environment. This lost resource may provide a trophic subsidy to wild fish in the vicinity of fish farms, yet the physiological consequences of the consumption of waste feed by wild fish remain unclear. In two regions in Norway with intensive aquaculture, the authors tested whether wild Sithe (*Pollachius virens*) and Atlantic Cod (*Gadus morhua*) associated with fish farms (F_{assoc}), where waste feed is readily available, had modified diets, condition and fatty acid (FA) compositions in their muscle and liver tissues compared to unassociated (UA) with farms. Stomach content analyses revealed that both cod and saithe consumed waste feed in the vicinity of farms (6–96% of their diet was composed of food pellets). This translated into elevated body and liver condition compared to fish caught distant from farms for cod at both locations and elevated body condition for Sithe at one of the locations. As a consequence of a modified diet, they detected significantly increased concentrations of terrestrial-derived fatty acids (FAs) such as linoleic (18:2ω6) and oleic (18:1ω9) acids and decreased concentrations of DHA (22:6ω3) in the muscle and/or liver of F_{assoc} cod and saithe when compared with UA fish. In addition, the ω3:ω6 ratio clearly differed between F_{assoc} and UA fish. Linear discriminant analysis (LDA) correctly classified 97% of fish into F_{assoc} or UA origin for both cod and saithe based on the FA composition of liver tissues, and 89% of cod and 86% of saithe into F_{assoc} or UA origin based on the FA composition of muscle. Thus, LDA appears a useful tool for detecting the influence of fish farms on the FA composition of wild fish. Ready availability of waste feed with high protein and fat content provides a clear trophic subsidy to wild fish in coastal waters, yet whether the accompanying side-effect of altered fatty acid compositions affects physiological performance or reproductive potential requires further research.

Sanchez-Jerez, P., D. Fernandez-Jover, I. Uglem, P. Arechavala-Lopez, T. Dempster, J.T. Bayle-Sempere, C. Valle Pérez, D. Izquierdo, P.A. Bjørn, and R. Nilsen. 2011. Coastal fish farms as fish aggregation devices (FADs). Artificial Reefs in Fishery Management. CRC Press. Taylor & Francis Group pages 187-208.

This review documents increased densities of native fishes it the locations of at least some of the numerous aquaculture facilities in various European countries.

Attachment D

Threatened and Endangered Species: 1990 Programmatic EIS Update

Attachment D to SEPA Checklist

1990 Programmatic EIS *Fish Culture in Floating Net Pens* Update:

Federally-Listed Threatened and Endangered Species and Species of Concern, and Washington State-Listed Species and Candidate Species

Cooke Aquaculture Pacific Marine Aquaculture Permit Application to Transition from Growing Atlantic Salmon to Growing All-Female Triploid Rainbow Trout/steelhead at the Cooke Existing Marine Net Pen Sites in Puget Sound, Washington

A. INTRODUCTION

The Washington Department of Fish & Wildlife (WDFW) plans to adopt the *Final Programmatic Environmental Impact Statement (PEIS): Fish Culture in Floating Net-Pens* (WDFW January 1990), or incorporate the PEIS by reference into the SEPA determination for the Cooke Aquaculture proposal to convert the species farmed at its existing Puget Sound marine net pens from Atlantic Salmon to farming an all-female triploid Rainbow Trout/steelhead (*Oncorhynchus mykiss*). The PEIS does not address threatened or endangered species that occur within the Puget Sound area, as these were listed after the PEIS was prepared. This additional information document has been prepared by the Cooke Aquaculture consultant team to address the potential effects of the species change proposal on Federally-listed threatened and endangered species, and Federal species of concern; and Washington State-listed species and candidates for listing.

PROPOSED ACTION

Cooke Aquaculture Pacific, LLC (Cooke) proposes to change the fish species being cultured at its seven (7) existing, permitted marine net pen aquaculture sites from Atlantic Salmon (*Salmo salar*) to domesticated stocks of mono-sex (all-female) sterile (triploid) Rainbow Trout/steelhead (*O. mykiss*).[1] All-female triploid Rainbow Trout/steelhead proposed to be raised in the net pen facilities is a sterile, mono-sex stock of fish that reduces the risk of genetic interference with native populations. Sterile fish stocks have also been used in numerous public agency recreational fish stocking programs for many years for this very same reason.

The existing Cooke Aquaculture Puget Sound marine net pen sites operate under the conditions of all required State agency permits for the commercial rearing of Atlantic Salmon in marine net pens. The farms have been raising almost exclusively Atlantic Salmon for the past 30 years. A recent change in Washington law (RCW 77.125.050), however, requires phasing out non-native finfish in marine net pen aquaculture. This change means that commercial marine finfish production in Washington will have to find a commercially viable and native species to farm in order stay in the business of growing seafood for human consumption. Rainbow Trout/steelhead are a native species to the Pacific Northwest region and have been commercially raised in Washington, primarily in freshwater facilities, for more than 80 years if not longer. Cooke is requesting re-approval of their Marine Finfish Aquaculture Permit (WAC 220-370-100) from the Washington Department of Fish and Wildlife (WDFW) that will allow the company to start raising domesticated stocks of all-female triploid Rainbow Trout/steelhead (*O. mykiss*) at their marine farms. Other than transitioning to the commercial cultivation of a different species of fish, the company is not planning

[1] While the common names Rainbow Trout and steelhead are often used interchangeably, this document refers to Rainbow Trout, which is the single official common name given to the species (*O. mykiss*) by the American Fisheries Society several years ago.

any alteration to the existing fish pen physical structures, site locations, supporting equipment, or general current practices, methods and cultivation techniques currently used for growing Atlantic Salmon in net pens. Domesticated stocks of all-female triploid Rainbow Trout/steelhead have very similar physiological and metabolic requirements to those of domesticated stocks of Atlantic Salmon. The basic difference is that all female, triploid (three chromosomes) Rainbow Trout/steelhead have been found to be reproductively sterile,[2] and thus convert their energy almost entirely to growth. By comparison, diploid populations of Rainbow Trout/steelhead eventually begin to sexually mature at certain age (maturation), and begin to expend energy toward the development of reproductive organs.

The company will use local stocks of all-female triploid Rainbow Trout/steelhead produced by Troutlodge hatcheries in Pierce County. Troutlodge brood stock are cultivated in Washington specifically for the production of ova to supply both private and public aquaculture operations. Troutlodge, a Washington-based company, has been producing Rainbow Trout/steelhead eggs for sale to private fish farms and public enhancement hatcheries throughout the world since 1945. Brood fish are raised in regulated pathogen-free conditions for their entire life cycle. The company utilizes a comprehensive health testing and disease-free certification program that exceeds World Organization of Animal Health (OIE) standards at their Washington facilities, allowing them to export live salmonid eggs throughout the world.

Troutlodge has been producing mono-sex (all-female) populations of Rainbow Trout/steelhead eggs since the mid-1990s. The all-female (XX only) ova are subsequently fertilized with X-only mono-milt. Triploidy is induced by mechanical pressure shock. For a short period of time, a high-pressure hydrostatic shock is applied to the newly fertilized eggs at a specified time point post-fertilization. The post-fertilization pressure treatment forces the fertilized egg to retain the third set of chromosomes that is normally ejected at this time. Pressure is then released and the triploid (3n) eggs are allowed to continue development. Ploidy is confirmed using a fluorescent nucleic acid label on either embryo or blood tissue using a flow cytometer at the Washington State University School of Veterinary Medicine. Testing results of Troutlodge triploid fish and eggs over a period of five (5) years (from 2013 to 2018) demonstrate a high rate of success in triploid induction (99.83% − 2,950 of 2,955 fish and/or eggs sampled); see SEPA Checklist Attachment A.

Cooke operates two freshwater hatcheries in the Scatter Creek area of Thurston County. The hatcheries raise and produce the juvenile fish that are eventually transferred to Cooke's Puget Sound marine net pens for final cultivation to the desired harvest size. Eyed all-female triploid Rainbow Trout/steelhead eggs would be supplied to the Cooke hatcheries from the Troutlodge hatchery under a WDFW Fin Fish Transport permit. The eggs would be hatched and cultured to a certain size in the Cooke hatcheries, and then transferred to the marine net pens after undergoing the necessary fish pathogen screening protocols, subject to review and approval by WDFW. Thereafter, a Fin Fish Transport Permit would be required from WDFW for each specified lot of fish to be transferred from Cooke hatcheries to the marine net pens.

Marine net pen cultivation and production protocols for Rainbow Trout/steelhead are basically the same as those used for Atlantic Salmon. Maximum cage density levels are expected to be managed at the same levels (approximately 0.9 to 1.2 lb/ft^3 or 15 to 20 kg/m^3), resulting in comparable maximum biomass levels that have historically been attained at each of the existing Cooke Aquaculture sites. Depending upon Rainbow Trout/steelhead size at harvest (targeted mean weights of approximately 7 to 9 lbs or 3.5 to 4.2 kg); the fish population sizes at each marine net pen site are expected to be similar to stocking levels for Atlantic Salmon. The fish feed composition for marine-reared trout diets will be the same or similar to

[2] Less than 1% (0.17%) of Troutlodge triploid fish and eggs tested over a period of five (5) years (from 2013 to 2018) were found to have the potential to still be reproductive; see discussion below and SEPA Checklist Attachment A.

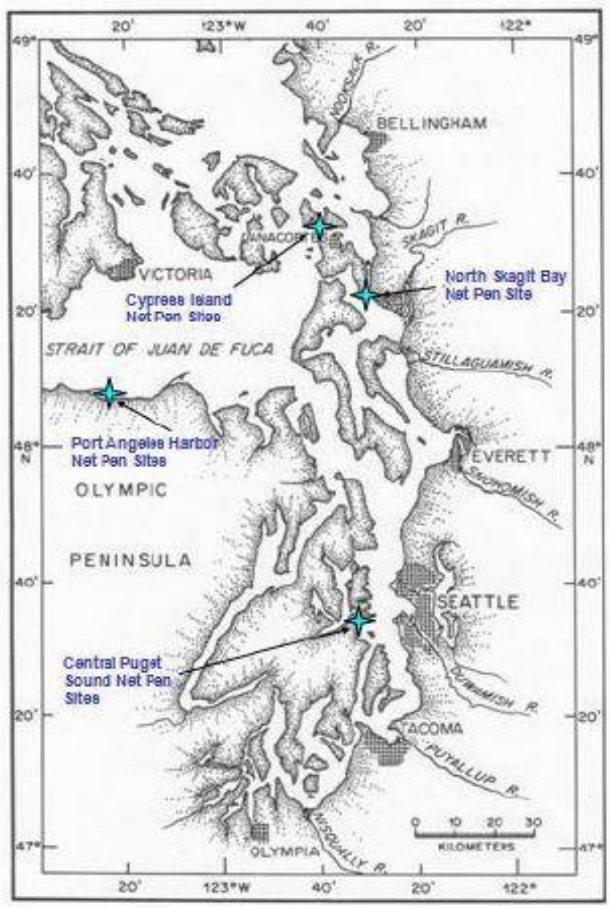
the currently-used marine salmon diets. Modern salmon and trout feeds are composed of highly digestible ingredients that are specifically formulated for optimal growth and feed conversion rates. No differences in water quality or sediment quality are expected to result from this change in species or the accompanying Rainbow Trout-specific feeds that would be used. Additional information on feed composition, expected feed conversion rates, projected growth rates, projected pen densities and production cycles is provided in SEPA Checklist Attachment B (Additional Information) in response to Information Request D.2.

All net pen support structures, stock nets, and predator exclusion nets will be the same types of equipment as that used for Atlantic Salmon production. Rainbow Trout/steelhead produced in this manner will be harvested, processed, packaged and shipped fresh to seafood customers throughout the United States.

PROJECT ACTION AREA

Existing Cooke Aquaculture marine net pen facilities with current, valid lease for State-owned aquatic lands are located at Fort Ward, Orchard Rocks, Clam Bay and Hope Island (see Vicinity Map). NPDES permits for these four facilities were recently renewed (on July 11, 2019) for these four facilities. Application materials for a modification to the NPDES permits to allow the cultivation of Rainbow Trout/steelhead in the net pens are currently being reviewed by the Washington Department of Ecology (Ecology). Cooke Aquaculture Pacific is working with the Washington Department of Natural Resources (WDNR) to resolve lease issues with existing marine net pen locations at Cypress Island (Site 1 and Site 3), and Port Angeles. If the situation is resolved, permits will also be pursued for these net pen sites to transition to native species aquaculture.

The project action area for the Cooke Aquaculture species conversion proposal corresponds to the location of the company's existing marine net pens which occur primarily within or adjacent to Central Puget Sound, North Puget Sound, the Strait of Juan de Fuca, and the Strait of Georgia. Literature indicates that potential effects of aquaculture are limited to 100 meters or less from the edge of net pens (Chamberlain and Stucchi 2007; Rooney and Podemski 2009; Noakes 2014). Minimum water depths of the existing net pens range from 35 feet to 95 feet below MLLW. Six pen sites are deeper than 55 feet, which is near the lower depths of nearshore salmon habitat (NMFS 2011). Likewise, fishes using deeper habitats (such as Pacific Cod, Pollock, and rockfishes) seldom occur within the shallower habitat where the existing net pens occupy a small amount of the available habitat.



Vicinity Map

EFFECTS ANALYSIS REQUESTED BY WDFW[3]

WDFW requested that the Federal- and State-listed species effects analysis take into consideration the extent to which changes in Puget Sound baseline light, sound, effluent (water quality), boat traffic, disease transmission, and escaped fish producing potential genetic and ecological effects (e.g., predation and competition) may affect listed species. The effects analysis for each listed species described in this Attachment D addresses these issues to the extent that they are applicable, and includes subsections specific to the species conversion proposal (see Attachment D, Section F below).

WDFW renewed the Cooke Aquaculture Marine Finfish Aquaculture permit to continue raising Atlantic Salmon *(S. salar)* for all existing net pen facilities that have a valid State-owned Aquatic Land lease (through the remaining term of those leases), and Ecology reissued the Section 402 NPDES permits for these sites on July 11, 2019. The SEPA Checklist prepared for this proposed action describes little to no change in the effect of the species conversion proposal overall on Puget Sound light (SEPA Checklist Section B.11), sound (SEPA Checklist Section B.7.b), water quality (SEPA Checklist Section B.3.a.6), or vessel traffic (SEPA Checklist Section B.14.f) because it will utilize existing marine net pens that have

3 Additional information request sent to Cooke Aquaculture by 5/21/19 e-mail communication from Ken Warheit, Ph.D., Supervisor, Washington Department of Fish & Wildlife Fish Health Section.

been installed and operational since the mid-1970s and mid-1980s.[4] No new facility siting or expansion is proposed. Consistent with the SEPA Guidelines, no global updates (1990 to 2019) to the 1990 Programmatic EIS have been prepared for light, sound, water quality, or vessel traffic since "*Elements of the environment that are not significantly affected need not be discussed"* (WAC 197-11-440(6)(a).

WDFW information requests regarding disease transmission, and regarding the potential for escaped Rainbow Trout/steelhead to cause genetic or ecological effects such as predation and competition, are thoroughly discussed in the Additional Information document (Attachment B) submitted with the July 2019 revised SEPA Checklist.

C. REGULATORY UPDATE SINCE 1990

Operational permits, approvals, and authorizations are required from multiple local, State, and Federal agencies for commercial finfish aquaculture. These permits and approvals are designed to protect ESA-listed species and habitats; protect historic and ongoing shoreline uses; assure compliance with Washington State sediment and water quality standards; regulate aquatic animal cultivation; and implement food safety regulations. This section describes current regulations involved with finfish aquaculture in Washington State, and provides a brief overview of the current requirements of these permits and approvals.

Local Jurisdiction Shoreline Substantial Development Permit (SSDP) or Shoreline Conditional Use Permit (SCUP). Washington State statute considers Aquaculture a water-dependent and preferred use of the shoreline environment and of statewide interest: *"This activity (Aquaculture) is of statewide interest. Properly managed, it can result in long-term over short-term benefit and can protect the resources and ecology of the shoreline. Aquaculture is dependent on the use of the water area and, when consistent with control of pollution and prevention of damage to the environment, is a preferred use of the water area"* ([RCW 90.58.020, WAC 173-26-201[2][d] and WAC 173-26-241[3][b][i][A]). Cities and Counties have jurisdiction over certain types of water-dependent uses within their boundaries. In addition, State-owned aquatic lands also fall under multiple State and Federal agency controls, authorities and regulations (described below). Marine aquaculture is unique and unlike upland land use permitting in many ways. Shoreline permits are issued under the regulations of the local jurisdiction Shoreline Master Program. Conditions are imposed through a SSDP or SCUP for construction and operation of new facilities. Shoreline Management Act (SMA) regulations provide for reasonable use of State shorelines and aquatic lands that facilitate public commerce while taking into consideration Statewide interests. The local jurisdiction Shoreline permitting process also requires compliance with Critical Area regulations, including protection standards for fish and wildlife habitat conservation areas and preparation of site-specific technical reports, and addresses land use issues such as light and glare, noise, view impacts, and compatibility with existing uses. Since there will be no new construction with the Cooke Aquaculture species change proposal, no Shoreline permits will be required.

State Environmental Policy Act (SEPA) Review and Determination. Cooke Aquaculture has prepared and submitted to WDFW an Environmental Checklist consistent with the SEPA Guidelines (WAC 197-11-315). The Checklist with Additional Information attachments constitutes an "expanded" SEPA Checklist. While not specifically defined in the State Guidelines, an expanded Checklist is accompanied by technical studies and/or additional information to address issues identified by the SEPA lead agency in

[4] Clam Bay approximately 1977; Fort Ward approximately 1975; Orchard Rocks 1977; Hope Island approximately 1985; Ediz Hook approximately 1985; Cypress Island Site 1 approximately 1985; Cypress Island Site 3 1986.

more detail, and to formulate proposed mitigation measures. After reviewing the SEPA Checklist and supporting documentation, WDFW will issue its SEPA Threshold Determination consistent with WAC 197-11-330 through -350. This proposed determination will be circulated for review by the Department of Ecology (Ecology), Department of Agriculture (DOA), Department of Natural Resources (WDNR) and other agencies with jurisdiction, affected Tribes, and interested groups and individuals for a public comment period.

Joint Aquatic Resource Permit Application. New finfish aquaculture facilities are required to submit a Joint Aquatic Resources Permit Application (JARPA) to all agencies involved in issuing permits and approvals related to the use of State or Federal waters. These include State and Federal agencies and Tribal governments in addition to the local Shoreline permit jurisdiction. The JARPA process provides for inter-agency coordination to address the overall features of the proposed action and its potential effects. The JARPA application describes the proposed project in specific detail regarding the project location, proposed construction methods, timing, dimensional characteristics, facility operational plans and any proposed mitigation measures. Mitigation measures can be incorporated by each agency with jurisdiction, through conditions of the various permits. There are also public involvement opportunities associated with the JARPA process. Since there will be no new construction with the Cooke Aquaculture species change proposal, no JARPA process will be required.

U.S. Army Corps of Engineers (USACE), Rivers and Harbors Act (RHA) Section 10 Permit, ESA Consultation, and Tribal Consultation. New projects (e.g., new net pen siting and construction) require a Section 10 Rivers and Harbors Act Authorization from the U.S. Army Corps of Engineers (USACE) for work and structures placed within the navigable waters of United States. Permit authorization is dependent on a thorough evaluation of potential effects to Endangered Species Act (ESA)-listed species and critical habitats found in the project vicinity. A project-specific Biological Evaluation (BE) to address new construction evaluates potential effects on ESA-listed species and critical habitats, and identifies proposed measures to minimize effects. The National Marine Fisheries Service (NMFS) and U.S. Fish and Service (USFWS) provide ESA Section 7 consultation to the Corps regarding potential effects to listed species under their jurisdiction, and describe mitigation measures to be included in the Section 10 permit. The Corps also seeks input from local Tribal governments with respect to treaty fishing rights and natural resources within their Usual and Accustomed (U&A) harvest area. Since there will be no new construction with the Cooke Aquaculture species change proposal, no RHA Section 10 permit will be required.

Washington Department of Ecology (Ecology) − Coastal Zone Management (CZM) Compliance. The Federal *Coastal Zone Management Act* requires all projects within coastal zones of the State of Washington to be certified for compliance by Ecology before any Federal agency (e.g., USACE) issues a permit authorization. State certification ensures that Federally-permitted projects are consistent with the State Coastal Zone Management Program, which has Federal approval. This applies to all shoreline activities in or affecting 15 coastal counties in Washington. Since there will be no new construction with the Cooke Aquaculture species change proposal, no CZM compliance determination is required.

Ecology − National Pollutant Discharge Elimination System (NPDES) Permit. Since 1996, Ecology has required aquaculture operators to obtain a National Pollutant Discharge Elimination System (NPDES) Individual permit for commercial net pen facilities within the State. This Clean Water Act permit requires monitoring, reporting, operational guidelines, Best Management Practices (BMPs), and Best Available Technology (BAT) for the facility to minimize pollution. NPDES measures are in addition to the monitoring and reporting requirements of other permits.

A key component of the NPDES permit for commercial net pen facilities is definition of a Sediment Impact Zone (SIZ) and Total Organic Carbon (TOC) monitoring. Organic carbon compounds are the main discharge of nutrients from salmonid net pen farm operations including Atlantic Salmon or Rainbow Trout/steelhead. The NPDES permit requires that the organic carbon levels of the sediments sampled at the 100-ft perimeter around the net pen facility (the Sediment Impact Zone boundary) be no higher than the normal organic carbon levels found in reference sample data collected at undisturbed locations throughout Puget Sound. These standards are set for the protection of benthic organisms and the surrounding marine environment. Sampling is carried out by a third-party consultant, and the analysis is conducted by a certified, independent laboratory. If a farm is out of balance with the natural assimilative properties of bottom sediments, such that carbon enrichment is identified, active mitigation will be required at the site. These measures may include additional TOC monitoring, a possible reduction in feeding amounts, reduction in fish stocking biomass, fallowing the site to allow recovery time of the sediments, possible reorientation/relocation of the site to change the hydrodynamics of a farm, or the complete removal and closure of the site if remediation does not cure the sediment standard exceedance. TOC tests will be carried out annually each summer, and additionally during the peak biomass period as the fish population is starting to be harvested from the facility if it occurs outside of the annual summer sampling period.

NPDES Individual permits for Cooke Aquaculture existing marine net pen operations in Puget Sound are based on current (2019) Ecology regulations. Specific conditions, restrictions, and monitoring and reporting procedures are designed to protect public health and safety, and to meet water and sediment quality standards that protect the environment. NPDES Individual permits for the Cooke Aquaculture Atlantic Salmon net pen facilities at Clam Bay, Fort Ward, Orchard Rocks, and Hope Island were recently reissued (July 11, 2019), conditionally authorizing discharges associated with commercial rearing of Atlantic Salmon, subject to closing out non-native fish rearing after the Washington Department of Natural Resources (WDNR) State-owned aquatic land leases expire in 2022. The renewed permits indicate that if the company elects to raise native fish at these locations in the future, Ecology will consider a permit modification and may apply the discharge limits and requirements of the existing NPDES permits to native finfish. The reissued permits become effective on August 10, 2019 and expire on August 9, 2024.

Washington Department of Fish and Wildlife (WDFW) Aquatic Farm Permit and Registration. Cooke Aquaculture existing marine net pen operations in Puget Sound are all registered with WDFW, as required by this permit. The Aquatic Farm Permit is specific to each individual aquatic farm location, and defines the type of species being reared at the facility. WDFW Aquatic Farm registration requires annual renewal by the Department. Cooke has requested the addition of Rainbow Trout/steelhead (*O. mykiss*) to their Aquatic Farm Permits for the facilities and the Department has made this modification to the permit.

WDFW Marine Finfish Aquaculture Permit. WDFW has the authority to approve, deny or approve with conditions the Marine Finfish Aquaculture Permit (MFAP). An aquatic farmer must have a valid MFAP that approves the species, stock and race of finfish being reared at that specific location. On March 19, 2019, WDFW approved the renewal of the Marine Finfish Aquaculture Permits for rearing Atlantic Salmon *(Salmo salar)* at the Cooke Aquaculture facilities. WDFW conditioned those permits to the Cooke net pen sites that have a valid existing WDNR lease for State-owned aquatic lands. These facilities include the Hope Island site in Skagit County, and the Clam Bay, Orchard Rocks and Fort Ward net pen sites in Kitsap County. Cooke also applied for renewal of the MFAP for the Port Angeles and Cypress Island net pen sites to raise both Atlantic Salmon and Rainbow Trout/steelhead; however, these sites do not currently have a valid lease for State-owned aquatic lands. The status of the WDNR lease agreement with these two locations is pending appeal.

The Aquaculture Finfish permit requires development of a plan of operation; accidental fish release prevention, response and reporting procedures; and other components. Copies of these plans and a Plan of Operation for a Finfish Permit Application were originally sent to the Department on January 18, 2019. Cooke is requesting approval of a Marine Finfish Aquaculture Permit from WDFW to allow the company to begin growing a native species, Rainbow Trout/steelhead, at their marine net pen facilities. These will be domesticated stocks of mono-sex (all-female) sterile (triploid) Rainbow Trout/steelhead, described above under the Proposed Action.

WDFW Finfish Transport Permit. Smolt transport from the Cooke Aquaculture hatchery near Olympia to existing marine net pen operations requires a WDFW Finfish Transport Permit to authorize the movement of live fish within Washington State. WDFW has regulatory authority and is responsible for enforcing the fish health laws and disease control regulations of Washington. Private finfish aquaculture facilities are subject to the same laws and regulations as State, Federal and Tribal fish hatcheries and fish enhancement facilities. Any findings of regulated pathogens at private or public finfish aquaculture facilities must be reported to WDFW fish health authorities within 24 hours. WDFW requires fish hatcheries to perform routine disease screening of adult brood fish as well as the offspring from spawning events. WDFW issues Finfish Transport permits based on the ability to verify that the brood fish and offspring tested free of any ecologically- or economically-important fish pathogens or parasites.

Additional Ecology and WDFW Regulatory Oversight. Ecology under the NPDES permit, and WDFW under the Finfish Aquaculture and Finfish Transport permits, require that accidental fish escapement be reported to them within 24 hours of knowledge of the event. As part of these State permits, the aquaculture industry has developed Fish Escape Prevention Plans and Accidental Fish Escape Response and Reporting Plans, as well as implemented other safeguards to minimize the risks of escapement. New laws became effective on June 7, 2018 with regard to non-native marine aquaculture. Those new laws phase out the growing of non-native marine fish in marine net pens over the next several years, which means that if there is any new marine finfish aquaculture to continue in Washington, native finfish species would have to be raised. The law also requires inspections of the physical net pen structures every two years by a licensed engineering firm with reports submitted to WDNR and WDOE. The industry has worked with the various agencies since the Cypress Island net pen collapse that occurred in August 2017 to implement various programs for routine inspections of the facilities by agency personnel and routine reporting to the agencies by the industry. The farmers have a financial interest in ensuring that all fish raised in a marine net pen facility make it to the market place. With this objective, companies engaged in the aquaculture industry have invested significantly to improve rearing equipment and cultivation practices to ensure that this occurs.

Since 2002, aquatic farmers have been required to mark each fish as identifiable to the individual aquatic farmer that grows it. Otolith (ear bone) marking of farmed stocks was developed in conjunction with WDFW technicians. Thermal otolith marking is also used by public enhancement hatcheries to identify the hatchery origins of the Pacific salmon they release into the wild. When these salmon return to spawn in Washington rivers, the hatchery origin of commercially- and recreationally-caught fish can be identified by WDFW.

Per Washington regulations also passed in 2002, no genetically-modified (trans-species) fish can be raised in commercial marine net pens in Washington State (WAC 220-370-100). No transgenic fish have ever been raised by the Washington marine net pen industry.

Washington Department of Natural Resources (WDNR) Aquatic Use Authorization. As mentioned above, the Cooke Aquaculture Hope Island, Clam Bay, Fort Ward and Orchard Rocks marine net pen operations have valid existing Aquatic Use Authorization and State-owned Aquatic Land Leases issued by WDNR. The status of the leases at the Cypress Island and Port Angeles net pen sites are being appealed, and until resolved, these sites do not have a valid aquatic use permit. Washington State owns the aquatic lands, and WDNR leases aquatic lands for water-dependent uses. These leases have conditions and rules for the allowable activities within the lease boundary areas, and are written to protect natural resources while creating economic activity from a public resource. Any vacated lease must have all physical improvements completely removed from them so that the lease area is returned to prior conditions. A certainty bond is required within the terms of the lease. Cooke Aquaculture prepares and submits quarterly harvested fish production reports to WDFW and WDNR. Lease payments are based on a flat annual rate and an additional royalty amount based on the harvest production each year.

U.S. Coast Guard Private Aids to Navigation (PATON) Permit. Operators of floating structures permanently moored in the navigable waters of the U.S. must obtain a Private Aids to Navigation (PATON) permit for placement of navigational warning lights on the structure. The exact latitude and longitudinal coordinates for each light and the light specifications are registered with the U.S. Coast Guard. These are entered onto updated navigational charts. Fish farm structures are required to install two or more 6-second flashing yellow navigation lights. Cooke Aquaculture existing marine net pen operations in Puget Sound have current PATON permits.

U.S. Food and Drug Agency (USFDA). Aquaculture facilities must comply with rules and regulations pertaining to the production of food fish for human consumption. Only USFDA-approved disease control chemicals are allowed to be used. Periodic random inspections of aquaculture products are carried out by the USFDA. Fish processing plants are periodically inspected by USFDA for compliance with current food safety regulations. USFDA requires a Hazards Analysis and Critical Control Points (HACCP) plan and strict record keeping to be licensed to process and sell fish products.

Siting Standards. Ecology, in conjunction with WDNR and WDFW, prepared *Recommended Interim Guidelines for Management of Salmon Net Pen Culture in Puget Sound* (1986) when commercial salmon pens were first being developed in Puget Sound in the mid-1980s. The *Interim Guidelines* initiated environmental standards and procedures for the proper location of fish pens in Puget Sound. Among other things, the *Guidelines* require that net pen sites be located in areas that meet minimum current velocity standards. By establishing minimum velocities, the standards ensure that re-suspension, oxygenation and assimilation of organic salmon farming waste products will occur so as to minimize potential benthic impacts.

Recommendations for Managing Commercial Net Pen Aquaculture in Washington's Straits and Estuaries. While the 1986 siting standards are generally still considered sound practices for locating new net pen aquaculture facilities, Ecology wanted to update these standards with state-of-the-art knowledge and best management practices, and to write recommendations to address operational issues for which there is now a performance history in Washington since commercial net pen aquaculture began in the mid-1970s. These issues included salmon interactions, marine life and other protected species interactions, disease management, sensitive habitats, water quality, feeds, predator control, antifoulants, transfer and transportation, escapes, marine debris, and emerging issues: climate change, ocean acidification, cumulative impacts, cultivation of non-salmonid finfish, and integrated multi-trophic aquaculture. A Planning Team was assembled in 2015, and a management recommendations outline was prepared for the guidance document.

The Washington State Legislature responded to the Cypress Island net pen collapse in August 2017 that resulted in the escapement of Atlantic Salmon to Puget Sound by passing Engrossed House Bill 2957, phasing out the remaining Atlantic Salmon net pens by 2022. The bill specifically directs Ecology WDFW, WDNR, and the Department of Agriculture (DOA) to complete the guidance effort initiated in

2015 for more State oversight of commercial net pen aquaculture. This effort receives substantial technical assistance from the NOAA National Centers for Coastal Ocean Science, the Northwest Indian Fisheries Commission, tribal governments, and academic institutions listed in the bill as project partners. The guidance project is designed to deliver recommendations for future consideration by State managers, and will not directly change State laws or regulations.

D. FEDERALLY-LISTED SPECIES AND SPECIES OF CONCERN

WDFW (May 21, 2019) identified the Federally-listed species to be addressed in the update of the 1990 *Programmatic EIS: Fish Culture in Floating Net Pens*, and in the SEPA analysis of the Cooke Aquaculture Puget Sound net pen species conversion proposal from rearing Atlantic Salmon to rearing Rainbow Trout/steelhead. Information provided in this section about each Federally-listed marine mammal, bird, fish and mollusc also includes the State species status; species description, distribution and habitat information; threats within the project action area; and, where applicable, a description of critical habitat. Additional State-listed species and candidate species are described in Section E. The effects of Atlantic Salmon marine net pen aquaculture, and of the Cooke Aquaculture species conversion proposal to rear all-female triploid Rainbow Trout/steelhead in the company's existing marine net pen facilities, is provided in Section F for both Federally-listed and State-listed species.

Common Name	Federal Species Status	State Species Status
Humpback Whale	Endangered	Endangered
Southern Resident Killer Whale	Endangered	Endangered
Northern Sea Otter	None[5]	Endangered
Stellar Sea Lion	None[6]	None
Bald Eagle	None[7]	None
Marbled Murrelet	Threatened	Threatened
Chinook Salmon (Puget Sound ESU)	Threatened	Candidate
Chum Salmon (Hood Canal Summer-run)	Threatened	Candidate
Chum Salmon (Puget Sound/Strait of Georgia ESU	None[8]	Candidate
Coho Salmon	Species of concern	None
Steelhead (Puget Sound ESU)	Threatened	Candidate
Bull Trout	Threatened	Candidate
Eulachon	Threatened	Candidate
Flathead Sole	Species of concern	None

5 Northern Sea Otter presently have no Federal listing status. They are protected under the Marine Mammal Protection Act (https://www.fws.gov/wafwo/articles.cfm?id=149489657).

6 Stellar Sea Lion presently have no Federal or State listing status. They are protected under the Marine Mammal Protection Act (WDFW, August 2008 [updated January 2019]: Priority habitats and species list).

7 Bald Eagle presently have no Federal or State listing status. They are protected under the Bald and Golden Eagle Protection Act (WDFW, August 2008 [updated January 2019]: Priority habitats and species list).

8 WDFW, August 2008 (updated January 2019).

Common Name	Federal Species Status	State Species Status
Green Sturgeon	Threatened	None
Pacific Cod	Species of concern	Candidate
Pacific Hake	Species of concern	Candidate
Pacific Lamprey	None[9]	None
River Lamprey	Species of concern	Candidate
Canary Rockfish	None[10]	Candidate
Bocaccio Rockfish	Endangered	Candidate
Yelloweye Rockfish	Threatened	Candidate
Pinto Abalone	Species of concern	Candidate

HUMPBACK WHALE (*Megaptera novaeangliae*)[11,12]

Status and Description

Humpback Whale were listed as endangered throughout their entire range under the Endangered Species Act on June 2, 1970 (35 FR 8491). They are also State-listed as endangered.[13] The Central North Pacific population has increased in abundance between the early 1980s and early 1990s; but the status of this population relative to its optimum sustainable population size is unknown (NOAA Fisheries. 2019a).

Humpback Whale are often identified by their large size, songs that can be heard many miles away, and their ability to breach in the water in spite of their large body size. Adults can grow to an average length of 40 to 60 feet and weigh as much as 44 tons.

Humpback Whale are filter feeders; they have no teeth. Their baleen plates have bristles that capture various small prey, and separate water and debris from their prey. Their only known natural predator is a pack of killer whales, though successful attacks are believed to be rare.

They hunt and feed during the summer months in cold waters, and migrate toward warmer tropical areas during the winter months to mate and bear offspring. They are known to travel as far as 16,000 miles. They are generally very solitary and non-social creatures that prefer traveling alone or in small groups of two to three, excepting when hunting or mating when they may be observed in large groups. Female Humpback Whale will bear a single offspring once every 2 to 3 years with the average gestation period lasting 11 to 12 months. This gestation period allows the female whale to return to warmer waters for birthing, where the calf can mature and prepare for the long migration back to the feeding grounds.

Distribution and Habitat

Surveys indicate that Humpback Whale occupy habitats around the world, with three major distinct populations: the North Atlantic, the North Pacific, and the southern oceans. These three populations do

9 WDFW, August 2008 (updated January 2019).

10 Genetic information collected by NMFS found that Canary Rockfish in the Puget Sound/Georgia Basin are not distinct from coastal Canary Rockfish, and therefore are not a Distinct Population Segment (DPS). To qualify for Federal ESA protections, a species must be discrete from the remainder of the species (NOAA Fisheries 2017).

11 USEPA 2010.

12 https://www.whalefacts.org/humpback-whale-facts/

13 WDFW 2008 (updated January 2019).

not interbreed. Humpbacks generally feed for 6 to 9 months of the year on their feeding grounds in Arctic and Antarctic waters. The animals then fast and live off their fat layer for the winter period while on the tropical breeding grounds (NOAA Fisheries 2019a). The North Pacific herd of Humpback Whale that typically occupies southeastern Alaska waters also migrates to Hawaii and Mexico in the winter months for breeding. Humpback Whale in the North Pacific are seasonal migrants feeding on zooplankton, and small schooling fish in coastal waters off the coastal waters of the western United States and Canada (NOAA Fisheries 2019a).
Humpback Whale are not expected to be routinely present in Washington waters or in the waters potentially affected by existing Puget Sound commercial net pens.

Threats within the Project Action Area
The most significant threats to the survival of Humpback Whale include entanglements in fishing gear, collisions with ship traffic, and pollution of their coastal habitat from human settlements (NOAA Fisheries 2019a). Noise pollution (e.g., sonar) is also a growing concern.

SOUTHERN RESIDENT KILLER WHALE (*Orcinus orca*)[14,15]

Status and Description
The distinct population segment (DPS) of Southern Resident Killer Whale (SRKW) was Federally-listed as endangered on November 18, 2005 (70 FR 69903). Prior to the ESA listing, NMFS determined that the SRKW stock was below its optimum sustainable population and designated it as depleted under the Marine mammal Protection Act in May 2003 (68 FR 31980). SRKW are also State-listed as endangered.[16]

Southern Resident Killer Whale use echolocation during foraging and feed primarily on salmonids. Based on a study that included both Northern and Southern DPS whales, salmon were found to represent more than 96% of the prey during summer and fall. Chinook Salmon were the preferred prey species comprising 70% of the species taken despite the relatively low abundance of Chinook in these areas compared to other species. Chum Salmon were consumed extensively in the fall. Other prey species of Southern Resident Killer Whale include flatfish, lingcod, greenling, and squid.

The SRKW population has fluctuated considerably over the 30 years that it has been studied. In 1974, it comprised 71 whales, peaked at 97 animals in 1996, and then declined to 79 in 2001. The population now numbers 76 individuals.

Distribution and Habitat
Resident killer whales in U.S. waters are distributed from Alaska to California, with four distinct communities recognized: Southern, Northern, Southern Alaska, and Western Alaska. The Southern Resident DPS consists of three pods named J, K, and L. These pods reside for part of the year in the inland waterways of Washington State and British Columbia (Strait of Georgia, Strait of Juan de Fuca, and Puget Sound), principally during late spring, summer, and fall. Pods visit coastal sites off Washington and Vancouver Island. Offshore movements and distribution are largely unknown for this DPS.

Critical Habitat
The final Critical Habitat Rule for the SRKW Distinct Population Segment (DPS) was issued 11/29/06 (50 CFR Part 226). SRKW critical habitat includes approximately 2,560 square miles of Puget Sound,

14 USEPA 2010.
15 NMFS 2008.
16 WDFW 2008 (updated January 2019).

excluding areas with water less than 20 feet deep relative to extreme high water. The Primary Constituent Elements (PCEs) for SRKW critical habitat are: 1) water quality to support growth and development; 2) prey species of sufficient quantity, quality, and availability to support individual growth, reproduction and development, as well as overall population growth; and 3) passage conditions to allow for migration, resting, and foraging (NMFS 2011). Three specific areas are designated: the summer core area in Haro Strait and waters around the San Juan Islands; Puget Sound; and the Strait of Juan de Fuca, excluding 18 military sites for national security purposes (approximately 112 square miles).

Threats within the Project Action Area

The SRKW population is at risk for both incremental small-scale impacts over time (e.g. reduced fecundity or sub-adult survivorship) or to major catastrophe (e.g. oil spill or disease outbreak). The small size of this DPS makes it potentially vulnerable to allele effects (e.g., inbreeding depression) that could cause decline.

There are limited numbers of reproductive-age SRKW males and several females of reproductive age are not having calves. The factors causing the decline of SRKWs are not well known. Some of the possible causes of decline are: reduced quantity and quality of prey; persistent pollutants that could cause immune or reproductive system dysfunction; oil spills; acoustic effects and disturbance from vessels.

NORTHERN SEA OTTER (*Enhydra lutris*) [17,18,19,20]

Status and Description

The Northern Sea Otter is protected from hunting and harassment by the Marine Mammal Protection Act of 1972 but has no Federal listing under ESA. They were State-listed as endangered in 1981, due to the small population size, restricted distribution, and vulnerability of the population in Washington State.

WDFW issued their Sea Otter Recovery Plan in December 2004, the goals of which are to implement strategies that will ensure a self-sustaining Sea Otter population in Washington through the foreseeable future, and to manage the Washington Sea Otter stock in a manner that is consistent with the Marine Mammal Protection Act, other Federal and State laws, court rulings, and Federal treaties with Native American tribes.

Between 1740 and 1900, Russian and American fur traders harvested Sea Otter almost to extinction. The International Fur Seal Treaty halted commercial hunting of Sea Otter in 1911, after which their populations rebounded and they re-colonized much of their former range between Prince William Sound, Alaska west to the Kuril Islands. However, by the 1950s, they became extinct along the Pacific coast from Prince William Sound south to Baja California, with the exception of one remnant population in California. During the 1960s and 1970s, the Alaska Department of Fish and Game, in collaboration with other State and Provincial wildlife management agencies, reintroduced Sea Otter into former habitat in Alaska, Canada, Washington and Oregon. Due to these efforts, Sea Otter populations in southeast Alaska, British Columbia, and Washington are currently stable and increasing, but have not yet rebounded to pre-commercial harvest levels.

Studies have found that genetic exchange between the British Columbia and Washington Sea Otter populations is occurring, but to an unknown degree. Given a steady and substantial increase in numbers

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[17] http://www.marinemammalcenter.org/education/marine-mammal-information/sea-otter.html
[18] https://www.fws.gov/wafwo/sea_otters_history.html
[19] https://www.fws.gov/wafwo/articles.cfm?id=149489657
[20] https://www.eopugetsound.org/articles/sea-otter-enhydra-lutris

between 2015 and 2017, and this evidence of genetic exchange with the British Columbia Sea Otter population, Sea Otter are no longer *"seriously threatened with extinction throughout all or a significant portion of its range within the state,"* which is the definition of an endangered species. It has been recommended that the sea otter be reclassified to a State threatened species in Washington (Sato 2018).

Sea Otter are members of the weasel (Mustelid) family. The have very thick fur, the thickest of any mammal. Their fur consists of two layers, an undercoat and longer guard hairs. This system traps a layer of air next to their skin to keep their skin dry. Adult sea otters average 4.5 feet (1.4 m) in length. Their average life span is approximately 15 to 20 years.

Sea Otter capture prey from the sea bottom, then carry it to the surface for handling and feeding. In Washington, prey items include urchins, clams, mussels, crabs, snails and chitons. Adult Sea Otter can eat 25% to 30% of their body weight in one day.

Distribution and Habitat

Sea Otter once ranged from Mexico to Alaska and even to Japan. They inhabit nearshore waters up to 20 fathoms deep, and seldom venture more than 1 to 2 km from land. They typically inhabit rocky habitats with kelp beds, but also occur at lower densities in soft-sediment areas without kelp. Kelp is generally considered an important element of their habitat, used for foraging and resting.

In Washington, Sea Otter historically occurred in estuarine and sandy habitats from the Columbia River to Point Grenville, along the rocky outer Olympic Peninsula coast, and into the Strait of Juan de Fuca. Few reached the San Juan Islands or Discovery Bay, and none were present in Puget Sound. The species was extirpated from the State by about 1910 (Scheffer 1940, Kenyon 1969).

Sea Otter were reintroduced to Washington in 1969 and 1970, when 59 animals were translocated from Amchitka Island, Alaska. The population grew by approximately 7.6% per year between 1991 and 2012 to 1,105 animals (Jameson and Jeffries 2013). At present, Sea Otter occur primarily in rocky habitats along the Olympia Peninsula coast from Destruction Island northward to Tatoosh Island. Colonization of the western Strait of Juan de Fuca has not yet occurred despite the presence of groups of animals using the area during fall and winter months until the year 2000 (Laidre et al. 2009).

Population growth increased to a 3-year running average of 1,753 individuals between 2015 and 2017. This exceeds the down-listing objective in the 2004 Recovery Plan of 1,640 Sea Otter over a three-year period (Sato 2018).

The distribution pattern of the Washington Sea Otter population has gradually changed in recent years with an increasing and larger proportion of the population now occurring south of La Push. In 2012, 73% of the population was south of La Push and 27% was north. Annual surveys do not extend east of Tongue Point, although credible sightings of scattered individual Sea Otter have come from the San Juan Islands and Puget Sound in recent years. No groups of multiple animals have been noted in these areas. In 2012, one individual was reported in South Puget Sound.[21] Currently there is no consensus on why Sea Otter are not clearly expanding into available habitat. [22]

Threats within the Project Action Area

Sea Otter are vulnerable to oil spills. They lack the blubber layer of other marine mammals, therefore relying on their fur to keep warm. If their fur is oiled, it loses its insulating qualities. Otters are also

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21 https://www.eopugetsound.org/articles/sea-otter-enhydra-lutris

22 https://wdfw.wa.gov/publications/01965

affected by oil fumes or poisoned by eating food exposed to oil. Other threats to Sea Otter include infectious diseases, parasites, boat strikes, entanglements in fishing nets, marine biotoxins, loss of kelp habitat, and reduced genetic diversity.

EASTERN POPULATION STELLER SEA LION (*Eumetopias jubatus*)[23]

Status and Description
Steller Sea Lion currently have no Federal listing status.[24] Neither are they listed in Washington State.[25]

In 1997, NMFS classified Steller Sea Lion into two distinct population segments divided by the 144°W latitude. The eastern population segment occupies habitat including southeastern Alaska and Admiralty Island. Currently, NMFS has classified the western population segment as endangered, while classifying the eastern population segment as threatened (62FR24345). Although the Steller Sea Lion population has declined steadily for the last 30 years, scientists have yet to identify the cause of the decline (NOAA Fisheries 2019b).

The worldwide Steller Sea Lion population is estimated at just under 200,000, with the majority occurring in Alaska. The range of the Steller Sea Lion extends around the North Pacific Ocean rim from northern Japan, the Kuril Islands and Okhotsk Sea, through the Aleutian Islands and Bering Sea, along Alaska's southern coast, and south to California (Kenyon and Rice 1961, Loughlin et al. 1984).

Steller Sea Lion are opportunistic feeders and consume a variety of fishes such as flatfish, code, and rockfish; and invertebrates such as squid and octopus. Demersal and off-bottom schooling fishes predominate (Jones 1981). Steller Sea Lion along the coasts of Oregon and California have eaten rockfish, hake, flatfish, cusk-eel, squid, and octopus (Fiscus and Baines 1966, Jones 1981, Treacy 1985); rockfish and hake are considered to be consistently important prey items (NMFS 1992). Feeding on lamprey in estuaries and river mouths has also been documented at sites in Oregon and California (Jones 1981, Treacy 1985). Spalding (1964) and Sinclair and Zepplin (2002) have documented Steller Sea Lion feeding on salmon.

Distribution and Habitat
Steller Sea Lion are polygamous and use traditional territorial sites for breeding and resting. Breeding sites, also known as rookeries, occur on both sides of the North Pacific, but the Gulf of Alaska and Aleutian Islands contain most of the large rookeries. Adults congregate for purposes other than breeding in areas known as haul-outs (University of Alaska 2019). Steller Sea Lion haul-out sites in Puget Sound include: Bangor Naval Base; east of Marrowstone Island; Toliva Shoals buoy; docks on Saltair Marina; navigation buoys and net pen floats near Orchard Rocks/NMFS Manchester;[26] old shipwreck on north side of Nisqually River Delta; and navigation buoys between Point Wilson and Point No Point.[27] Other

[23] USEPA 2010.
[24] https://www.fisheries.noaa/gov/species/steller-sea-lion
[25] WDFW 2008 (updated January 2019).
[26] A WDFW site-specific Priority Habitats and Species database search maps priority seal and sea lion haul-outs in a relatively large area along the south side of Rich Passage that includes the existing Cooke Aquaculture Clam Bay marine net pens, and at Orchard Rocks near the existing Cooke Aquaculture Orchard Rocks marine net pen (WDFW July 10, 2019). The database documents the presence of Steller Sea Lion and Harbor Seal at these locations.
[27] Personal communication between Matthew Szelag, EPA and Teresa Mongillo, NOAA. September 22, 2010. Provided information from Jeffries et al. 2000: Navy; WDFW; NMML.

rocks, reefs, and beaches as well as floating docks, navigational aids, jetties, and breakwaters are also used as haul-out areas (NOAA Fisheries 2019b).

Steller Sea Lion may be observed in Puget Sound year-around, but they are most abundant during the fall and winter months. No breeding rookeries have been identified in Washington waters (NOAA Fisheries 2019b).

Critical Habitat
Steller Sea Lion critical habitat has been designated in Alaska, California, and Oregon and includes a 20-nautical-mile buffer around all major haul-outs and rookeries, as well as associated terrestrial, air, and aquatic zones, and three large offshore foraging areas. No critical habitat has been designated in Washington.

Threats within the Project Action Area
Responses to various types of human-induced disturbances have not been specifically studied. Close approach by humans, boats, or aircraft will cause hauled-out Steller Sea Lion to go into the water. Disturbances that cause stampedes on rookeries may cause trampling and abandonment of pups (Lewis 1987). Areas subjected to repeated disturbance may be permanently abandoned (Kenyon 1962), and/or the repeated disturbance may negatively affect the condition or survival of pups through interruption of normal nursing cycles. Low levels of occasional disturbance may have little long-term effect (NOAA Fisheries 2019).

BALD EAGLE (*Haliaeetus leucocephalus*)

Status and Description
The U.S. Fish and Wildlife Service removed Bald Eagle from the ESA threatened and endangered species list in 2007, but they remain protected under the Bald and Golden Eagle Protection Act, the Migratory Bird Act, and the Lacey Act which prohibit the killing, selling, or otherwise harming of eagles, their nests, and their eggs. The Washington Fish and Wildlife Commission changed the State status of Bald Eagle from "threatened" to "sensitive" in 2011, thereby eliminating many of the State's special protective measures. The Washington Department of Fish & Wildlife recommended in 2016 that the "sensitive" status for Bald Eagle is no longer appropriate and that the species be removed from the State list of endangered species (Kalasz and Buchanan 2016). The U.S. Fish and Wildlife Service now has primary responsibility for managing Bald Eagle under the provisions of the Federal regulations listed above.[28]

A review of all known Bald Eagle territories in the Washington Species Data Management system indicates that the number of territories has increased by an average of 28 per year since 2005 when the species was down-listed to "sensitive" in the State of Washington. As of 2015, the total number of known territories in the State was 1,334, though this total reflects the cumulative number of sites and not the number that are known to be active in any particular year (Kalasz and Buchanan 2016).

Distribution and Habitat
Bald Eagles are well distributed in Washington, but the majority of the population is found west of the Cascade Mountain Range. They are found in association with marine environments and nearly all major waterways, inland lakes, and reservoirs. They are largely piscivorous (fish eaters), and also take birds and mammals. They also commonly practice scavenging.

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28 https://wwwwdfw.wa.gov/species-habitats/at-risk/species-recovery/bald eagle

Threats within the Project Action Area
When Bald Eagle was first listed as endangered under the Federal ESA in 1978, the primary reason for its imperiled status was due to the effects of chemical contaminants and, to a lesser extent, habitat loss. With the restrictions placed on the use of DDT and the decline in use of other environmental contaminants, the Bald Eagle population has rebounded. While there are still threats to Bald Eagles, including the presence of environmental contaminants, none of the threats that previously impacted them are presently having known deleterious effects. Current population analyses indicate that Bald Eagle populations will continue to grow despite these effects (Kalaz and Buchanan 2016).

MARBLED MURRELET (*Brachyramphus marmoratus*)[29,30,31]

Status and Description
Marbled Murrelet were declared by the U.S. Fish and Wildlife Service to be a threatened species in California, Oregon and Washington under the Endangered Species Act in 1992. Marbled Murrelet were State-listed as threatened in 2013. Due to the magnitude of population decline since State listing in 1993, WDFW recently recommended that Marbled Murrelet be listed as a State endangered species (Desimone 2016).

In Washington, nesting habitat losses due to timber harvest since 1993 have been substantial, with an estimated loss on non-Federal lands of 30%. At-sea population monitoring from 2001 to 2015 indicated a 4.4% decline annually, which represents a 44% reduction of the population since 2001. The 2015 population estimate for Washington is about 7,500 birds.

Marbled Murrelet are members of the seabird family Alcidae. Like other alcids, they forage by diving, using their wings for underwater propulsion. The typical flight speed of a Marbled Murrelet is 43 mph (70 km/hr) or faster (Burger 1997). They feed primarily on a variety of forage fishes, and sometimes on larger zooplankton. Their main prey items include immature Pacific herring, northern anchovy, Pacific sand lance, capelin, surf smelt, juvenile rockfish, and immature salmon. They will also prey upon squid and pelagic crustaceans (amphipods, mysids, and euphausiids), especially during the non-breeding season (Sealy 1975, Carter 1984, Burkett 1995, Nelson 1997).

Distribution and Habitat
In Washington, Marbled Murrelet are found in nearshore marine areas (within 1.2 mi [2 km] of shoreline), with the greatest concentrations in North Puget Sound (WDW 1993). Marbled Murrelet spend most of their lives in or near marine waters, except during nesting. Nests are located in large branches or other large platforms in conifer trees, but the species prefers mature, old-growth forests (Nelson et al. 2006, WDFW 2012). Terrestrial nesting habitat distribution includes western Washington within 55 miles of marine waters (Falxa and Raphael 2016). Nest locations in Washington have been documented from near sea level to 4,200 feet elevation and inland to 36.5 miles from the nearest marine water. Parent Marbled Murrelet will travel between the nests sites and coastal marine forage areas to exchange incubation or chick-rearing duties. They exhibit strong site fidelity to nesting areas, appear to nest in alternate years, on average, and have a naturally low reproductive rate.

Threats within the Project Action Area
The largest threat to Marbled Murrelet populations is deforestation and loss of old-growth forests used for nesting habitat. Fragmented forests are more vulnerable to excessive wind at the edges, and more exposed

29 RPS ASA 2016.
30 Hamer Environmental 2016.
31 Desimone 2016.

to predation from jays, ravens and crows, making the loss of thickly settled forests unfavorable for the Marbled Murrelet (WDW 1993). Oil pollution has also posed a threat to Marbled Murrelet and their coastal environment, with the *Seagate* (1956), *Arco Anchorage* (1985), *Nestucca* (1988) and *Tenyu Maru* (1991) oil spills resulting in oiled Murrelets (WDW 1993). Marbled Murrelet entanglement and mortalities in gill nets and hook-and-line fisheries have been observed but not quantified. Irrespective of the risk, the low reproductive rate of the Marbled Murrelet species limits the ability of the population to recover after an episodic or chronic threat. A nest success rate in Washington of 20% for the period 2004-2008 was attributed to nestling starvation or adults abandoning eggs before completing incubation, suggesting low prey availability.

CHINOOK SALMON (*Oncorhynchus tshawytscha*)[32]

Chinook Salmon are easily distinguished from other *Oncorhynchus* species by their large size. Adults weighing over 120 pounds have been caught in North American waters. Chinook Salmon are very similar to Coho Salmon in appearance while at sea (blue-green back with silver flanks), except for their large size, small black spots on both lobes of the tail, and black pigment along the base of the teeth. Chinook Salmon are anadromous and semelparous; i.e., adults, they migrate from a marine environment into the freshwater streams and rivers of their birth (anadromous) where they spawn and die (semelparous).

Juvenile stream-type and ocean-type Chinook Salmon have adapted to different ecological niches. Ocean-type Chinook Salmon tend to utilize estuaries and coastal areas more extensively for juvenile rearing. The brackish water areas in estuaries also moderate physiological stress during parr-smolt transition. The development of the ocean-type life history strategy may have been a response to the limited carrying capacity of smaller stream systems and glacially-scoured, unproductive watersheds, or a means of avoiding the impact of seasonal floods in the lower portion of many watersheds.

Puget Sound Chinook Salmon[33]

Status and Description

Puget Sound Chinook Salmon are Federally-listed as threatened. This Evolutionary Significant Unit (ESU) is a candidate species for listing in Washington State.[34]

Distribution and Habitat

The boundaries of the Puget Sound Chinook Salmon evolutionarily significant unit (ESU) correspond with the Puget Lowland Ecoregion. This ESU encompasses all runs of Chinook Salmon in the Puget Sound region from the North Fork Nooksack River to the Elwha River on the Olympic Peninsula, including Hood Canal.

Chinook Salmon in this area all exhibit an ocean-type life history. Although some spring-run Chinook Salmon populations in the Puget Sound ESU have a high proportion of yearling smolt emigrants, the proportion varies substantially from year to year and appears to be environmentally mediated rather than genetically determined. Puget Sound stocks all tend to mature at ages 3 and 4 and exhibit similar, coastally-oriented, ocean migration patterns (Meyers et al. 1998).

Chinook Salmon prefer to spawn and rear in the mainstem of rivers and larger streams (Healey 1991). Although the incubation period is determined by water temperatures, fry typically hatch in about 8 weeks

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32 USEPA 2010.
33 USEPA 2010.
34 WDFW 2008 (updated January 2019).

(Wydoski and Whitney 1979, Healey 1991). After emergence, Puget Sound juvenile Chinook Salmon migrate to the marine environment during their first year.

Rearing and development to adulthood occurs primarily in estuarine and coastal waters (NOAA Fisheries 2019c). The amount of time juvenile Chinook Salmon spend in estuarine areas depends upon their size at downstream migration and rate of growth. While residing in upper estuaries, juvenile prey mainly on benthic and epibenthic organisms such as amphipods, mysids, and cumaceans. Juveniles typically move into deeper waters when they reach approximately 65 to 75 mm in fork length. As the juveniles grow and move to deeper waters with higher salinities, their main prey changes to pelagic organisms such as decapod larvae, larval and juvenile fish, drift insects, and euphausids (Simenstad et al. 1977).

Hatchery Influence
Hatchery fish are known to spawn in the wild and are not considered discrete stocks from the wild fish (WDFW and WWTIT 1994). State-run and Federally-run Chinook Salmon hatcheries were operating in the Puget Sound Chinook Salmon ESU by 1908. Transfers of Chinook Salmon eggs to Puget Sound from other regions, especially the Lower Columbia River, were common practices of early hatcheries (Myers et al. 1998). By the 1920s, several million Chinook Salmon had been released into Puget Sound tributaries (Cobb 1930). Recently, stock integrity and genetic diversity have become important objectives. New policies have been initiated to reduce the impact of hatchery fish on natural populations (Levin and Williams 2002). The abundance of Chinook Salmon in watersheds throughout this ESU has been closely related to hatchery efforts (Myers et al. 1998).

WDFW classified 11 out of 29 stocks in this ESU as being sustained, in part, through artificial propagation. Nearly 2 billion fish have been released into Puget Sound tributaries since the 1950s. The vast majority of these have been derived from local returning fall-run adults. Returns to hatcheries have accounted for 57% of the total spawning escapement, although the hatchery contribution to spawner escapement is probably much higher than that, due to hatchery-derived strays on the spawning grounds (Good et al. 2005, NMFS-NWFSC-66).

Critical Habitat
Critical habitat designation for Puget Sound Chinook (PS Chinook) and Hood Canal Summer Run (HCSR) Chum Salmon was finalized September 2, 2005 (70 FR 52630) and includes Puget Sound marine areas − the South Sound, Hood Canal, and North Sound to the international boundary at the outer extent of the Strait of Georgia, Haro Strait, and the Strait of Juan de Fuca to a straight line extending north from the west end of Freshwater Bay, inclusive. Critical habitat consists of the water, substrate, and the adjacent riparian zone of accessible estuarine and riverine reaches. Cooke Aquaculture existing Puget Sound net pens sited in less than 98 feet depth (below MLLW) fall within designated critical habitat for PS Chinook and HCSR Chum Salmon. Six of the existing seven net pens are sited in less than 98 feet of water, with one near that depth at low tide. The applicable primary constituent element (PCE) for the PS Chinook and HCSR Chum Salmon designated critical habitat in this action area is: nearshore marine areas free of obstruction and excessive predation with water quality and quantity conditions and forage, including aquatic invertebrates and fishes, supporting growth and maturation, and natural cover such as submerged and overhanging large wood, aquatic vegetation, etc.[35]

Threats within the Project Action Area
Chinook Salmon were abundant in Washington State near the turn of the century, when estimates based on peak cannery pack suggested peak runs of near one million fish in the Oregon Coast, Washington Coast, and Puget Sound ESUs. However, Chinook Salmon in this region has been strongly affected by

[35] NMFS 2011.

losses and alterations of freshwater habitat. Timber harvesting and associated road building have occurred throughout this region. Agriculture is also widespread in the lower portions of river basins and has resulted in widespread removal of riparian vegetation, rerouting of streams, degradation of streambanks, and summer water withdrawals. Urban development has substantially altered watershed hydrodynamics and affected stream channel structure in many parts of Puget Sound.

Widespread stream blockages have reduced available spawning habitat, and widespread release of hatchery fish from limited stocks has increased the risks of loss of genetic diversity and fitness to natural populations. In addition, the large numbers of hatchery releases masks natural population trends, making it difficult to determine their sustainability. Forestry practices, farming and urbanization have blocked or degraded fresh water habitat (Myers et al. 1998).

HOOD CANAL SUMMER-RUN CHUM SALMON (*Oncorhynchus keta*)[36]

Chum Salmon have the widest natural geographic distribution of all Pacific salmon species, ranging in Asia from Korea to the Russian Arctic coast and west to the Lena River, and in North America from Monterey, California, to the Arctic coast and east to the Mackenzie River (Beaufort Sea). Historically, they may have constituted up to 50% of the annual biomass of the seven species of Pacific salmon in the North Pacific Ocean (Sato et al. 2009).

Chum Salmon spawn successfully in streams of various sizes, and the fry migrate directly to the sea soon after emergence. Immature chum distribute themselves widely over the North Pacific Ocean, and maturing adults return to the home streams at various ages, usually at 2 through 5 years, and in some cases up to 7 years (Groot and Margolis 1991). Common to virtually every region of the Chum Salmon’s area of distribution is the occurrence of early and late returning stocks to the natal stream. In North America, the only true summer Chum Salmon may be in the Yukon River, where summer Chum have the distinguishing characteristics of the Asian summer Chum. From western Alaska south to British Columbia and Washington, there are runs referred to as “summer” Chum that spawn from June to early September; these Chum are characterized by large body size, older age composition, and high fecundity, and are probably early autumn Chum (Groot and Margolis 1991).

In general, early-run Chum Salmon spawn in stream main stems, while late spawners seek out spring water that has more favorable temperatures through the winter. The timing of the runs varies from north to south, as does age at maturity and absolute (and probably, relative) fecundity (Groot and Margolis 1991).

Hood Canal Summer-run Chum Salmon

Status and Description

The Hood Canal Summer-run Chum Salmon (HCSR) ESU was Federally-listed as threatened on August 2, 1999. The Puget Sound/Strait of Georgia ESU is not Federally-listed, but is a candidate species for listing in Washington State.[37]

Most Hood Canal Summer-run Chum spawn in early September to mid-October. The Union River Summer-run Chum is an exception as they have an earlier spawning timing (September – early October). Fry emerge from February to June. In Washington, Chum may reside in freshwater for as long as a month before migration to estuarine habitats where they remain for about a month before migrating to deeper water (Johnson et al. 1997).

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36 USEPA 2010.

37 WDFW 2008 (updated January 2019).

Distribution and Habitat
This ESU includes Summer-run Chum Salmon populations in Hood Canal in Puget Sound and in Discovery and Sequim Bays on the Strait of Juan de Fuca. It may also include summer-run fish in the Dungeness River, but the existence of that run is uncertain. Distinctive life-history and genetic traits were the most important factors in identifying this ESU. Hood Canal Summer-run Chum Salmon are defined as fish that spawn from mid-September to mid-October in the mainstems of rivers (Johnson et al. 1997).

Hatchery Influence
Very few Summer-run Chum Salmon have been artificially propagated in Hood Canal, and the only releases in recent years have been from newly established restoration programs. These recent releases totaled about 241,000 Chum Salmon fry into Hood Canal in 1993 and 1994 and about 85,000 fry into Discovery Bay on the Strait of Juan de Fuca in 1992.

Critical Habitat
Critical habitat for Hood Canal Summer-run Chum Salmon was designated September 2, 2005 (70 FR 52630), concurrent with critical habitat designation for Puget Sound Chinook Salmon, as described above.

Threats within the Project Action Area
This ESU is in danger of extinction. Of 12 streams in Hood Canal identified as recently supporting spawning populations of summer Chum Salmon, five may already have become extinct, six of the remaining seven showed strong downward trends in abundance, and all were at low levels of abundance. Threats to the continued existence of these populations include degradation of spawning habitat, low water flows, and incidental harvest in salmon fisheries in the Strait of Juan de Fuca and Coho Salmon fisheries in Hood Canal (Johnson et al. 1997).

COHO SALMON (*Oncorhynchus kisutch*)

Status and Description
The Puget Sound/Strait of Georgia population of Coho Salmon are Federally-listed as a species of concern. They have no special status listing in Washington State.[38] A NMFS Biological Review Team found in 1995 that population abundance was near historical levels, and recent trends in overall population abundance had not been downward; however, there was substantial uncertainty relating to several of the risk factors considered (described below under Threats within the Project Action Area).

Coho have a very regular life history. Eggs are deposited in stream gravel in the fall, alevins emerge from the gravel the next spring, and in their second spring juveniles go to sea, about 18 months after hatching. Adults range in size from 6 to 12 pounds, and up to 31 pounds.

Distribution and Habitat
The Coho Salmon Puget Sound/Strait of Georgia ESU utilizes drainages in Puget Sound and Hood Canal, the eastern Olympic Peninsula (east of Salt Creek), and the Strait of George from the eastern side of Vancouver Island and the British Columbia mainland (north to and including Campbell River and Powell River), excluding the upper Fraser River above Hope (Weitkamp, L. et al. 1995).

This species uses coastal streams and tributaries, and can even be found in urban settings if their needs for cold, clean, year-around water are met. Coho spawn in small coastal streams and in the tributaries of larger rivers. They prefer areas of mid-velocity water with small to medium-sized gravels. Because they

38 WDFW 2008 (updated January 2019).

use small streams with limited space, they must use many such streams to successfully reproduce. Returning Coho often gather at the mouths of streams and wait for the water flow to rise, such as after a rain storm, before heading upstream. The higher flows and deeper water enable the fish to pass obstacles, such as logs across the stream or beaver dams that would otherwise be impassable.[39]

The WDFW Priority Habitats and Species (PHS) database (July 10, 2019) maps Coho Salmon as a priority fish presence in Beaver Creek that discharges to Clam Bay in the vicinity of the Cooke Aquaculture existing marine net pen site at this location in Rich Passage. The PHS database also maps Coho Salmon as a priority fish presence in Tumwater Creek that discharges to Port Angeles Harbor approximately 1.5 miles south of the Cooke Aquaculture existing net pen location near the eastern terminus of Ediz Hook within Port Angeles Harbor.

Threats within the Project Action Area

Threats to the Coho Salmon Puget Sound/Strait of Georgia population include widespread and intensive artificial propagation, high harvest rates, extensive habitat degradation, and unfavorable ocean conditions (Weitkamp, L. et al. 1995).

STEELHEAD (*Oncorhynchus mykiss*)[40]

Steelhead are the anadromous form of the Rainbow Trout (*O. mykiss*), which occurs in two subspecies, *O. mykiss irideus* and *O. mykiss gaidneri*. Whereas stream-resident Rainbow Trout may complete their life cycle in a limited area of a small stream and attain a length of only 8 inches or so, steelhead generally spend half their lives at sea, roaming for thousands of miles in the North Pacific Ocean. Steelhead return to spawn at sizes ranging from about 24 inches and 5 pounds to about 36 to 40 inches or more and 20 pounds or more (Behnke 2002).

Biologically, steelhead can be divided into two reproductive ecotypes, based on their state of sexual maturity at the time of river entry. These two ecotypes are termed “stream-maturing” and “ocean-maturing.” Stream-maturing steelhead enter fresh water in a sexually immature condition and require from several months to a year to mature and spawn. These fish are often referred to as “summer-run” steelhead. Ocean-maturing steelhead enter fresh water with well-developed gonads and spawn shortly after river entry. These fish are commonly referred to as “winter-run” steelhead. Ocean-maturing fish are the predominate ecotype in coastal streams within the action area.

Puget Sound populations begin spawning in February or March, with summer-run steelhead spawning from March into June.

Juvenile steelhead generally spend two years in freshwater before smolting and migrating to the ocean at lengths of about 6 to 8 inches. Most steelhead return to their natal rivers to spawn after spending 15 to 30 months in the ocean. Unlike other Pacific salmonids, steelhead do not all die soon after spawning, but the rate of survival to repeat spawning is generally low – about 10% (Behnke 2002).

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39 https://wdfw.wa/gov/species-habitats/species/oncorhynchus-kisutch

40 USEPA 2010.

Puget Sound Steelhead ESU[41]

Status and Description

The Puget Sound steelhead ESU was officially listed as "threatened" by NOAA Fisheries Service on March 11, 2007. This species is a candidate for listing in Washington State.[42] The Puget Sound distinct population segment (DPS) of steelhead originates downstream from natural and manmade impassable barriers from rivers flowing into Puget Sound from the Elwha River eastward, including rivers in Hood Canal, the South Sound, the North Sound and the Strait of Georgia. Numerous juvenile steelhead are also produced at six artificial hatchery facilities.

In the 1980s, the Puget Sound steelhead run size was estimated as 100,000 winter-run and 20,000 summer-run. In the 1990s, the total run size for major stocks in this ESU was greater than 45,000 with natural escapement estimates of 22,000 steelhead.

Distribution and Habitat

The Puget Sound steelhead DPS includes all naturally spawned anadromous winter-run and summer-run *O. mykiss* (steelhead) populations in streams of the Strait of Juan de Fuca, Puget Sound, and Hood Canal, basins. This area is bounded to the west by the Elwha River (inclusive) and to the north by the Nooksack River and Dakota Creek (inclusive), as well as the Green River natural and Hamma Hamma winter-run steelhead hatchery stocks.

Hatchery Influence

Releases of hatchery-propagated steelhead into Puget Sound waters began in the 1900s. By the 1940s, extensive hatchery rearing programs were developed. Hatchery fish were widespread, spawning naturally throughout the region, and were largely derived from a single stock (Chambers Creek). In the 1980s, the hatchery portion of the population based on ocean catches was 70%. During the two decades that followed, release levels of hatchery steelhead remained relatively constant. Hatchery-produced winter steelhead were released in nearly every basin in the ESU, except for the Cedar River and some smaller tributaries.

The risk posed by artificial production programs to natural production in the Puget Sound steelhead ESU is not clear as definitive information is not available. However, the genetic and life-history relationships between the Chambers Creek Hatchery and Skamania Hatchery and the naturally-spawning populations indicate that these hatchery effects could be substantially detrimental. Approximately six million hatchery-produced steelhead have been released in Washington waters annually during recent years (WDFW web site). These planted fish are diploid steelhead capable of natural reproduction and degradation of wild steelhead populations.

Critical Habitat

Puget Sound steelhead Distinct Population Segment (DPS) critical habitat was designated by the National Marine Fisheries Services on March 25, 2016 (81 FR 9251: 9251-9325). Specific areas include approximately 2,031 miles of freshwater and estuarine habitat in Puget Sound. A number of particular areas (e.g., military properties) are excluded from the critical habitat designation because the benefits of exclusion outweigh the benefits of inclusion, and exclusion will not result in extinction of the species. Much of the designated critical habitat is fresh water spawning, rearing, and migratory habitat. This includes estuarine areas free of obstruction with water quality and salinity conditions that support juvenile

41 NMFS 2005.

42 WDFW 2008 (updated January 2019).

and adult physiological transitions between fresh water and saltwater, and juvenile and adult forage habitat that support growth and maturation.

Threats within the Project Action Area

Habitat loss, hatchery steelhead introgression, and harvest are the major contributors to the decline of steelhead in this ESU. Habitat utilization by steelhead has been affected by a number of large dams in Puget Sound basins. Besides eliminating access to habitat, dams affect habitat quality by changing river hydrology, temperature profiles, gravel recruitment, and large woody debris movement and stability. Urban development and suburbanization have resulted in the loss of historical land cover, often replacing it with imperious surface. Combined with loss of wetland/riparian habitat, hydrology of many urban streams has changed dramatically. Flood frequency and peak flow during storm events has increased and groundwater derived summer flows have decreased. Land development for agriculture has also altered historical land cover. Diking, riprapping of banks, and channelization have resulted in river constriction which increases gravel scour, decreases habitat complexity, and alters amplitude of high flow events.

BULL TROUT (*Salvelinus confluentus*)

Status and Description

Bull Trout are Federally-listed as threatened. They are a candidate species for listing in Washington State.[43] Bull Trout are members of the char subgroup of the family Salmonidae that are native to waters of western North America.

Distribution and Habitat

Bull Trout have more specific habitat requirements than most salmonids. Habitat characteristics that particularly influence their distribution and abundance include water temperature, cover, channel form and stability, spawning and rearing substrate conditions, and migratory corridors. Juvenile Bull Trout usually rear from 1 to 4 years in their natal stream before migrating to either a larger river (fluvial) or lake (adfluvial). Some Bull Trout (amphidromous) produced in Puget Sound streams migrate to estuarine waters for rearing prior to returning to their natal stream to spawn or for additional rearing.

Critical Habitat

The U.S. Fish and Wildlife Service designated 19,729.0 miles (31,750.8 km) of streams including 754.0 miles (1,213.2 km) of marine shoreline) as Bull Trout critical habitat (Federal Register 2010/Vol. 75, No. 200) in Washington, Oregon, Nevada, Idaho, and Montana. Bull Trout have more specific habitat requirements than most other salmonids (Rieman and McIntyre 1993). Habitat components that particularly influence their distribution and abundance include water temperature, cover, channel form and stability, spawning and rearing substrate conditions, and migratory corridors (Fraley and Shepard 1989; Goetz 1989; and Watson and Hillman 1997). The Primary Constituent Elements (PCE) of Bull Trout critical habitat are: space for individual and population growth and for normal behavior; food, water, air, light, minerals, or other nutritional or physiological requirements; cover or shelter; sites for breeding, reproduction, or rearing (or development) of offspring; and habitats that are protected from disturbance or are representative of the historical, geographical, and ecological distributions of a species (USFWS 2010, FR 75(20): 63898-64070).[44]

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[43] WDFW 2008 (updated January 2019).

[44] USEPA 2010.

Threats within the Project Action Area
The decline in Bull Trout populations is primarily due to fresh water habitat degradation and fragmentation, blockage of migratory corridors, poor water quality, past fisheries management practices, impoundments, dams, water diversions, and the introduction of nonnative species.

EULACHON (*Thaleichthys pacificus*)[45]

Status and Description
Eulachon (also known as Columbia River smelt or hooligan) are Federally-listed as threatened. They are State-listed as a candidate species for further status designation at a later time. Eulachon are a small anadromous fish, typically weighing 2.4 ounces (0.07 kg) with a length of 8.5 in (0.22 m; NOAA 2014d). The maximum reported age for the species is 5 years, spending 3 to 5 years at sea before returning to rivers for spawning (Hugg 1996, NOAA 2014d). Most Eulachon are semelparous[46], dying soon after spawning. Eggs hatch in 20 to 40 days, and larvae are transported downstream for dispersion in estuary and ocean bottom waters (NOAA 2011b; NOAA 2014d). Larvae feed on phytoplankton, copepod eggs, mysid shrimp, barnacle larvae, and worm larvae (NOAA 2011b). They also feed on large zooplankton, krill, and small crustaceans. Adults will not feed while they are spawning (Hart and McHugh 1944).

Distribution and Habitat
Eulachon live in nearshore waters over the continental shelf (up to a maximum depth of 980 ft [300 m] but often shallower), except for when the species returns to its natal river to spawn (NOAA 2014d). Eulachon occur along the eastern Pacific Ocean, extending from northern California to the southeast Bering Sea (Gustafson et al. 2010). In U.S. waters, most Eulachon originate from the Columbia River basin and its tributaries (NOAA 2011b). Eulachon have been occasionally reported in coastal Washington rivers.

Threats within the Project Action Area
The greatest threat to Eulachon is reduction in suitable habitat availability, particularly in the Columbia River basin (NOAA 2014d). The major threats to Eulachon habitat include damn construction, which prevents spawning runs and degrades demersal spawning habitat, and dredging, which exposes the water column to sediment particulates. Global climate change is likely to pose a threat to Eulachon, with increasing water temperatures making presently-occupied spawning grounds and rivers unsuitable habitat. While fishing pressure has been reduced, by-catch and specific fishing methods may still pose a threat to eulachon.

FLATHEAD SOLE (*Hippoglossoides elassodon*)[47]

Status and Description
Flathead Sole are Federally-listed as a species of concern. They have no State-listing status.[48] They are a right-eyed flatfish with a very flat, oval body shape. They have a medium to large mouth with one row of sharp conical teeth in both jaws. The Flathead Sole is very similar to the Petrale Sole, with the distinguishing factors of Petrale Sole being two rows of teeth in the upper jar and a rounded rear ridge on the lower jaw.

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45 RPS ASA 2016.
46 A species is considered semelparous if it is characterized by a single reproductive episode before death.
47 WDFW 2019.
48 WDFW 2008 (updated January 2019).

Flathead Sole can grow up to 22 inches (56 cm) in length, and approximately 3.4 pounds (1.56 kg) in weight. Maximum age is at least 27 years for females and 30 years for males.

Distribution and Habitat
Flathead Sole range from the seas of Japan and Okhotsk, across the Bering Sea and to the coast of North America as far south as Point Reyes, California. This species lives on soft, silty or muddy bottoms at depths of up to approximately 3,500 feet (1,050 m). They are most commonly encountered within the water depth range of approximately 325 to 800 feet (100 to 247 m). Flathead Sole are occasionally caught off the Washington coast by commercial harvesters using otter-trawls and long-line gear. They are rarely caught by recreational fishermen within Puget Sound.

Threats within the Project Action Area
The action area is in the southern end of the Flathead Sole range. Flathead Sole in the Gulf of Alaska are not overfished and are not subject to overfishing. Flathead sole are quite abundant in Alaska, and populations are well above target levels. On the West Coast, Flathead Sole make up only a small percentage of groundfish harvests. They are part of the "other flatfish" complex on the West Coast, and are not subject to overfishing (NOAA 2019c).[49] The normal depth distribution of Flathead Sole is 325 feet and deeper, which is not within the more shallow range of depths potentially affected by the proposed action.

GREEN STURGEON (*Acipenser medirostris*)[50]

Status and Description
Green Sturgeon are Federally-listed as threatened, but do not have a priority status in Washington State. They are an anadromous fish with olive green dorsal coloring and yellowish green-white coloring underside. They are more marine-oriented than salmonids or any other sturgeon species (Adams et al. 2002), and live primarily at sea or in estuaries, except when spawning. Unlike salmon, they can spawn several times within their lifetime and will return to natal rivers every 2 to 5 years to do so (Moyle 2002). They are a long-lived, slow-growing species, reaching maturity around age 15, with a reported maximum rage between 60 and 70 years old. Prey items include shrimp, mollusks, amphipods, and small fish. The only known spawning locations are the Klamath, Rogue, and Sacramento Rivers along the U.S. west coast. Within rivers, adults will spawn in cool waters with deep, turbulent flow and hard clean substrate (Moyle et al. 1992).

Distribution and Habitat
Green Sturgeon range from nearshore Mexico waters north to the Bering Sea, with the majority of Green Sturgeon occurring in U.S. waters located from Monterey, California north to Puget Sound. The species is separated into two distinct population segments. The northern DPS (nDPS) includes sturgeon that spawn from the Klamath River in Northern California through the Rogue River in Oregon (NOAA 2015). This nPDS has been listed by NOAA as a species of concern. The southern DPS (sDPS) consists of Green Sturgeon that spawn in the Sacramento River. These are Federally-listed as threatened (NOAA 2015). Green Sturgeon use both marine and freshwater habitats throughout their life cycle and are believed to migrate long distances after leaving estuarine waters.

Critical Habitat
The northern range of Green Sturgeon critical habitat includes the southern portion of the Strait of Juan de Fuca and coastal Port Angeles waters. While the Puget Sound region is not a spawning area for Green

49 NOAA 2019c. https://www.fisheries.noaa.gov/species/flathead-sole
50 RPS ASA 2016.

Sturgeon, the species spends significant time in coastal regions of Washington and open ocean waters (NOAA 2014e). Thus, the relatively shallow Puget Sound waters where existing marine net pen sites are located are unlikely to provide habitat used by Green Sturgeon for feeding or migration.

Threats within the Project Action Area

Some of the greatest threats to Green Sturgeon are impediments to migration up or down rivers, and larval survival in river systems. Some of these threats include low freshwater flow from dams, contaminants introduced to river waters, poor water quality, entrainment from water projects, and impassable barriers. By-catch and fishing mortality are other sources of concern, as historical overfishing of a small population has contributed to their low population numbers today. With changes in climate, increased water temperatures and introduction of invasive exotic species to natal grounds could also be detrimental. Reduction in spawning area within the Sacramento River has been a great concern for the sDPS stock.

PACIFIC COD (*Gadus macrocephalus*)[51]

Status and Description

Pacific Cod are Federally-listed as a species of concern, and State-listed as a candidate species for further status designation at a later time. An Endangered Species Act status review for Pacific Cod (Gustafson et al. 2000, NOAA 2011c) stated that the population structure for this species includes a distinct population segment (DPS) along the West Coast of North America including Puget Sound and coastal British Columbia north to at least Dixon Entrance.

Pacific Cod are a commercially-important fish species in some parts of their size range (NOAA 2011c). They feed on krill, shrimp, sand lance and crabs and are prey for seals, halibut and other fishes (Love 1996, NOAA 2011c).

Distribution and Habitat

The Salish Sea population of Pacific Cod is found in Puget Sound, the Strait of Juan de Fuca, and the Strait of Georgia in the northwestern U.S. and southeastern Canada (NOAA 2011c).

Pacific Cod are schooling fish that live near the ocean bottom (NOAA 2011c). In the Salish Sea, they can be found over sand and mixed coarse bottom substrates (Palsson 1990, NOAA 2011c). Adult Pacific Cod occur as deep as 2,870 ft (875 m), but the vast majority occurs between 160 and 980 ft (50 to 300 m) (WDWF and NMFS 2005). In recent years, Pacific Cod have become rare or extinct in most Puget Sound waters, in part due to slight increases in water temperatures at this southern edge of their North Pacific range.

Threats within the Project Action Area

The primary threats for Pacific Cod within the project action area include small population size due to past overfishing (NOAA 2011c). While they were once an abundant species important in the sport and commercial fisheries in the Salish Sea, they became overfished in the early 1990s and the population has still not rebounded (Palsson 1990, NOAA 2011c). Other threats to this population of Pacific Cod include warmer waters due to global climate change, and predation (Gustafson et al. 2000, Beamish 2008, NOAA 2011c).

51 RPS ASA 2016.

PACIFIC HAKE (*Merluccius productus*)[52]

Status and Description

Pacific Hake are Federally-listed as a species of concern, and State-listed as a candidate species for further status designation at a later time. They are a groundfish of the order Gadiformes. They can reach up to 36 inches (91 cm) in length and 15 years of age (NOAA 2009). Pacific Hake larvae prey on calanoid[53] copepod eggs, nauplii[54], and adults (McFarlane and Beamish 1986, Sumida and Moser 1984, Gustafson et al. 2000). Juveniles and small adults mostly prey on euphausiids[55] (NOAA 1990, Gustafson et al. 2000). Large adults also eat amphipods, squid, Pacific Herring, smelt, crabs, shrimp, and sometimes juvenile Pacific Hake (Dark and Wilkins 1994, McFarlane and Beamish 1986, NOAA 1990, Gustafson et al. 2000).

Distribution and Habitat

There are three recognized stocks of Pacific Hake, one of which is the Georgia Basin DPS, which include both the Puget Sound and Strait of Georgia stocks (NOAA 2009). They are found near the bottom or in the water column to depths of 3,000 feet (914 m); however, they are most common in water shallower than 750 feet (229 m) (WDWF 2016c). There are several spawning locations for Pacific Hake in Puget Sound (Gustafson et al. 2000).

Threats within the Project Action Area

One of the major factors for the decline of Pacific Hake within the Puget Sound area is overfishing (NOAA 2009). Other threats for this species include pinniped predation, habitat alteration or loss, and environmental changes (Gustafson et al. 2000). Though the potential effects of habitat loss or degradation are unknown, it has been speculated that juvenile Hake survival could be reduced through loss or degradation of nearshore nursery habitats (West 1997, Gustafson et al. 2000).

PACIFIC LAMPREY (*Entosphenus tridentatus*)[56]

Status and Description

Pacific Lamprey have no current Federal or State listing status.[57]

Lampreys belong to a primitive group of fishes that are eel-like in form and lack the jaws and paired fins of true fishes. Pacific Lamprey have a round, sucker-like mouth, no scales, and gill openings. Lamprey identification depends largely on the number, structure, and position of teeth in adults. Adult Pacific Lamprey have three large anterior teeth and many smaller posterior teeth on the oral disc.

As adults in the marine environment, Pacific Lamprey are parasitic and feed on a variety of marine and anadromous fish, including Pacific salmon, flatfish, rockfish, and pollock. They are preyed upon by sharks, sea lions, and other marine animals.

After spending 1 to 3 years in the marine environment, Pacific Lamprey cease feeding and migrate to freshwater between February and June. Most upstream migration takes place at night. They are thought to overwinter and remain in freshwater habitat for approximately 1 year before spawning. During that time,

52 RPS ASA 2016.
53 Calanoid copepods are a kind of zooplankton in the order Calanoida.
54 Nauplii are the first larval stage of many crustaceans, with an unsegmented body and a single eye.
55 Euphausiids are a type of small, shrimp-like crustaceans (e.g., krill).
56 https://www.fws.gov/oregonfwo/articles.cfm?id=149489457
57 WDFW 2008 (updated January 2019).

they may shrink in size by up to 20%. Spawning occurs between March and July, depending on location within their range.

Distribution and Habitat

Historically, Pacific Lamprey were thought to be distributed wherever salmon and steelhead occurred. However, recent data indicate that the distribution of the Pacific Lamprey has been reduced in many river drainages. They no longer exist upstream from many dams and other impassable barriers in west coast streams, including many larger rivers throughout coastal Washington, Oregon, and California. In the marine environment, they have been caught at depths ranging from 300 to 2,600 ft, and as far off the west coast as 62 miles in ocean-haul nets.

Pacific Lamprey spawn in habitat similar to that of salmon: gravel-bottomed streams at the upstream end of riffle habitat. The degree of homing is unknown, but adult Lamprey cue in on ammocoete areas that release pheromones that are thought to aid adult migration and spawning location.

Threats within the Project Action Area

Pacific Lamprey face a variety of threats to various life history stages, including barriers to upstream and downstream passage; dewatering and reduced stream flows; poisoning from accidental spills or chemical treatments; poor water quality; dredging activities; stream and floodplain degradation due to channelization, loss of side channel habitat, and scouring; changes in ocean conditions such as prey reduction and an increase in predators; and predation by non-native fish species.

RIVER LAMPREY (*Lampetra fluviatilis*)[58]

Status and Description

River Lamprey presently have no Federal listing status, but are State-listed as a candidate species for further status designation at a later time . Available information on the abundance of River Lamprey indicates some potential local declines; however, data are lacking to substantiate a significant decline in abundance or distribution.

Like the Pacific Lamprey described above, the River Lamprey is a member of a primitive group of fishes that are eel-like in form and lack jaws and paired fins. They have a round, sucker-like mouth (oral disc), no scales, and breathing holes instead of gills. Adult River Lamprey have two teeth (cusps) and no posterior teeth on the oral disc. Their life span is 6 to 7 years.

Similar to Pacific Lamprey, River Lamprey spawn in gravel-bottomed streams, at the upstream end of riffle habitat. Both sexes construct the nests, often moving stones with their mouths. River Lamprey lay approximately 11,000 to 37,000 eggs per adult female. Adults typically die after the eggs are deposited and fertilized. After the eggs hatch, young ammocoetes drift downstream to areas of low velocity and silt or sand substrate. They remain burrowed in the stream bottom, living as filter feeders on algae and detritus for 2 to 7 years. Metamorphosis from the ammocoete to macropthalmia life stage occurs between July and April. At this time, macropthalmia are thought to live deep in the river channel, which may explain why they are rarely observed. As adults, their oral disc develops just before they enter the ocean between May and July. They are at sea for approximately 10 weeks in a parasitic phase. During this time, they remain close to shore, feeding primarily on smelt and herring near the surface. After the adult feeding phase, River Lamprey migrate to spawning areas and stop feeding. Their degree of fidelity to their natal streams is unknown.

58 https://www.fws.gov/wafwo/species/Factsheets/Riverlampreyfinal.pdf

Distribution and Habitat
River Lamprey are found from just north of Juneau, Alaska, to San Francisco Bay, California; however, detailed information on their distribution and abundance is lacking. River Lamprey are associated with large river systems such as the Fraser, Columbia, Klamath, Eel, and Sacramento Rivers, and tend to occur only in the lower portions of these rivers.

In Washington, this species probably historically occurred in most major rivers. The current distribution of River Lamprey in the State includes rivers and streams along the coast from the mouth of the Columbia River to the mouth of the Hoh River (though not elsewhere on the Olympic Peninsula), throughout Puget Sound, and in the Lake Washington basin. Adults feed in nearshore marine and estuarine habitat.

Threats within the Project Action Area
Potential threats to River Lamprey include artificial barriers to migration; poor water quality; harvest; predation by non-native species; stream and floodplain degradation; loss of estuarine habitat; decline in prey, ocean conditions, dredging and dewatering.

CANARY ROCKFISH (*Sebastes pinniger*)[59,60]

Status and Description
Canary Rockfish of the Puget Sound/Georgia Basin were determined by the National Marine Fisheries Service (NMFS) in 2010 to be a DPS. NMFS subsequently collected genetic information to better understand the Yelloweye Rockfish, Canary Rockfish, and Bocaccio listed under ESA, and found that Puget Sound/Georgia Basin Canary Rockfish are not discrete from coastal Canary Rockfish. For this reason, Puget Sound/Georgia Basin Canary Rockfish do not qualify for protection under ESA, and they were delisted on January 23, 2017.[61] Though NMFS does not know the abundance of Canary Rockfish in the Puget Sound/Georgia Basin, the overall population has improved dramatically.[62]

Canary Rockfish are a candidate for listing in Washington State. WDFW rockfish regulations in-place since 2010 to protect all species of rockfish are independent of the Federal de-listing of Canary Rockfish. Recreational anglers currently may not target, possess, or retain any type of rockfish in Puget Sound, including the San Juan Islands area, and there still are no commercial fisheries that may target any species of rockfish in Puget Sound.

Rockfishes are unusual among the bony fishes in that fertilization and embryo development is internal and female rockfish give birth to live larval young. Larvae are found in surface waters and may be distributed over a wide area extending several hundred miles offshore. Fecundity in female Canary Rockfish ranges from 260,000 to 1.9 million eggs, considerably more than many other rockfish species. Larvae and small juvenile rockfish may remain in open waters for several months, being passively dispersed by ocean currents.

Larval rockfish feed on diatoms, dinoflagellates, tintinnids, and cladocerans, and juveniles consume copepods and euphausiids of all life stages. Adults eat demersal invertebrates and small fishes, including other species of rockfish, associated with kelp beds, rocky reefs, pinnacles, and sharp dropoffs. Approximately 50% of adult Canary Rockfish are mature at 14 inches (36 cm) total length (about 5 to 6 years of age). They can live to be 75 years old.

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59 NOAA 2019b. http://www.nmfs.noaa.gov/pr/species/fish/canaryrockfish.htm
60 NOAA Fisheries, West Coast Region 2017.
61 http://www.westcoast.fisheries.noaa.gov/publications/fm/2017/82fr7711.pdf
62 http://www.nmfs.noaa.gov/sfa/fisheries_eco/status_of_fisheries/archive/2015/2015_status_of_stocks_updated.pdf

Distribution and Habitat
Canary Rockfish range between Punta Colonett, Baja California, and the Western Gulf of Alaska. Within this range, Canary Rockfish are most common off the coast of central Oregon.

Canary Rockfish primarily inhabit waters 160 to 820 feet (50 to 250 m) deep, but may be found to 1,400 feet (425 m). Juveniles and sub-adults tend to be more common than adults in shallow water and are associated with rocky reefs, kelp canopies, and artificial structures, such as piers and oil platforms. Adults generally move into deeper water as they increase in size and age, but usually exhibit strong site fidelity to rocky bottoms and outcrops where they hover in loose groups just above the bottom.

Critical Habitat
A final rule designating critical habitat for Yelloweye Rockfish, Canary Rockfish, and Bocaccio was issued by NOAA on February 11, 2015 (79 FR 68041). However, since Canary Rockfish were subsequently de-listed on January 23, 2017, critical habitat is no longer applicable to this species.

Threats within the Project Action Area
Primary stressors to rockfish populations include fishery removals, derelict fishing gear, hypoxia and food web interactions (Palsson et al. 2009). Canary Rockfish are fished directly and are often caught as by-catch in other fisheries, including those for salmon. Adverse environmental factors led to recruitment failures in the early- to mid-1990s. Because this species is slow growing, late to mature, and long-lived, recovery from threats will take many years, even if the threats are no longer affecting the species.

BOCACCIO ROCKFISH (*Sebastes paucispinis*)[63]

Status and Description
Bocaccio Rockfish are Federally-listed as endangered, and are currently State-listed as a candidate species for further status designation at a later time.[64]

Recreational catch and effort data spanning 12 years from the mid-1970s to mid-1990s suggests possible declines in abundance in Washington. Additional data over this period show the number of angler trips increased substantially and the average number of rockfish caught per trip declined. Taken together, these data suggest declines in the population over time. Currently there are no survey data being taken for this species, but few of these fish are caught by fishermen and none have been caught by Washington State biological surveys in 20 years, suggesting a very low population abundance. They are thought to be at less than 10% of their unfished abundance.

A 2005 stock assessment by NOAA Fisheries, however, suggests that Bocaccio have higher populations than was thought to be the case.

Current regulations in Washington State, where the species is most at risk, limit the daily rockfish catch to three rockfish total (of any species, except as noted below in the discussion of Washington State rockfish species of concern).

Rockfishes are unusual among the bony fishes in that fertilization and embryo development is internal, and female rockfish give birth to live larval young. Larvae are found in surface waters, and may be distributed over a wide area extending several hundred miles offshore. Fecundity in female Bocaccio

63 NOAA 2019a. http://www.nmfs.noaa.gov/pr/species/fish/bocaccio.htm
64 WDFW 2008 (updated January 2019).

ranges from 20,000 to more than 2 million eggs, considerably more than many other rockfish species. Larvae and small juvenile rockfish may remain in open waters for several months, being passively dispersed by ocean currents.

Larval rockfish feed on diatoms, dinoflagellates, tintinnids, and cladocerans, and juveniles consume copepods and euphausiids of all life stages. Adults eat demersal invertebrates and small fishes (including other species of rockfish) associated with kelp beds, rocky reefs, pinnacles, and sharp dropoffs. Approximately 50% of adult Bocaccio mature within 4 to 6 years. They are difficult to age but are suspected to live as long as 50 years.

Distribution and Habitat
Bocaccio range from Punta Blanca, Baja California, to the Gulf of Alaska off Krozoff and Kodiak Islands. They are most common between Oregon and northern Baja California. Bocaccio were once common on steep walls in portions of Puget Sound, now they are very rare.

Bocaccio commonly occur at depths between 160 and 820 ft (50 to250 m). Adults generally move into deeper water as they increase in size and age but usually exhibit strong site fidelity to rocky bottoms and outcrops. Juveniles and sub-adults may be more common than adults in shallower water, and are associated with rocky reefs, kelp canopies, and artificial structures, such as piers and oil platforms.

Critical Habitat
A final rule designating critical habitat for Yelloweye Rockfish, Canary Rockfish,[65] and Bocaccio was issued by NOAA on February 11, 2015 (79 FR 68041). Specific locations within five DPS basins have been mapped: the San Juan/Strait of Juan de Fuca basin; Whidbey Basin, Main Basin (areas between Point Wilson near Port Townsend and the Tacoma Narrows), South Puget Sound and Hood Canal. The specific areas in the final designation include 590.4 square miles of nearshore habitat for Bocaccio, and 414.1 square miles of deep-water habitat for Bocaccio and Yelloweye Rockfish. Areas essential to the conservation of Bocaccio include benthic habitats or sites deeper than 98 ft (30 m) that possess or are adjacent to areas of complex bathymetry consisting of rock and or highly rugose habitat. These features support growth, survival, reproduction, and feeding opportunities by providing the structure for rockfishes to avoid predation, seek food, and persist for decades.[66]

Threats within the Project Action Area
Primary stressors to rockfish populations include fishery removals, derelict fishing gear, hypoxia and food web interactions (Palsson et al. 2009). Bocaccio are fished directly and are often caught as by-catch in other fisheries, including those for salmon. Adverse environmental factors led to recruitment failures in the early- to mid-1990s. Because Bocaccio Rockfish are so slow-growing, late to mature, and long-lived, recovery from the above threats will take many years, even if the threats are no longer affecting the species.

[65] No longer applicable to Canary Rockfish; see the Status and Description of Canary Rockfish above.
[66] https://www.federalregister.gov/documents/2014/11/13/2014/endangered-and-threatened-species-designation-of-critical-habitat-for-the-puget-soundgeorgia-basin.

YELLOWEYE ROCKFISH (*Sebastes ruberrimus*)[67]

Status and Description

The Puget Sound/Georgia Basin DPS of Yellow Rockfish in Washington State is Federally-listed as threatened, and State-listed as a candidate species for further status designation at a later time (WDFW 2008, updated January 2019).

Recreational catch and effort data spanning 12 years from the mid-1970s to mid-1990s suggests possible declines in abundance. While catch data are generally constant over time, the number of angler trips increased substantially, and there was a decline in the average number of rockfish caught per trip. Taken together, these data suggest declines in the population over time. Currently there are no survey data being taken for Yelloweye Rockfish, but few of these fish are caught by fishermen, suggesting a low population abundance.

Various Washington State restrictions on fishing have been put in place over the years leading to the 2003 ban on retention of Yelloweye Rockfish. Because this species is slow-growing, late to mature, and long-lived, recovery from these threats will take many years, even if the threats are no longer affecting the species.

Yelloweye Rockfish are very large rockfish that reach up to 3.5 feet (approximately 1 m) in length.

Rockfishes are unusual among the bony fishes in that fertilization and embryo development is internal and female rockfish give birth to live larval young. Larvae are found in surface waters and may be distributed over a wide area extending several hundred miles offshore. Fecundity in female Yelloweye Rockfish ranges from 1.2 to 2.7 million eggs, considerably more than many other rockfish species. Larvae and small juvenile rockfish may remain in open waters for several months being passively dispersed by ocean currents.

Larval rockfish feed on diatoms, dinoflagellates, tintinnids, and cladocerans, and juveniles consume copepods and euphausiids of all life stages. Adults eat demersal invertebrates and small fishes, including other species of rockfish, associated with kelp beds, rocky reefs, pinnacles, and sharp dropoffs. Approximately 50% of adult Yelloweye Rockfish are mature by 16 inches (41 cm) total length (about 6 years of age). They are among the longest-lived of rockfishes, living up to 118 years old.

Distribution and Habitat

Yelloweye Rockfish range from northern Baja California to the Aleutian Islands, Alaska, but are most common from central California northward to the Gulf of Alaska.

Juveniles and sub-adults tend to be more common than adults in shallower water, and are associated with rocky reefs, kelp canopies, and artificial structures such as piers and oil platforms. Adults generally move into deeper water as they increase in size and age, but usually exhibit strong site fidelity to rocky bottoms and outcrops. Yelloweye Rockfish occur in waters 80 to 1,560 feet (25 to 475 m) deep, but are most commonly found between 300 to 590 feet (91 to 180 m).

Critical Habitat

A final rule designating critical habitat for Yelloweye Rockfish, Canary Rockfish,[68] and Bocaccio was issued by NOAA on February 11, 2015 (79 FR 68041). Specific locations within five DPS basins have

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67 NOAA 2019d. http://www.nmfs.noaa.gov/pr/species/fish/yelloweyerockfish.htm

68 No longer applicable to Canary Rockfish; see the Status and Description of Canary Rockfish above.

been mapped: the San Juan/Strait of Juan de Fuca basin; Whidbey Basin, Main Basin (areas between Point Wilson near Port Townsend and the Tacoma Narrows), South Puget Sound and Hood Canal. The specific areas in the final designation include 414.1 square miles of deep-water habitat for yelloweye rockfish and bocaccio. Areas essential to the conservation of Yelloweye Rockfish include benthic habitats or sites deeper than 98 ft (30 m) that possess or are adjacent to areas of complex bathymetry consisting of rock and or highly rugose habitat. These features support growth, survival, reproduction, and feeding opportunities by providing the structure for rockfishes to avoid predation, seek food, and persist for decades.[69]

Threats within the Project Action Area

Primary stressors to rockfish populations include fishery removals, derelict fishing gear, hypoxia and food web interactions (Palsson et al. 2009). Yelloweye Rockfish are fished directly and are often caught as by-catch in other fisheries, including those for salmon. Adverse environmental factors led to recruitment failures in the early- to mid-1990s.

Rockfish are vulnerable to overfishing because many species do not begin to reproduce until they are 5 to 20 years old, and very few of their young survive to adulthood. They are also susceptible to overfishing and habitat degradation as a result of their long life-span.[70]

PINTO ABALONE (*Haliotis kamtschatkana*)[71,72]

Status and Description

Pinto Abalone was Federally-listed as a species of concern in 2004 (WDFW 2008, updated January 2019). WDFW identified Pinto Abalone as a candidate species for further status designation at a later time. WDFW recommends in the April 2019 status report that Pinto Abalone be listed as endangered in Washington State (Carson and Ulrich 2019).

There was no historical commercial fishing for Pinto Abalone in Washington State, and the recreational fishery was closed in 1994 due to declines in abundance. Densities at all but one site are below or within the minimum range for successful fertilization (NOAA/NMFS 2007).

WDFW regularly monitors the abundance of Pinto Abalone at 10 index stations throughout the San Juan Archipelago. Because Pinto Abalone are highly patchy, cryptic and frequently associate with microhabitats such as rock crevices or patches of coralline algae that may themselves be sparsely distributed, total abundances are not measured. Repeated surveys at a system of index sites are conducted in order to detect temporal trends in abalone abundance. These surveys indicate an 83% decline in abalone abundance between 1992 and 2009 (Rothaus et al. 2008).

Known for their large, muscular foot and the pearlescent oval shell, Pinto Abalone are slow-growing, long-lived marine snails. Pinto (or Northern) Abalone is the only species found in Washington State.

Rothaus et al. (2008) found an increase in mean shell length of 10.4 mm between 1992 and 2006, indicating a substantial shift in the size distribution of abalone populations. This signifies a shift in abalone population age structure from younger to older animals, indicative of repeated recruitment

[69] https://www.federalregister.gov/documents/2014/11/13/2014/endangered-and-threatened-species-designation-of-critical-habitat-for-the-puget-soundgeorgia-basin.
[70] https://www.westcoast.fisheries.noaa.gov/protected_species/rockfish/rockfish_in_puget_sound
[71] NOAA/NMFS. 2007. https://www.westcoast.fisheries.noaa.gov/publications/SOC/pintoabalone_detailed.pdf
[72] Encyclopedia of Puget Sound. https://www.eopugetsound.org/science-review/2-pinto-abalone

failure. Many aspects of abalone biology and ecology not yet well understood may be important in explaining both the decline and the recovery potential for Pinto Abalone in the Puget Sound region (Encyclopedia of Puget Sound).

Since 2004, a program of hatchery-based rearing and out-planting aimed at restoring abalone populations in Washington State has been led by the Puget Sound Restoration Fund and several local partners. In the summer of 2000, nearly 2,000 abalone were out-planted near Anacortes and Port Angeles (Encyclopedia of Puget Sound).

Pinto Abalone broadcast spawn from April to June. The larvae are planktonic and settle after approximately 7 to 10 days in response to cues from both crustose coralline algae and from adults. Limited larval dispersal seems to make abalone species in general difficult to manage. There may be an unusual genetic form in the Puget Sound/Strait of Georgia region (Jamieson 1999).

Pinto Abalone are important herbivores in nearshore habitats, feeding primarily on drift macroalgae such as kelp and benthic diatom films. They can structure subtidal communities through the maintenance of substrata dominated by crustose coralline algae and through the facilitation of conspecific settlement.

Distribution and Habitat

Pinto Abalone were once widely distributed throughout the waters of British Columbia and Washington State. In recent decades, populations have undergone sharp declines, likely in response to the combined stressors of overharvest, poaching, and sub-optimal environmental conditions (Campbell 2000).

Pinto Abalone are typically found in nearshore rocky habitats in semi-exposed or exposed coastal regions. They typically occupy the low intertidal zone to a depth of 30 ft (9 m), but may be found as deep as 330 feet (100 m). In Washington State, they range from Admiralty Inlet to the San Juan Islands and the Strait of Juan de Fuca, and are typically found at depths of about 60 feet (20 m) (Buoma 2007).

Representative Pinto Abalone habitat at the WDFW San Juan Archipelago index station sites is primarily comprised of bedrock and boulders encrusted with coralline algae, supporting assemblages of kelp and other macroalgae (Rothaus et al. 2008).

Threats within the Project Action Area

Threats to the population success of Pinto Abalone include overharvest, suspected illegal harvest, and predation by reintroduced and recovering sea otter (*Enhydra lutris*).

E. STATE-LISTED SPECIES AND CANDIDATE SPECIES[73]

WDFW provided the table below to identify Washington State-listed species and candidates for listing to be addressed in the update of the 1990 *Programmatic EIS: Fish Culture in Floating Net Pens*, and in the SEPA analysis of the Cooke Aquaculture Puget Sound net pen species conversion proposal from rearing Atlantic Salmon to rearing all-female triploid Rainbow Trout/steelhead. The proposed action, project action area, and elements of the effects analysis are the same for State-listed species as described in Section A preceding the effects analysis for Federally-listed species. Information provided in this section about each State-listed marine mammal, fish and mollusc includes species status and description, distribution and habitat, and threats within the project action area. The effects of Atlantic Salmon marine net pen aquaculture, and of the Cooke Aquaculture species conversion proposal to rear all-female triploid

[73] Some State-listed species are described above in Section D, Federally-Listed Species and Species of Concern.

Rainbow Trout/steelhead in the company's existing marine net pen facilities, is provided in Section F for both State-listed and Federally-listed species.

Common Name	Animal Type	State Species Status
Pacific Harbor Porpoise	Mammals	Candidate
Gray Whale	Mammals	Sensitive
Pacific Herring	Fish	Candidate
Black Rockfish	Fish	Candidate
Brown Rockfish	Fish	Candidate
China Rockfish	Fish	Candidate
Copper Rockfish	Fish	Candidate
Quillback Rockfish	Fish	Candidate
Redstripe Rockfish	Fish	Candidate
Tiger Rockfish	Fish	Candidate
Yellowtail Rockfish	Fish	Candidate
Walleye Pollock (South Puget Sound)	Fish	Candidate
Olympia Oyster	Molluscs	Candidate

PACIFIC HARBOR PORPOISE (*Phocoena phocoena*)[74]

Status and Description

The Washington Department of Fish and Wildlife lists Pacific Harbor porpoises as a "candidate" species (WDFW 2015). This status refers to species that are to be reviewed by the State for possible listing as endangered, threatened, or sensitive. NOAA has not listed harbor porpoises, but they are protected under the Marine Mammal Protection Act.

Harbor Porpoise may weigh up to 170 pounds (77 kg) and measure 5 feet in length. Their diet includes schooling fish (e.g., herring, capelin) and invertebrates (e.g., cephalopods). The species general travels in small groups of one to eight individuals. Sexual maturity is reached in 3 to 4 years, and females may give birth every year for several years in a row. Lifespan is approximately 24 years.

Distribution and Habitat

In the eastern North Pacific Ocean, Harbor Porpoise range from Point Barrow in northern Alaska south to Point Conception in California. The species is distributed in coastal and inland waters. Harbor Porpoise are known to occur year-around in the Strait of Juan de Fuca and Puget Sound (NOAA 2011a). The species is most commonly found in bays, estuaries and harbors less than 650 feet (200m) deep.

Threats within the Project Action Area

The 2003 estimated abundance of the inland Washington waters stock of Harbor Porpoise was 10,682 individuals, with a minimum population estimate of 2,545 (NOAA 2011a). There is no more recent abundance estimate available for this stock, and there are no reliable data on long-term population trends on the Harbor Porpoise.

[74] RPS ASA 2016.

Threats to Harbor Porpoise include by-catch in fishing gear (particularly gillnets, trawls, and herring weirs) and boat strikes. Additionally, Unusual Mortality Events (UME) have been recorded for this species in Puget Sound, including 114 strandings in 2006 (NOAA 2011a).

GRAY WHALE (*Eschrichtius robustus*)[75]

Status and Description

The Washington Department of Fish and Wildlife lists Gray Whale as "sensitive" (WDFW 2008, updated January 2019). This status refers to species that are vulnerable or declining, and likely to become endangered or threatened in significant portions of their range if threats persist or if cooperative management does not occur. While the Western Pacific population of Gray Whale is Federally-listed as endangered, the U.S. Fish and Wildlife Service delisted the Eastern Pacific population in 1994; therefore, it does not currently have a Federal listing status. However, Gray Whale, like all marine mammals, are protected under the Marine Mammal Protection Act (MMPA).

Gray Whale roll on their sides and swim slowly along the sea floor to feed. They filter amphipods from bottom sediments using their baleen plates. They reach sexual maturity at an average of 8 years old and give birth to single calves after 12 to 13 months of gestation.

Distribution and Habitat

Gray Whale are found mainly in shallow coastal waters in the North Pacific Ocean. Genetically distinct Eastern North Pacific (ENP) and Western North Pacific (WNP) populations are recognized. Most of the ENP population feeds in the Chukchi and Bering Seas, but some gray whales have also been reported feeding along the Pacific coast during the summer, in waters off of Southeast Alaska, British Columbia, Washington, Oregon, and northern California. In the fall, gray whales migrate south along the coast of North America from their summer feeding grounds to winter bear calves in areas off the coast of Baja California. The most recent population estimate of the ENP is about 27,000 whales (NOAA 2019e).

Threats within the Project Action Area

General nearshore threats to Gray Whale include vessel collisions, entanglement in fishing gear, and disturbance from ecotourism, noise, and whale-watching. Collisions with all sizes and types of vessels are one of the primary threats to marine mammals, particularly large whales. Gray whales are one of the most vulnerable species to vessel strikes because they feed and migrate along the U.S. west coast, which has some of the world's heaviest vessel traffic associated with some of the largest ports in the country.

Entanglement in fishing gear can result in fatigue, compromised feeding ability, or severe injury, which would ultimately lead to death.

Underwater noise threatens whale populations, interrupting their normal behavior and driving they away from areas important to their survival.

PACIFIC HERRING (*Clupea pallasii*)[76]

Status and Description

Pacific Herring are State-listed in Washington as a candidate species for further status designation at a later time.

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[75] https://www.fisheries.noaa.gov/species/gray-whale

[76] https://www.fisheries.noaa.gov/Alaska/endangered-species-conservation/pacific-herring.

Distribution and Habitat
Pacific Herring is a coastal schooling species found on both the eastern and western sides of the Pacific Ocean at depths ranging from the surface to 1,300 ft (400 m). Adult Pacific Herring migrate inshore, entering estuaries to breed once per year, with timing varying by latitude. Herring spawn in shallow areas along shorelines, between the subtidal and intertidal zones. Eggs are deposited on kelp, eelgrass (*Zostera marina*), and other available structures. Herring feed on phytoplankton and zooplankton in nutrient-rich wasters associated with oceanic upwelling.

Threats within the Project Action Area
Threats for Pacific Herring include destruction of herring spawning grounds, juvenile and feeding habitat and rearing/foraging habitat. None of these habitat requirements occur directly within areas where Cooke Aquaculture existing marine net pens are located. Global climate change, recovering populations of predators and fishing exploitation in Southeast Alaska are also threats to Pacific Herring.

BLACK ROCKFISH (*Sebastes melanops*)[77]

Status and Description
Black Rockfish are State-listed as a candidate species for further status designation at a later time. Recreational harvest within Puget Sound has been closed, with the exception of restricted fishing in the Strait of Juan de Fuca. They can grow up to approximately 27 inches (69 cm) in length, and weigh up to 11 pounds (5 kg). Maximum age is 50 years old.

Distribution and Habitat
Black Rockfish range from Amchitka and Kodiak Islands, Alaska, to Huntington Beach in southern California. They have been found at water depths up to 1,200 feet (366 m), but are most commonly found in waters shallower than 180 feet (55 m). Black Rockfish are known to form large schools in and around kelp and artificial structures. They are commonly caught by recreational harvesters off the Washington coast.

Threats within the Project Action Area
Primary stressors to rockfish populations include fishery removals, derelict fishing gear, hypoxia and food web interactions (Palsson et al. 2009). Rockfish species are often caught as by-catch in other fisheries. Because they are slow growing, late to mature, and long-lived, recovery from threats take many years, even if the threats are no longer affecting the species.

BROWN ROCKFISH (*Sebastes auriculatus*)[78]

Status and Description
Brown Rockfish are State-listed as a candidate species for further status designation at a later time. Recreational harvest within Puget Sound has been closed. In Puget Sound, the apparent hybridization of Quillback, Copper and Brown Rockfish makes species identification difficult. Brown Rockfish can grow up to 22 inches (56 cm) in length. Maximum age is at least 34 years old.

Distribution and Habitat
Brown Rockfish range from Prince William Sound, Alaska, to southern Baja California. They were once abundant in central and South Puget Sound. They range in depth from shallow inshore waters to approximately 400 feet (135 m) deep. They are most commonly distributed above 400 feet (120 m).

[77] WDFW 2019.
[78] WDFW 2019.

Brown Rockfish can be found a few meters off the bottom, and are common on both low and high relief areas, and occasionally within eelgrass or other marine vegetation. They are rarely caught by recreational harvesters off the Washington coast.

Threats within the Project Action Area
Primary stressors to rockfish populations include fishery removals, derelict fishing gear, hypoxia and food web interactions (Palsson et al. 2009). Rockfish species are often caught as by-catch in other fisheries. Because they are slow growing, late to mature, and long-lived, recovery from threats take many years, even if the threats are no longer affecting the species.

CHINA ROCKFISH (*Sebastes nebulosus*)[79]

Status and Description
China Rockfish are State-listed as a candidate species for further status designation at a later time. Recreational harvest within Puget Sound has been closed. They can grow up to 18 inches (45 cm) in length and have a relatively small mouth. Maximum age is at least 79 years old.

Distribution and Habitat
China Rockfish are found from Kechemak Bay, Cook Inlet, Alaska, to San Nicolas Island in southern California. They are found at water depths between approximately 10 and 420 feet (3 and 128 m). This is a solitary species inhabiting high-energy, high relief rocky outcrops with numerous crevices. They are very territorial and rarely move more than about 30 feet (10 m) from their home site. China Rockfish are commonly caught by recreational harvesters off the northern Washington coast.

Threats within the Project Action Area
Primary stressors to rockfish populations include fishery removals, derelict fishing gear, hypoxia and food web interactions (Palsson et al. 2009). Rockfish species are often caught as by-catch in other fisheries. Because they are slow growing, late to mature, and long-lived, recovery from threats take many years, even if the threats are no longer affecting the species.

COPPER ROCKFISH (*Sebastes caurinus*)[80]

Status and Description
Copper Rockfish are State-listed as a candidate species for further status designation at a later time. Recreational harvest within Puget Sound has been closed. Hybridization between Brown, Copper and Quillback Rockfish occurs in Puget Sound, sometimes making species identification difficult. Copper Rockfish can grow up to approximately 26 inches (66 cm) in length. Males grow larger than females. Maximum age is at least 50 years old.

Distribution and Habitat
Copper Rockfish range from the northern Gulf of Alaska to Isla San Benito, near the center of Baja California. They are found from subtidal waters to about 600 feet (183 m) in depth. Adults are found primarily in boulder fields and over high-relief rocks, either in schools or as single individuals. They are commonly caught by recreational harvesters off the northern Washington coast and occasionally by recreational fishermen in the Strait of Juan de Fuca.

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[79] WDFW 2019.
[80] WDFW 2019.

Threats within the Project Action Area

Primary stressors to rockfish populations include fishery removals, derelict fishing gear, hypoxia and food web interactions (Palsson et al. 2009). Rockfish species are often caught as by-catch in other fisheries. Because they are slow growing, late to mature, and long-lived, recovery from threats take many years, even if the threats are no longer affecting the species.

QUILLBACK ROCKFISH (*Sebastes maliger*)[81]

Status and Description

Quillback Rockfish are State-listed as a candidate species for further status designation at a later time. Recreational harvest within Puget Sound has been closed. Hybridization between Brown, Copper and Quillback Rockfish occurs in Puget Sound, sometimes making species identification difficult. Quillback Rockfish can grow up to approximately 24 inches (61 cm) in length. Maximum age is 95 years old.

Distribution and Habitat

Quillback Rockfish range from Kenai Peninsula in the Gulf of Alaska to Anacapa Passage in southern California. They can be found from subtidal waters to a depth of 900 feet (approximately 275 m). Juveniles are usually shallower than adults, and can be found on bull kelp-covered rocky outcrops, while adults tend to live in deeper water as solitary individuals. They are bottom dwellers that prefer high-relief, broken rock with flat-bladed kelps. In Puget Sound, Quillback Rockfish living on high-relief areas have a very limited home range and have high fidelity to their home sites. They are commonly caught off the Washington coast by recreational harvesters and occasionally caught by recreational fishermen in the Strait of Juan de Fuca.

Threats within the Project Action Area

Primary stressors to rockfish populations include fishery removals, derelict fishing gear, hypoxia and food web interactions (Palsson et al. 2009). Rockfish species are often caught as by-catch in other fisheries. Because they are slow growing, late to mature, and long-lived, recovery from threats take many years, even if the threats are no longer affecting the species.

REDSTRIPE ROCKFISH (*Sebastes proriger*)[82]

Status and Description

Redstripe Rockfish are State-listed as a candidate species for further status designation at a later time. Recreational harvest within Puget Sound has been closed. They can grow up to approximately 20 inches (51 cm) in length. Maximum age is at least 55 years old.

Distribution and Habitat

Redstripe Rockfish range from the Bering Sea and Amchitka Island, Alaska, to southern Baja California. They are found at water depths from 40 to 1,400 feet (12 to 425 m), and are most commonly found between approximately 500 and 900 feet (150 to 275 m). This species usually lives over high-relief, rugged bottoms, and may form dense schools that rise off the bottom during the day and disperse at night. They are occasionally caught off the Washington coast by commercial otter-trawls and longline gear. They are rarely caught by recreational fishermen within Puget Sound.

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81 WDFW 2019.

82 WDFW 2019.

Threats within the Project Action Area

Primary stressors to rockfish populations include fishery removals, derelict fishing gear, hypoxia and food web interactions (Palsson et al. 2009). Rockfish species are often caught as by-catch in other fisheries. Because they are slow growing, late to mature, and long-lived, recovery from threats take many years, even if the threats are no longer affecting the species.

TIGER ROCKFISH (*Sebastes nigrocinctus*)[83]

Status and Description

Tiger Rockfish are State-listed as a candidate species for further status designation at a later time. Recreational harvest within Puget Sound has been closed. They can grow up to 24 inches (61 cm) in length. Maximum age is 116 years old.

Distribution and Habitat

Tiger Rockfish range from Kodiak Island and Prince William Sound, Alaska, to Tanner and Cortes Banks in southern California. They occur at water depths between approximately 60 and 1,000 feet (18 to 298 m). Adult Tiger Rockfish live on rock outcrops that have caves and crevices. They are rarely observed in the open during the day. They are occasionally caught off the Washington coast by commercial harvesters using otter-trawls and longline gear. They are rarely caught by recreational fishermen in Puget Sound.

Threats within the Project Action Area

Primary stressors to rockfish populations include fishery removals, derelict fishing gear, hypoxia and food web interactions (Palsson et al. 2009). Rockfish species are often caught as by-catch in other fisheries. Because they are slow growing, late to mature, and long-lived, recovery from threats take many years, even if the threats are no longer affecting the species.

YELLOWTAIL ROCKFISH (*Sebastes flavidus*)[84]

Status and Description

Yellowtail Rockfish are State-listed as a candidate species for further status designation at a later time. Recreational harvest within Puget Sound has been closed. Under water, Yellowtail Rockfish resemble Olive Rockfish, making identification somewhat difficult. Yellowtail Rockfish can grow up to approximately 26 inches (66 cm) in length. Maximum age is at least 64 years old.

Distribution and Habitat

Yellowtail Rockfish range from Unalaska Island, Alaska, to San Diego, California. Only juvenile yellowtails have been found in Puget Sound. Older juveniles and adults are usually found over high relief, such as boulders and sheer rock walls, although they are seen rarely over cobble-mud bottoms. Yellowtail Rockfish are a schooling species, sometimes swimming well off the bottom in schools of thousands. They can be found from the surface to approximately 1,800 feet (549 m) in water depth. They are commonly caught by commercial harvesters off the Washington coast, and occasionally caught in the Strait of Juan de Fuca by recreational fishermen.

Threats within the Project Action Area

Primary stressors to rockfish populations include fishery removals, derelict fishing gear, hypoxia and food web interactions (Palsson et al. 2009). Rockfish species are often caught as by-catch in other fisheries.

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[83] WDFW 2019.

[84] WDFW 2019.

Because they are slow growing, late to mature, and long-lived, recovery from threats take many years, even if the threats are no longer affecting the species.

SOUTH PUGET SOUND WALLEYE POLLOCK (*Theragra chalcogramma*)[85]

Status and Description
Walleye Pollock are State-listed as a candidate species for further status designation at a later time.

Walleye Pollock adults are generally a semi-demersal species. Quinnell and Schmidtt (1991) found that the mean length of Walleye Pollock collected in North Puget Sound was approximately 5.5 inches (14 cm), suggesting they were largely young-of-the-year. Walleye Pollock collected in South Puget Sound had a mean length of 6.3 inches (16 cm), which suggests the presence of a spawning population in or near South Puget Sound (Gustafson et al. 2000). Early-stage Walleye Pollock feed on copepod nauplii (Nakatani 1988, Canino et al. 1991, Gustafson et al. 2000), and juveniles mostly feed on euphausiids, copepods, decapod[86] larvae, and larvaceans[87] (Grover 1990, Merati and Brodeur 1996, Bailey et al. 1999, Gustafson et al. 2000). Predators of Walleye Pollock eggs and larvae include a variety of invertebrates and fish. Juvenile Walleye Pollock are preyed upon by seabirds (e.g., common murre) and marine mammals (e.g., harbor seals) (Bailey et al. 1999, Hunt et al. 1996, Lowry et al. 1996, Gustafson et al. 2000).

Distribution and Habitat
Various life stages of Walleye Pollock inhabit nearshore areas, large estuaries (including Puget Sound), coastal embayments and open ocean basins. Adults occur as deep as 1,160 feet (366 m), but the vast majority occurs in depths between 330 and 980 feet (100 to 300 m). Juvenile pollock have been found in a variety of habitat types, including eelgrass (over sand and mud), gravel and cobble; however, because of their pelagic mode, they are not thought to consistently associate with many types of substrates. The nearest known spawning ground to the project action area is to the east in Port Townsend, Jefferson County (Gustafson et al. 2000).

Threats within the Project Action Area
The threats for Walleye Pollock are similar to those for Pacific Cod and include destruction of herring spawning grounds, juvenile and feeding habitat, and rearing/foraging habitat, which do not directly occur within the project action area. Global climate change, recovering populations of predators, and fishing exploitation are additional identified threats.

OLYMPIA OYSTER (*Ostrea lurida*)

Status and Description
Olympia Oyster are State-listed as a candidate species for further status designation at a later time (WDFW 2008, updated January 2019).

Native populations of Olympia Oyster were initially decimated by human harvest. By 1870, overharvesting had depleted native oyster stocks in both Willapa Bay and Puget Sound. Olympia. Oyster populations were maintained at low levels by water pollution, particularly effluent discharge from pulp

[85] RPS ASA 2016.
[86] Decapods are an order of crustaceans including many familiar groups, such as crayfish, crabs, lobsters, prawns and shrimp.
[87] Larvaceans are solitary, free-swimming tunicates (transparent marine invertebrates) found throughout the world's oceans.

and paper mills that severely limited reproductive success extending into the 1970s. Non-native oyster predators, particularly the Japanese oyster drill (*Ocenebra japonica*) and a parasitic crustacean (*Mytilicola orientalis*) were accidentally introduced along with the Pacific oysters early in the 1900s (Pacific Biodiversity Institute 2019).

Olympia Oysters usually inhabit low tidelands that remain inundated during low tides. Improving water quality over the past 40 years with the closure of all but two of pulp mills in Puget Sound, and recent restoration efforts, have failed to restore Olympia Oyster populations.

Distribution and Habitat

The historic range of the Olympia Oyster extended along the coast of North America from southeast Alaska to Baja California. They usually inhabit low tidelands or estuaries that remain inundated with water during low tide, although they can also be found on anthropogenic surfaces such as the undersides of floats an on pilings (Pacific Biodiversity Institute 2019).

Native Olympia Oyster beds created complex, three-dimensional habitat and foraging locations for invertebrates and fish, including salmonids and Dungeness crab (*Metacarcinus magister*). Olympia Oyster are filter feeders that filter phytoplankton, bacteria, excess nutrients, pathogens, and other pollutants from the water at a rate of 5 gallons per hour. Historic Olympia Oyster beds would thereby provide a substantial clean water benefit and provide the water quality necessary to allow for healthy eelgrass and kelp growth (Northwest Straits Commission. 2019, Blake and Bradbury 2013, Pacific Biodiversity Institute 2019).

Overall, Olympia Oyster are thought to occur throughout the extent of their historic range in low densities. In the Pacific Northwest, Olympia Oyster populations have been classified as poor or functionally extinct, with 90 to 99% of the population lost. Less than 4% of the historic core populations remain in Salish Sea (Peabody and Davis 2013).

Threats within the Project Action Area

According to the 2013 WDFW report on rebuilding Olympia Oyster populations in the Salish Sea (Blake and Bradbury 2013), this species is sensitive to the presence of non-native, invasive predators, including the Japanese oyster drill (*Ocinebrellus inornatus*); shoreline and tideland modifications, including nearshore or estuarine restoration projects; by-catch mortality from Pacific Oyster commercial harvest; turbidity, siltation, and increased nutrient inputs from upland practices; and genetic fitness impacts from unrestricted distribution of generic hatchery-origin native oysters.

Extreme temperatures from storm and freezing events that exceed the tolerance range of Olympia Oyster, and water pollution in the form of heavy metals, oils, pesticides and other chemicals may also impact the recovery of Olympia Oyster (Capital Regional District 2019).

The recovery and reestablishment of historic levels of the Olympia Oyster is restricted by a lack of suitable habitat where beds once occurred. The lack of habitat is mostly due to presence of eelgrass or aquaculture in tideland areas, loss of tidelands due to filling or diking, and the diminished densities that would allow for effective reproduction (Blake and Bradbury 2013).

F. EFFECTS ANALYSIS

The potential effects of floating net pen culture on Federally-listed species and State-listed species and candidate species are described in this section based on features and operations of Cooke Aquaculture existing marine net pens in Puget Sound in which primarily Atlantic Salmon have been raised since the 1980s. These effects have been described in hundreds of technical studies performed by Federal agencies and independent researchers, some of which are cited here. Other than transitioning to the commercial cultivation of a different species of fish, the company is not planning any alteration to the existing fish pen physical structures, site locations, supporting equipment, or general current practices, methods and cultivation techniques used for growing Atlantic salmon in net pens.

The effects analysis in this section addresses two scenarios for each species: the effects of Atlantic Salmon net pen culture (to update the 1990 Programmatic EIS), followed by the effects of the species conversion proposal to transition the species reared in Cooke Aquaculture net pens from Atlantic Salmon to all female triploid Rainbow Trout/steelhead. Concerns about the potential effects of escapement of domesticated stocks of mono-sex sterile Rainbow Trout/steelhead are discussed in SEPA Checklist Attachment B, Additional Information.

Features and Operations of Cooke Aquaculture Floating Net Pens Considered in the Effects Analysis of Threatened and Endangered Species, Candidate Species, and Species of Concern

Fish Containment (Stock Nets) and Predator-Exclusion Nets. Cooke Aquaculture floating marine net pens use three different net types:

- Fish containment or stock nets are the primary containment net the fish are reared in. The net extends from above the surface of the water (approx., 5 feet) where it is attached to the hand railing and walkway attachment points around the inside perimeter of each pen. The net then extends below the surface from approximately 25 to 50 feet, where it is attached to a heavy pipe frame weighting system. The stock nets are made of a strong nylon/polypropylene blend material approximately one-eighth inch in diameter and the webbing is approximately 1-inch mesh on the square. The containment nets are pulled taught by a weighted pipe frame that helps to keep the shape of the growing environment uniform in tidal currents, and to maintain the physical separation between the fish growing nets and the predator exclusion barrier netting. Fish containment nets have high visibility due to their bright colors and thick twine diameters (compared to gillnet monofilament, for example). Following harvest, the containment nets are lifted from each net pen site to the surface and then transported by boat to a land-based facility for cleaning and repairs. The predation exclusion or barrier net are either removed or maintained in place between generations. The same procedure would occur if there was a scheduled removal of the predator barrier net (lifted to the surface at the pens, put onto a vessel and then transported on the deck of a boat to a shore facility for unloading and shipping to a net maintenance facility). None of the nets are towed through the water, to or from shore, when they are being transported.
- Marine mammal predator exclusion/barrier nets (4-inch mesh on the square with a twine diameter of one-quarter inch) are attached around the outside perimeter of the entire net pen facility walkway and to the pipe weight frame below the surface (varies by site from approximately 30 to 60 feet deep). The predation barrier net surrounds the entire net pen structure and is lashed to the subsurface pipe weight frame, creating a rigid wall and floor panel that protects the fish from seals and sea lions attacks and reaching the fish through the barrier. The predation net is attached to poles that extend approximately 5 to 6 feet above the surface of the water around the entire perimeter of the net pen system. This jump fence keeps marine mammals such as seals and sea

lions from jumping onto the net pen floats and accessing the fish containment nets located on the inside perimeter of the walkway. Like the fish containment nets, predator exclusion nets are stretched taught with a heavy steel pipe frame that maintains the distance (approximately 4 to 8 feet) between the containment net and barrier net. These nets are also made of brightly-colored, thick-diameter twine that provides high visibility.

- Avian exclusion nets (2-inch mesh on the square with a twine diameter of one-eighth inch) are tied securely to the inner perimeter hand railing of each fish containment pen and stretched tightly across the top of the net, 4 feet high at the point of attachment to the hand railings. These nets are used primarily to keep gulls and scavenger birds from trying to reach the fish food pellets as they are being distributed to the surface of each fish pen while the fish are being fed.

Stocking, Grow-out and Harvest. Fish rearing units (farm sites) are planted with a single-stock (generation) of cultured smolts delivered from a company-owned freshwater hatchery near Olympia. The all-female triploid Rainbow Trout/steelhead smolts would be fed and reared in the marine net pens for approximately 12 to 18 months before reaching their targeted harvest weight. The fish are harvested and transported by a marine fishing vessel directly to an existing land-based fish processing facility. Harvesting a generation of fish would take approximately 2 to 6 months to complete depending on growing conditions, market demands and other operational factors. Once the fish stock has been completely harvested out, the marine net pen facility would be fallowed (no fish stocks on site) for a minimum of 30 days from the date that the last fish containment net was removed from the farm facility. Fish containment nets are removed by boat and transported to an approved upland facility for routine cleaning and maintenance repairs. The clean nets would then be reinstalled into the net pens and the site is prepared for restocking with the next generation of smolts.

Feeding and Fish Health Maintenance. Cultivated fish stocks are fed daily using mechanical feeding equipment. Detailed records are kept on feeding behavior, fish mortalities and the daily food consumption within each pen. Trained staff feed the fish, and fish health technicians conduct routine inspections of the fish to monitor the health status of the fish stocks. Size and temperature dependent feeding rates and digestion rates for the most efficient feed conversion into fish biomass growth has been extensively researched by the aquaculture industry and are incorporated into the feed management strategy of the farm sites. Feed usage is tracked daily and tracked in a database that calculates expected growth rates. Periodic fish weight samples are used to verify growth rates and feed conversion rates are following projected biological budgets. Underwater cameras in each pen allow technicians to closely monitor feeding behavior and feeding technicians adjust the feed delivery rate to meet the specific appetite of each fish pen population and to minimize loss of uneaten feed.

Routine diving and fish mortality removal occurs at each fish farm each week. Fish mortalities removed from the pens are transferred to sealed containers (plastic fish totes) and are transported to an approved, land-based top-soil composting facility. The plastic fish mortality totes are cleaned and disinfected at the off-site composting facility before being returned to the individual farms. Mortalities are recorded and checked carefully by trained fish health technicians to determine and document the cause of mortality. Should a fish disease occur within a net pen operation, treatment may be carried out using medicated fish feed. Veterinary guidelines are closely followed to ensure proper treatment and dosage to the fish population. Medicated feed usage (if any) is recorded in the feed database, and a monthly summary report of feed usage (including any medicated feed) is sent to the Washington Department of Ecology (Ecology), Washington Department of Fish and Wildlife and the Washington Department of Natural Resources.

Water Quality and Sediment Quality Compliance. Since 1996, Ecology has required marine net pen aquaculture operations to obtain a National Pollutant Discharge Elimination System (NPDES) Individual permit for commercial net pen facilities within the State. This Clean Water Act permit requires monitoring, reporting, operational guidelines, Best Management Practices (BMPs), and Best Available Technology (BAT) for the operation of a facility to minimize pollution. NPDES measures are in addition to the monitoring and reporting requirements of other permits.

A key component of the NPDES permit is definition of a Sediment Impact Zone (SIZ) and Total Organic Carbon (TOC) monitoring. Organic carbon compounds are the main discharge of nutrients from a salmon net pen operation. The NPDES permit requires that organic carbon levels of the sediments sampled at the 100-ft perimeter around a net pen facility (the SIZ boundary) meet sediment management standards and be the same as or less than the natural organic carbon levels that are found in sediments from Puget Sound reference locations. These sediment standards are set for the protection of benthic organisms and the surrounding marine environment. Sampling is carried out by a third-party consultant, and the analysis is conducted by a certified, independent laboratory. If a farm is out of balance with the natural assimilative capacity of nutrients by the surrounding benthic environment, active mitigation is required. These measures would include additional TOC monitoring, a possible reduction in feeding amounts, a reduction in fish stocking biomass, further fallowing of the site to allow additional sediment recovery time, or possible reorientation/relocation of the site into more oxygenated waters, or the complete removal and closure of the site if remediation measures do not cure the exceedance of sediment standards.

NPDES permits are renewed and updated every 5 years based on the most current Ecology regulations. Specific conditions, restrictions, and monitoring and reporting procedures incorporated into each Individual permit are designed to protect public health and safety, and to meet water and sediment quality standards protective of the environment. NPDES Individual permits for the Cooke Aquaculture Atlantic Salmon net pen facilities at Clam Bay, Fort Ward, Orchard Rocks, and Hope Island were recently reissued (July 11, 2019), conditionally authorizing discharges associated with commercial rearing of Atlantic Salmon, subject to closing out non-native fish rearing after the Washington Department of Natural Resources (WDNR) State-owned Aquatic Land leases expire in 2022. The renewed permits indicate that if the company elects to raise native fish at these locations in the future, Ecology will consider a permit modification and may apply the discharge limits and requirements of the existing NPDES permits to native finfish. The reissued permits become effective on August 10, 2019 and expire on August 9, 2024.

Potential Effects on Federally-Listed Species and Species of Concern

Species are listed below in the same order in which they are described above in Sections D and F, with Federally-listed species described first, followed by State-listed and State candidate species. The effects analysis for existing Atlantic Salmon floating net pen culture on Federally-listed species and species of concern has been summarized from various sources, for the purpose of updating the *Final Programmatic EIS: Fish Culture in Floating Net Pens* (WDF 1990). These paragraphs have been reviewed and edited by Don Weitkamp, Ph.D., Aquatic Biologist and Water Quality Expert, who was one of the original authors of the 1990 Programmatic EIS. Dr. Weitkamp is also the author of all statements of effect (below) for the Cooke Aquaculture proposal to convert the species reared in existing marine net pens from Atlantic Salmon to sterile (triploid) all-female Rainbow Trout/steelhead. Don's curriculum vitae (CV) is provided in SEPA Checklist Attachment E, along with the CV for Dr. Walton Dickhoff who also reviewed and contributed to the analysis of potential effects of the Cooke Aquaculture species conversion proposal in the Additional Information document (SEPA Checklist Attachment B).

HUMPBACK WHALE

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture[88]

Humpback Whale are not often found in Washington, especially within Puget Sound. They are more common off the Pacific Coast of Washington, which is a primary migratory corridor. Existing Cooke Aquaculture Puget sound marine finfish rearing facilities do not impact the major migratory corridor of Humpback Whale since there are no existing floating marine net pens on Washington's Pacific Coast. In addition, Humpback Whale do not rely heavily on benthic feeding, so effects to the benthic environment are considered minimal. Therefore, Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Humpback Whale (NMFS 2011).

Potential Effects from the Proposed Net Pen Species Conversion

Conversion of the net pen facilities from Atlantic Salmon to triploid all-female Rainbow Trout/steelhead will not change significant threats to Humpback Whales through entanglement in fishing gear, collisions with ship traffic, pollution, or any other means. Therefore, the action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Humpback Whales.

SOUTHERN RESIDENT KILLER WHALE

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture[89]

Southern Resident Killer Whale (SRKW) are regular inhabitants of Puget Sound. Marine net pens are insignificant in their overall size and are therefore not expected to impact SRKW habitat. Vessels that service these facilities may cause short-term and localized disturbances but are not expected to have any lasting effects. There is adequate space to accommodate SRKW passage around the existing net pen facilities so that any effects on passage are expected to be insignificant (Thom 2010).

No outbreaks of parasites related to net pen operations have been observed in Puget Sound. Therefore, NMFS concludes that the operation of net pen facilities would have insignificant and discountable effects on salmonids regarding sea lice infestation. In addition, NMFS anticipates discountable effects on prey quality because net pen operators comply with NPDES permit requirements related to maintaining water quality and sediment quality. A Not Likely to Adversely Affect (NLAA) determination was supported for listed salmonids in Puget Sound; therefore, SRKW are also not likely to be adversely affected since salmonids are a primary prey base (NMFS 2011).

Other than limited and non-lethal predator control permitted by NMFS, the technical memorandums do not state any concerns of adverse effects to marine mammals in Puget Sound in relation to Atlantic Salmon rearing facilities (Nash 2001). Furthermore, the Washington Pollution Control Hearings Board (PCHB) specifically noted in its 1997 ruling that the operation of net pen facilities in Puget Sound does not have a negative impact on marine mammals (Ecology 2007). Therefore, Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Southern Resident Killer Whale.

NMFS also concurs with the EPA effect determination of May Affect, Not Likely to Adversely Affect SRKW critical habitat. Existing Cooke Aquaculture Puget Sound marine net pens are required to comply with Washington State water quality standards through NPDES permit compliance. NMFS anticipates effects on prey quantity and quality will be discountable or insignificant within the action area and within designated critical habitat of SRKW. And the potential for vessels or the net pens themselves to interfere with passage

[88] USEPA 2010.
[89] USEPA 2010.

within SRKW critical habitat is expected to be short-term and localized, and therefore insignificant (NMFS 2011).

Potential Effects from the Proposed Net Pen Species Conversion

Conversion of the net pen facilities from Atlantic Salmon to triploid all-female Rainbow Trout/steelhead will not change significant threats to Southern Resident Killer Whales through reduced quantity and quality of prey; persistent pollutants that could cause immune or reproductive system dysfunction; oil spills; and noise disturbance from vessels or any other means. Therefore, the action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Southern Resident Killer Whales.

NORTHERN SEA OTTER

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture

Northern Sea Otter utilize nearshore habitat, commonly away from human activity. They are commonly found in areas of bull kelp or other floating macroalge where they anchor themselves to rest. Northern Sea Otter are marine mammals that primarily prey on marine invertebrate resources but, they will consume any easily available prey. If provided access to concentrated fish in net pens they are likely to include this food resource in their diet. However, marine mammal exclusion nets installed around the outside of net pens effectively exclude otter access to fish inside the net pens. Thus, net pens offer no useful habitat to Sea Otter and do not occupy habitat that Sea Otter require. The soft substrate at depths of 50 feet and more under net pens is not likely to support the prey organisms (sea urchins, sea stars, crabs) that Otters commonly prefer.

Potential Effects from the Proposed Net Pen Species Conversion

The action of converting the net pen facilities from Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing will not change significant threats to Sea Otters within the action area. The action will not increase the risk of oil spills, diseases, parasites, boat strikes, entanglements in fishing nets, marine biotoxins, loss of kelp habitat, reduced genetic diversity or any other potential effect. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Sea Otters.

STELLER SEA LION

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture[90]

Steller Sea Lion occur in Washington but there are no breeding rookeries in the State. They have been observed around floating marine net pens in Rich Passage. Vessels servicing these net pens may cause short-term and localized disturbances, but they are not expected to have any lasting effects. There is adequate space to accommodate sea lion passage around the existing net pen facilities so that any effects on passage are expected to be insignificant. Steller Sea Lion typically feed on fish and large invertebrates such as squid and octopus, so effects to the benthic environment are considered minimal to the Steller Sea Lion prey base. Other than limited and non-lethal predator control permitted by NMFS, the technical memorandums do not state any concerns of adverse effects to marine mammals in Puget Sound in relation to Atlantic Salmon rearing facilities (Nash 2001). Furthermore, the Washington Pollution Control Hearings Board (PCHB) specifically noted in its 1997 ruling that the operation of net pen facilities in Puget Sound does not have a negative impact on marine mammals (Ecology 2007). Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Steller Sea Lion (NMFS 2011).

[90] USEPA 2010.

Potential Effects from the Proposed Net Pen Species Conversion

The action of converting the net pen facilities from Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing will not change significant threats to Steller Sea Lions within the action area. The eastern DPS of the Steller Sea Lion has increased at a rate of 4.76% per year since 1989 (NOAA Fisheries 2019b). No rookeries exist within the action area. The action will not increase the risk of oil spills, disturbance of rookeries, or any other potential effect. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Steller Sea Lions.

BALD EAGLE

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture[91]

The most likely impact to Bald Eagle from marine net pen aquaculture could be increased risk of shoreline oiling and pollution from increased vessel traffic. In general, for bird species, the interaction with fish pen cages and predator exclusion nets is likely to be insignificant, as the nets cover the pens themselves in order to reduce potential bird interactions with the fish in rearing cages. Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Bald Eagle.

Potential Effects from the Proposed Net Pen Species Conversion

The action to convert the net pen facilities from Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing will not change significant threats to Bald Eagles within the action area. The action will not increase shoreline development, clearcutting, chemical pollution, loss of prey, and illegal killing or any other potential effect. The availability of salmonid prey from the net pens is not expected to increase with the proposed action. Therefore, the action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Bald Eagles.

MARBLED MURRELET

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture[92]

Marbled Murrelets could be attracted to forage near salmon net pen complexes if their primary forage fish were attracted to these sites either due to the presence of overhead cover from the net pens, foraging availability from fish feed, or availability of suitable substrate for the forage on the seafloor below the pens. However, Marbled Murrelets are likely to avoid net pen complexes due to the visual presence (disturbance) of the structures, noise and associated human activity. They would be unlikely to be attracted to farmed salmon smolts within the net pens as prey items due to their larger size and weight in relation to their preferred prey items.

There are no known documented instances of Marbled Murrelet entrapment, entanglement, or mortality in nets used in aquaculture sites (Rueggeburg and Booth 1989). The 4-inch square mesh size of marine mammal predator exclusion netting is likely too small to entangle Marbled Murrelets since these birds have a total wing span of 11 to 12.5 inches and would be "flying" underwater with wings somewhat outstretched to catch prey. Since avian exclusion nets are highly visible, stretched taught and flat across net pens, extending only 6 feet above the surface of the water, and surrounded by a visible hand railing, it is extremely unlikely that Marbled Murrelets would collide with these nets, and Marbled Murrelets do not perch on structures like these. The small mesh size of the avian exclusion nets meets the recommendations by Ruggeberg and Booth (1980) that nets have a mesh size no greater than approximately 4 inches (10 cm) after stretching to avoid entangling birds. Unlike the invisible mono-filament gillnets used for commercial fishing that can cause seabird entanglements, salmon net pen

91 RPS ASA 2016.
92 Hamer Environmental 2016.

facilities use a brightly-colored thick polypropylene twine that is both a visual and physical deterrent to flying and diving sea-birds. The net transport process to an upland facility for cleaning and repair after a net pen site is harvested-out avoids any risk of colliding or interacting with diving or foraging Marbled Murrelets.

The overall conclusion of the Marbled Murrelet specialist (Tom Hamer, Hamer Environmental) is that Cooke Aquaculture Atlantic Salmon net pen operations May Affect but are Not Likely to Adversely Affect Marbled Murrelet.

Potential Effects from the Proposed Net Pen Species Conversion

Converting the net pen facilities from Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing is not an action that will change significant threats to Marbled Murrelets within the action area. The action will not increase deforestation, oil spill, entanglement in gill nets or hook-and-line fisheries, or illegal killing or any other potential effect. No aspect of the action is expected to adversely affect the low reproductive rate of Marbled Murrelets. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Marbled Murrelets.

CHINOOK SALMON

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture[93]

Sub-adult salmonid consumption of benthic organisms near net pen facilities is expected to be insignificant since facility siting by WDNR is restricted to deeper waters to limit negative impacts to benthic communities. Since Cooke Aquaculture existing marine net pens with permits to rear Atlantic Salmon are restricted to deeper waters to minimize benthic community impacts, effects on juvenile salmonid nearshore habitat (less than 66 feet MLLW) are also expected to be insignificant. Potential impacts to the migration corridors of listed salmonids is considered to be low since the number and size of net pens in Puget Sound is insignificant in relation to the width and/or water surface area of the channels or bays where they are located, and in relation to the total water surface area of Puget Sound. NOAA technical memorandums do not mention any migration concerns related to the location of existing net pen facilities. For these reasons, Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Chinook Salmon.

NMFS concurs with EPA that the potential impacts of Atlantic Salmon marine net pens on sediment quality in Puget Sound will be insignificant because existing net pens are located mostly in water that is more than 66 feet deep. While nutrient enrichment may occur in beneath the pens, nearshore Chinook Salmon migration, feeding, rearing and predator avoidance will not be affected by sediment quality beneath net pens, because there is no overlap between sediment quality and these PCE (NMFS 2011). NPDES permits issued by Ecology for each of the existing marine net pen operations designate an explicit sediment impact zone (SIZ) 100 feet from the outer edge of the net pen rearing area, and a total organic carbon (TOC) standard used as a screening tool for determining compliance with Washington State Sediment Management Standards (Chapter 173-204 WAC). Organic carbon compounds (from uneaten feed and fish wastes) are the main discharge of nutrients from a salmon net pen operation. If routine sediment monitoring indicates a violation of listed standards, exceedance monitoring and benthic infaunal abundance analysis is required. If these limitations are not met, enhanced monitoring is required and additional mitigation such as a reduction in biomass, fallowing or other measures may be required.

Potential Effects from the Proposed Net Pen Species Conversion

Converting the existing net pen facilities from rearing Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing is not an action that will change significant threats to Chinook Salmon within the

93 USEPA 2010.

action area. The action will not occur in fresh water habitats where Chinook spawn and undergo initial rearing. The proposed action will not alter shoreline marine habitat used by Chinook for early marine rearing. No aspect of the action is expected to adversely affect the reproduction or early rearing of young Chinook prior to their offshore migration. Therefore, the action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Puget Sound Chinook Salmon.

In the event of an inadvertent release of as many as 250,000 Rainbow Trout/steelhead (such as occurred with the Cypress Site 2 collapse in 2017), there is no direct evidence that this would produce an adverse effect on a local Chinook Salmon population. See the response to Escapement Issues and Potential Interactions in Sections A and B of the Additional Information document (SEPA Checklist Attachment B).

- Many, perhaps half of the escaped triploid Rainbow Trout/steelhead would be harvested within the first few days following escapement because they would tend to remain in the vicinity of the rearing pens.
- Many of the triploid Rainbow Trout/steelhead would be trapped in the netting of a collapsed pen or incur physical damage as they egress the pen structure, suffering injuries to the point that their survival time outside of the pens would be reduced.
- Triploid Rainbow Trout/steelhead escaped from marine net pens have essentially no predator experience with the natural prey of Chinook Salmon, placing the triploid Rainbow Trout/steelhead at a competitive disadvantage.
- Steelhead provide no reproductive competition to Chinook since they use different substrate and hydraulic conditions (e.g., smaller gravel, shallower depths, lower water velocities).
- Inadvertent release of 200,000 to 250,000 triploid Rainbow Trout/steelhead would result in far fewer released steelhead than the number annually stocked by State, Federal and Tribal hatcheries (more than 6,000,000 per year) that are available to compete or mate with wild steelhead (due to recapture, poor survival, predation, lack of a homing instinct).
- Inadvertent release of a relatively small number of triploid Rainbow Trout/steelhead (200,000 to 250,000 fish) is unlikely to produce a detectable adverse effect to Washington's Chinook Salmon population in the presence of the intentional (i.e., hatchery) release of approximately 6,000,000 diploid steelhead per year into waters of the State from WDFW, Federal and Tribal hatcheries.
- If a major escapement of triploid Rainbow Trout/steelhead (200,000 to 250,000 fish) were to occur once in 20 years, this would constitute approximately 0.2% of hatchery steelhead released by from State, Tribal and Federal hatcheries during the same time period at the current rate of releases.

CHUM SALMON (Hood Canal Summer-Run)

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture
NMFS concurs with EPA that the potential impacts of Atlantic Salmon marine net pens on Chum Salmon are unlikely to be different from those assessed above for Chinook Salmon. While nutrient enrichment may occur in these areas, nearshore HCSR Chum Salmon migration, feeding, rearing and predator avoidance will not be affected by sediment quality beneath net pens, because there is no overlap between sediment quality and these PCE (NMFS 2011). Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Hood Canal Summer-run Chum Salmon.

Potential Effects from the Proposed Net Pen Species Conversion

Conversion of the existing net pen facilities from Atlantic Salmon rearing to triploid all-female Rainbow Trout/steelhead rearing is not an action that will change significant threats to Hood Canal Chum Salmon within the action area. The action will not increase degradation of spawning habitat, low water flows, or incidental harvest in salmon fisheries or any other potential adverse effect. No aspect of the action is expected to adversely affect the low reproductive rate of Chum Salmon in Hood Canal tributaries. Therefore, the action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Hood Canal Chum Salmon.

COHO SALMON

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture

Since salmonids migrate over long distances, and their most sensitive habitats are the riverine systems in which they spawn, off-shore Atlantic Salmon marine net pen operations likely have little direct impact on their physical habitat. There is unlikely to be an adverse effect due to decrease dissolved oxygen in the vicinity as such conditions would be self-limiting on the concentrated Atlantic Salmon being reared in the net pens. Also, these pens are sited in areas with good circulation, and it is a requirement of the NPDES permit for each marine net pen site that DO monitoring be performed during the critical summer period (August 15 – September 30). Therefore, water quality impacts are unlikely to be an issue. Fish pens could act as an attraction to juvenile salmon during their migration out to sea, but this effect is mitigated through careful feeding practices. Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Coho Salmon.

Potential Effects from the Proposed Net Pen Species Conversion

Converting the existing net pen facilities from rearing of Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing is not an action that will change threats to Coho Salmon within the action area. The action will not occur in fresh water habitats where Coho spawn and undergo initial rearing. The proposed action will not alter shoreline marine habitat used by Coho for early marine rearing. No aspect of the action is expected to adversely affect the reproduction or early rearing of young Coho prior to their offshore migration. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Puget Sound Coho Salmon.

STEELHEAD

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture

The potential effects of Atlantic Salmon floating net pen culture on steelhead are based on features and operations of Cooke Aquaculture existing marine net pens in Puget Sound in which primarily Atlantic Salmon have been raised since the 1990s. The effects to naturally-produced steelhead from Atlantic Salmon floating net pen aquaculture would be the same as for the other species of Pacific salmon.

The risk posed by artificially-produced Atlantic Salmon to naturally-produced steelhead in the Puget Sound steelhead ESU is only through potential competition of accidentally released Atlantic Salmon for prey resources or spawning habitat. However, cultured Atlantic Salmon would only have experienced pellet food and not fish prey, making them poor competitors for naturally-produced steelhead. Any surviving Atlantic Salmon that reach sexual maturity would likely spawn at different times than native steelhead, thereby minimizing or avoiding competition for spawning habitat. Native steelhead populations are currently at a very low level in Puget Sound making it unlikely that Atlantic Salmon would sufficiently reduce available spawning habitat even if there was overlap in their spawning times. Thus, Atlantic Salmon floating net pen culture May Affect but Not Likely Adversely Affect the genetics of naturally-produced steelhead.

Potential Effects from the Proposed Net Pen Species Conversion

Conversion of the existing net pen facilities from rearing of Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing is an action that will not change threats to steelhead within the action area. The action will not occur in fresh water habitats where steelhead spawn, undergo initial rearing and migrate to estuarine waters. The proposed action will not alter shoreline marine habitat used by steelhead for early marine rearing and migration. No aspect of the action is expected to adversely affect the reproduction or early rearing of juvenile steelhead prior to their offshore migration. Therefore, the action to convert from Atlantic Salmon to Rainbow Trout rearing is Not Likely to Adversely Affect Puget Sound Steelhead.

Also see the description of potential effects to Chinook Salmon (above) in the event of inadvertent release of triploid Rainbow Trout/steelhead due to escapement. Potential effects to wild steelhead would be similar, though it is possible that a small number of the overall all-female triploid Rainbow Trout/steelhead population would have reproductive potential. (See the C.4 response in the Additional Information document, SEPA Checklist Attachment B.) Given the reduced population of wild steelhead, it is unlikely that a small number of reproductive Rainbow Trout/steelhead that might successfully escape from a net pen failure would compete with wild steelhead for spawning habitat. However, if any of the escaped Rainbow Trout/steelhead were successful in spawning with wild steelhead, the survival fitness of the offspring would be reduced, as is the case with a much larger number of hatchery diploid steelhead intentionally released by the State each year.

BULL TROUT

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture

Some Bull Trout (amphidromous) produced in Puget Sound streams migrate to estuarine waters for rearing prior to returning to their natal stream to spawn or for additional rearing. Bull Trout occurring in estuarine waters tend to be large juveniles and adults. The decline in Bull Trout populations is primarily due to fresh water habitat degradation and fragmentation, blockage of migratory corridors, poor water quality, past fisheries management practices, impoundments, dams, water diversions, and the introduction of nonnative species. Effects for estuarine habitat are the same as described above for Coho Salmon. Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Bull Trout.

Potential Effects from the Proposed Net Pen Species Conversion

Conversion of the existing net pen facilities from rearing of Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing is an action that will not change threats to Bull Trout and their estuarine habitat within the action area. The action will not occur in fresh water habitats where most Bull Trout spawn, undergo initial rearing and migrate. The proposed action will not alter shoreline marine habitat used by Bull Trout for early marine rearing and migration of the amphidromous life form. No aspect of the action is expected to adversely affect the Bull Trout. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Puget Bull Trout or their estuarine habitat.

EULACHON

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture[94]

Eulachon prey (primarily phytoplankton and zooplankton) could be affected by changes in water quality from nutrient enrichment; however, no significant adverse water quality effects are expected from

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[94] RPS ASA 2016.

Atlantic Salmon net pen aquaculture because existing Cooke Aquaculture Puget Sound facilities are located in areas with good circulation, and discharges are regulated by site-specific NPDES permits and monitored for compliance. No significant adverse effect is anticipated on phytoplankton levels in the areas surrounding existing marine net pens. For these reasons, Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Eulachon.

Potential Effects from the Proposed Net Pen Species Conversion

Changing the existing net pen facilities operation of Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing is an action that will not change threats to Eulachon and their access to spawning habitat within the action area. The action will not occur in fresh water habitats where Eulachon spawning occurs. The proposed action will not alter access or degrade spawning habitat. No aspect of the action is expected to adversely affect Eulachon or their habitat. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Eulachon.

Flathead Sole

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture

The existing net pens are located in Puget Sound which is on the southern extreme of the Flathead Sole range. Flathead Sole in the Gulf of Alaska are not overfished and are not subject to overfishing. Flathead sole are quite abundant in Alaska, and populations are well above target levels. On the West Coast, Flathead Sole make up only a small percentage of groundfish harvests. Flathead sole are part of the "other flatfish" complex on the West Coast, and are not subject to overfishing (NOAA 2019c). The normal depth distribution of Flathead Sole is 325 ft and deeper, which is not within the shallow range of depths potentially affected by Cooke Aquaculture existing Atlantic Salmon net pens in Puget Sound. Net pen rearing of Atlantic Salmon is Not Likely to Adversely Affect Flathead sole.

Potential Effects from the Proposed Net Pen Species Conversion

Changing the existing net pen facilities operation of Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing is an action that will not change threats to Flathead Sole or any of their habitat within the action area. The action will occur in estuarine water habitats where Flathead Sole spawning is not likely to occur. The proposed action will not alter access or degrade any aspect of Flathead Sole habitat. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Flathead Sole. Nutrient enrichment and increased benthic invertebrate production in sediments under net pens is likely to include an area that is too small to produce any effect on the Flathead Sole population of the NE Pacific.

Green Sturgeon

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture[95]

The benthic prey of Green sturgeon (shrimp, clams, amphipods, and Dungeness crabs), sand lance and other small fishes could be affected by adverse impacts to water quality and/or sediment quality. No significant adverse water quality effects are expected from Atlantic Salmon net pen aquaculture because discharges are regulated by site-specific NPDES permits and monitored for compliance. No significant adverse sediment impacts are expected from Atlantic Salmon net pen culture because existing Cooke Aquaculture facilities are located in areas with good circulation, and each site is monitored for organic enrichment of the substrate within a Sediment Impact Zone (a 100-ft perimeter around the net pen array) for compliance with site-specific NPDES permits. Potential sediment impacts are further mitigated by the fallow period following each 18-month grow-out period. Green sturgeon critical habitat could be

95 RPS ASA 2016.

impacted if net pen arrays and/or vessel traffic to/from marine net pens were to impede their migration routes. Green sturgeon use multiple water depths in coastal areas. Potential conflict with net pen arrays and/or aquaculture-related vessel traffic is not likely to be significant due to the relatively small footprint of the marine net pen sites, and the tendency of Green Sturgeon to occur in coastal waters more than 330 feet from shore. Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Green Sturgeon.

Potential Effects from the Proposed Net Pen Species Conversion

Changing the existing net pen facilities operation of Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing is an action that will not change threats to Green Sturgeon or access to their spawning habitat. The action will not occur in fresh water habitats where Green Sturgeon spawn. The proposed action will not alter access to or degrade spawning habitat. No aspect of the proposed action is expected to adversely affect Green Sturgeon or their habitat. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Green Sturgeon.

PACIFIC COD

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture[96]

The pelagic prey of Pacific Cod (krill, shrimp and sand lance) could be affected by changes in water quality; however, no significant adverse water quality effects are expected from Atlantic Salmon net pen aquaculture because discharges are regulated by site-specific NPDES permits and monitored for compliance. The demersal prey of Pacific Cod (crabs) could potentially be affected by sediment impacts; however, no significant adverse sediment impacts are expected from Atlantic Salmon net pen culture because existing Cooke Aquaculture facilities are located in areas with good circulation, and each site is monitored for organic enrichment of the substrate within a Sediment Impact Zone (a 100-ft perimeter around the fish pen array) for compliance with site-specific NPDES permits. Pacific Cod primarily occupy deeper waters than the existing Cooke Aquaculture marine net pen sites. Therefore, Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Pacific Cod.

Potential Effects from the Proposed Net Pen Species Conversion

Changing the existing net pen facilities operation of Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing is an action that will not change threats to Pacific Cod or access to their habitat. The action will not occur in deep-water (50 to 300 m) habitats where Pacific Cod spawn. The proposed action will not alter access to or degrade spawning habitat. No aspect of the action is expected to adversely affect Pacific Cod or their habitat. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Pacific Cod.

PACIFIC HAKE

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture[97]

The pelagic prey of Pacific Hake (zooplankton, fish, and squid) could be affected by changes in water quality; however, no significant adverse water quality effects are expected from Atlantic Salmon net pen aquaculture because discharges are regulated by site-specific NPDES permits and monitored for compliance. The demersal prey of Pacific Hake (crustaceans) could potentially be affected by sediment impacts; however, no significant adverse sediment impacts are expected from Atlantic Salmon net pen culture because existing Cooke Aquaculture facilities are located in areas with good circulation, and each site is monitored for organic enrichment of the substrate within a Sediment Impact Zone (a 100-ft

[96] RPS ASA 2016.
[97] RPS ASA 2016.

perimeter around the fish pen array) for compliance with site-specific NPDES permits. Since Pacific hake primarily occupy deeper waters than the floating marine net pens, it is unlikely that Atlantic Salmon net pen culture has a significant impact to this species. Therefore, Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Pacific Hake.

Potential Effects from the Proposed Net Pen Species Conversion

Changing the existing net pen facilities operation of Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing is an action that will not change threats to Pacific Hake. The action will occur in open water of Puget Sound that includes a very small portion of the lightly populated habitat occupied by Pacific Hake. The proposed action will not alter access to or degrade spawning habitat. No aspect of the action is expected to adversely affect Pacific Hake or their habitat. The action to convert from Atlantic Salmon to Rainbow Trout rearing is Not Likely to Adversely Affect Pacific Hake.

PACIFIC LAMPREY

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture

Historically, Pacific Lamprey were thought to be distributed wherever salmon and steelhead occurred. However, recent data indicate that the distribution of Pacific Lamprey has been reduced in many river drainages. They no longer exist upstream from many dams and other impassable barriers in west coast streams, including many larger rivers throughout coastal Washington, Oregon, and California. In the marine environment, they have been caught at depths ranging from 300 to 2,600 ft, and as far off the west coast as 62 miles in ocean-haul nets.

Pacific Lamprey face a variety of threats to various life history stages, including barriers to upstream and downstream passage; dewatering and reduced stream flows; poisoning from accidental spills or chemical treatments; poor water quality; dredging activities; stream and floodplain degradation due to channelization, loss of side channel habitat, and scouring; changes in ocean conditions such as prey reduction and an increase in predators; and predation by non-native fish species.

The potential effects of Atlantic Salmon floating net pen culture on Pacific Lamprey are based on features and operations of Cooke Aquaculture existing marine net pens in Puget Sound in which primarily Atlantic Salmon have been raised since the 1980s. These effects have been described in numerous technical studies, some of which are cited in this document. Pacific Lamprey in estuarine and marine waters tend to generally occupy greater depths (>300 ft) then the net pens which are all under 180 feet in maximum depth. The existing rearing of Atlantic Salmon in net pens does not result in any adverse effects to Pacific Lamprey, although escaped Atlantic Salmon may provide a small amount of prey for Pacific Lamprey. Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Pacific Lamprey.

Potential Effects from the Proposed Net Pen Species Conversion

Changing the existing net pen facilities operation of Atlantic Salmon to Rainbow Trout/steelhead rearing is an action that will not change threats to Pacific Lamprey or access to their freshwater spawning habitat. The action will not occur in deep-water (300 to 2,600 ft) marine habitats where Pacific Lamprey reside. The proposed action will not alter their freshwater spawning habitat. No aspect of the action is expected to adversely affect Pacific Lamprey or their habitat. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Pacific Lamprey.

RIVER LAMPREY

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture

In Washington, this species probably historically occurred in most major rivers. The current distribution of River Lamprey in the State includes rivers and streams along the coast from the mouth of the Columbia River to the mouth of the Hoh River (though not elsewhere on the Olympic Peninsula), throughout Puget Sound, and in the Lake Washington basin. Adults feed in nearshore marine and estuarine habitat.

Potential threats to River Lamprey include artificial barriers to migration; poor water quality; harvest; predation by non-native species; stream and floodplain degradation; loss of estuarine habitat; decline in prey, ocean conditions, dredging and dewatering.

The potential effects of Atlantic Salmon floating net pen culture on River Lamprey are based on features and operations of Cooke Aquaculture existing marine net pens in Puget Sound in which primarily Atlantic Salmon have been raised since the 1990s. These effects have been described in numerous technical studies, some of which are cited in this document. River Lamprey tend to remain in fresh water and estuarine habitats where the net pens are not located. The existing rearing of Atlantic Salmon in net pens does not result in any adverse effects to River Lamprey, although escaped Atlantic Salmon could provide a small amount of prey for River Lamprey. Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect River Lamprey.

Potential Effects from the Proposed Net Pen Species Conversion

Changing the existing net pen facilities operation of Atlantic Salmon to Rainbow Trout/steelhead rearing is an action that will not change threats to River Lamprey or access to their freshwater spawning habitat. The action will not occur in deep-water marine habitats where River Lamprey are likely to reside during their marine rearing. The proposed action will not alter their freshwater spawning habitat. No aspect of the action is expected to adversely affect River Lamprey or their habitat. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect River Lamprey.

CANARY ROCKFISH

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture[98]

Canary Rockfish inhabit waters at depths between 160 and 820 feet which is deeper, overall, than the waters under existing Cooke Aquaculture Puget Sound Atlantic Salmon net pens. Due to the deficiency of scientific evidence that existing salmon net pen facilities in Puget Sound harm rockfish species through escape, disease transfer, or other indirect effects; the overall lack of an overlap between existing net pen facilities and primary rockfish habitat; and the small quantity of net pen operations in Puget Sound, EPA has concluded that the existing net pen facilities carry an insignificant risk of negatively affecting rockfish. Therefore, Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Canary Rockfish.

Potential Effects from the Proposed Net Pen Species Conversion

Changing the existing net pen facilities operation of Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing is an action that will not change threats to Canary Rockfish or access to their marine spawning habitat. The action will not occur in deep-water (160 to 820 ft) marine habitats where Canary Rockfish tend to reside. The proposed action will occur in relatively shallow habitat where they are rarely found. Juveniles and sub-adults tend to be more common than adults in shallow water where they are commonly associated with rocky reefs, kelp canopies, and artificial structures. Larval rockfish

98 http://www.nmfs.noaa.gov/pr/speces/fish/canaryrockfish.htm. In USEPA 2010.

feed on diatoms, dinoflagellates, tintinnids, and cladocerans, and juveniles consume copepods and euphausiids that are commonly produced as fouling organisms on the substrate provided by net pens. However, the net pens are a sufficiently small part of the available habitat that they are not likely to produce a detectable positive affect. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Canary Rockfish.

BOCACCIO ROCKFISH

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture[99]

Bocaccio Rockfish are typically found at depths between 160 and 820 feet which is deeper, overall, than the waters under existing Cooke Aquaculture Puget Sound Atlantic Salmon marine net pens. Due to the deficiency of scientific evidence that existing salmon net pen facilities in Puget Sound harm rockfish species through escape, disease transfer, or other indirect effects; the overall lack of an overlap between existing net pen facilities and primary rockfish habitat; and the small quantity of net pen operations in Puget Sound, EPA has concluded that the existing net pen facilities carry an insignificant risk of negatively affecting rockfish. Therefore, Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Bocaccio Rockfish.

Potential Effects from the Proposed Net Pen Species Conversion

Changing the existing net pen facilities operation of Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing is an action that will not change threats to Bocaccio or access to their marine spawning habitat. The action will not occur in deep-water (160 to 820 ft) marine habitats where Bocaccio tend to reside. The proposed action will occur in relatively shallow habitat where they are rarely found. Juveniles and sub-adults tend to be more common than adults in shallow water where they are commonly associated with rocky reefs, kelp canopies, and artificial structures. Larval rockfish feed on diatoms, dinoflagellates, tintinnids, and cladocerans, and juveniles consume copepods and euphausiids that are commonly produced as fouling organisms on the substrate provided by net pens. However, the net pens are a sufficiently small part of the available habitat that they are not likely to produce a detectable positive affect. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Bocaccio.

YELLOWEYE ROCKFISH

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture[100]

Yelloweye Rockfish are typically found at depths between 80 and 1,560 feet, but more commonly at 300 to 590 feet, which is deeper overall than the waters under existing Cooke Aquaculture Puget Sound Atlantic Salmon marine net pens. Due to the deficiency of scientific evidence that existing salmon net pen facilities in Puget Sound harm rockfish species through escape, disease transfer, or other indirect effects; the overall lack of an overlap between existing net pen facilities and primary rockfish habitat; and the small quantity of net pen operations in Puget Sound, EPA has concluded that the existing net pen facilities carry an insignificant risk of negatively affecting rockfish. Therefore, Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Yelloweye Rockfish.

Potential Effects from the Proposed Net Pen Species Conversion

Changing the existing net pen facilities operation of Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing is an action that will not change threats to Yelloweye Rockfish or access to their marine spawning habitat. The action will not occur in deep-water (300 to 590 ft) marine habitats where

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99 http://www.nmfs.noaa.gov/pr/speces/fish/bocaccio.htm. In USEPA 2010.

100 http://www.nmfs.noaa.gov/pr/speces/fish/yelloweyerockfish.htm. In USEPA 2010.

Yelloweye Rockfish are commonly found. The proposed action will occur in relatively shallow habitat where they are rarely found. Juveniles and sub-adults tend to be more common than adults in shallow water where they are commonly associated with rocky reefs, kelp canopies, and artificial structures. Larval rockfish feed on diatoms, dinoflagellates, tintinnids, and cladocerans, and juveniles consume copepods and euphausiids that are commonly produced as fouling organisms on the substrate provided by net pens. However, the net pens are a sufficiently small part of the available habitat that they are not likely to produce a detectable positive affect. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Yelloweye Rockfish.

PINTO ABALONE

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture

There was no historical commercial fishing for Pinto Abalone in Washington State, and the recreational fishery was closed in 1994 due to low abundance. WDFW regularly monitors the abundance of Pinto Abalone at 10 index stations throughout the San Juan Archipelago.

Because Pinto Abalone are highly patchy, cryptic and frequently associate with microhabitats such as rock crevices or patches of coralline algae that may themselves be sparsely distributed, total abundances are not measured. Repeated surveys at a system of index sites are conducted in order to detect temporal trends in abalone abundance. These surveys indicate an 83% decline in abalone abundance between 1992 and 2009 (Rothaus et al. 2008). Densities at all but one site are below or within the minimum range for successful fertilization (NOAA/NMFS 2007).

The existing rearing of Atlantic Salmon in net pens does not result in any adverse effects to Pinto Abalone. The abalone reside in hard substrate habitats with irregular surface that are not suitable for net pens. The algae consumed by Pinto Abalone do not occur in the soft bottom habitats where net pens are sited. Atlantic Salmon culture in floating marine net pens is Not Likely to Adversely Affect Pinto Abalone.

Potential Effects from the Proposed Net Pen Species Conversion

Changing the existing net pen facilities operation of Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing is an action that will not change threats to Pinto Abalone or their habitat. The action will occur in areas of soft bottom habitat where Pinto Abalone are not found. Pinto Abalone are found in nearshore rocky habitats in semi-exposed or exposed coastal regions. They typically occupy the low intertidal zone to a depth of 30 feet (9 m), but may occur as deep as 330 feet (100 m). The existing net pens are not present in or adjacent to habitats of this nature. Therefore, the action to convert from Atlantic Salmon to Rainbow Trout rearing is Not Likely to Adversely Affect Pinto Abalone.

Potential Effects on State-Listed Species and Candidate Species

The effects analysis for existing Atlantic Salmon floating net pen culture on State-listed species and candidate species has been summarized from various sources, for the purpose of updating the *Final Programmatic EIS: Fish Culture in Floating Net Pens* (WDF 1990). All of these paragraphs have been reviewed by Don Weitkamp, Ph.D., Aquatic Biologist and Water Quality Expert, who was one of the original authors of the 1990 Programmatic EIS. Dr. Weitkamp is also the author of all statements of effect (below) for the Cooke Aquaculture proposal to convert the species reared in existing marine net pens from Atlantic Salmon to sterile (triploid) all female Rainbow Trout/steelhead.

PACIFIC HARBOR PORPOISE

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture[101]

Pacific harbor porpoise prey includes schooling fish (e.g., herring, capelin) and invertebrates (e.g., cephalopods). Pelagic prey such as these species could be affected by changes in water quality; however, no significant water quality changes are expected from Atlantic Salmon net pen aquaculture because discharges are regulated by site-specific NPDES permits and monitored for compliance.

The chance of disturbance by vessels traveling to and from net pen sites to transport workers and provide services to the net pens is considered discountable due to the low number of trips (two to four round trips per day). Harbor porpoise echolocation, agile swimming capabilities, and being accustomed to vessel traffic in Puget Sound waters make the risk of vessel strikes unlikely to occur.

The risk of direct interaction with fish pen cages or predator exclusion nets is considered discountable.

The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Pacific Harbor Porpoise.

Potential Effects from the Proposed Net Pen Species Conversion

Changing the existing net pen facilities operation of Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing is an action that will not change threats to Harbor Porpoise access to their marine habitat. Although the action will occur in bays, estuaries and harbors less than 200m (650 ft) deep habitats where Harbor Porpoise are commonly found, the existing physical, water quality, and prey conditions will not change. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Pacific Harbor Porpoise.

GRAY WHALE

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture[102]

Gray whales feed primarily on small crustaceans and other organisms living within soft, shallow water sediment. The depth of predator exclusion nets hanging from the net pens are likely to discourage Gray Whale from feeding under the pens, which occur in a small portion of the available habitat. Potential nutrient enrichment due to fish pen discharges is not expected to alter the benthic community beneath Atlantic Salmon net pens because existing Cooke Aquaculture facilities are located in areas with good circulation, and each site is monitored for organic enrichment of the substrate within a Sediment Impact Zone (a 100-ft perimeter around the net pen array) for compliance with site-specific NPDES permits. Therefore, sediment quality and benthic impacts are unlikely to be an issue. Sediments are also allowed to rest after each grow-out period during the fallow period that follows each 18-month grow-out. Gray Whales are unlikely to interact with the net pen array and can avoid vessels in transit if disturbed. For these reasons, Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Gray Whale.

Potential Effects from the Proposed Net Pen Species Conversion

Changing the existing net pen facilities operation of Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing is an action that will not change threats to Gray Whale access to estuarine habitat in Puget Sound where only small numbers of Gray Whales occur during their migration between Alaska and Mexico. Although the action will occur in near-shore habitats where Gray Whales that enter Puget

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101 RPS ASA 2016.
102 RPS ASA 2016.

Sound occur, the existing physical, water quality, and prey conditions of these habitats will not change due to the proposed action. General nearshore threats to Gray Whale include vessel collisions, entanglement in fishing gear, and disturbance from ecotourism, noise, and whale-watching. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Gray Whale.

PACIFIC HERRING

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture[103]
The pelagic prey of Pacific Herring (zooplankton, fish, and larvae) could be affected by changes in water quality; however, the Cooke Aquaculture floating marine net pens are sited in areas with good circulation, and it is a requirement of the NPDES permit for each site that water quality monitoring be performed. Therefore, water quality impacts are unlikely to be an issue. Since Pacific Herring only temporarily occur as a schooling coastal fish around floating marine net pens, it is unlikely that direct impacts would occur. Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Pacific Herring.

Potential Effects from the Proposed Net Pen Species Conversion
Changing the existing net pen facilities operation of Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing is an action that will not change threats to Pacific Herring or access to their shoreline spawning habitat or juvenile rearing habitat. The action will occur in moderate depth shoreline habitats where Pacific Herring migrate and rear. However, the proposed action will not change conditions affecting Pacific Herring spawning and rearing in Puget Sound. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Pacific Herring.

BLACK ROCKFISH

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture[104]
Rockfish generally use habitat deeper than the areas over which net pens are located. Juvenile rockfish commonly inhabit nearshore areas with structures that offer protection. Young rockfish may include net pens as habitat they use during their transition from shorelines to deep-water habitats. Due to the deficiency of scientific evidence that existing salmon net pen facilities in Puget Sound harm rockfish species through escape, disease transfer, or other indirect effects; the overall lack of an overlap between existing net pen facilities and primary rockfish habitat; and the small quantity of net pen operations in Puget Sound, EPA has concluded that the existing net pen facilities carry an insignificant risk of negatively affecting rockfish. Therefore, Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Black Rockfish.

Potential Effects from the Proposed Net Pen Species Conversion
Changing the existing net pen facilities operation of Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing is an action that will not change threats to Black Rockfish or access to their marine spawning habitat. The action will not occur in deep-water (180 to 1,200 ft) marine habitats where Black Rockfish are commonly found. The proposed action will occur in relatively shallow habitat where they are rarely found. Juveniles and sub-adults tend to be more common than adults in shallow water where they are commonly associated with rocky reefs, kelp canopies, and artificial structures. Larval rockfish feed on diatoms, dinoflagellates, tintinnids, and cladocerans, and juveniles consume copepods and euphausiids that are commonly produced as fouling organisms on the substrate provided by net pens.

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103 RPS ASA 2016.
104 USEPA 2010.

However, the net pens are a sufficiently small part of the available habitat that they are not likely to produce a detectable positive affect. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Black Rockfish.

BROWN ROCKFISH

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture[105]
Rockfish generally use habitat deeper than the areas over which net pens are located. Juvenile rockfish commonly inhabit nearshore areas with structures that offer protection. Young rockfish may include net pens as habitat they use during their transition from shorelines to deep-water habitats. Due to the deficiency of scientific evidence that existing salmon net pen facilities in Puget Sound harm rockfish species through escape, disease transfer, or other indirect effects; the overall lack of an overlap between existing net pen facilities and primary rockfish habitat; and the small quantity of net pen operations in Puget Sound, EPA has concluded that the existing net pen facilities carry an insignificant risk of negatively affecting rockfish. Therefore, Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Brown Rockfish.

Potential Effects from the Proposed Net Pen Species Conversion
Changing the existing net pen facilities operation of Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing is an action that will not change threats to Brown Rockfish or access to their marine spawning habitat. The action will occur in water less than 400 ft deep where Brown Rockfish commonly occur. Juveniles and sub-adults tend to be more common than adults in shallow water where they are commonly associated with rocky reefs, kelp canopies, and artificial structures. Larval rockfish feed on diatoms, dinoflagellates, tintinnids, and cladocerans, and juveniles consume copepods and euphausiids that are commonly produced as fouling organisms on the substrate provided by net pens. However, the net pens are a sufficiently small part of the available habitat that they are not likely to produce a detectable positive affect. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Brown Rockfish.

CHINA ROCKFISH

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture[106]
Rockfish generally use habitat deeper than the areas over which net pens are located. Juvenile rockfish commonly inhabit nearshore areas with structures that offer protection. Young rockfish may include net pens as habitat they use during their transition from shorelines to deep-water habitats. Due to the deficiency of scientific evidence that existing salmon net pen facilities in Puget Sound harm rockfish species through escape, disease transfer, or other indirect effects; the overall lack of an overlap between existing net pen facilities and primary rockfish habitat; and the small quantity of net pen operations in Puget Sound, EPA has concluded that the existing net pen facilities carry an insignificant risk of negatively affecting rockfish. Therefore, Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect China Rockfish.

Potential Effects from the Proposed Net Pen Species Conversion
Changing the existing net pen facilities operation of Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing is an action that will not change threats to China Rockfish or access to their marine spawning habitat. The action will occur in water less than 420 ft deep where China Rockfish commonly occur. Juveniles and sub-adults tend to be more common than adults in shallow water where

[105] USEPA 2010.
[106] USEPA 2010.

they are commonly associated with rocky reefs, kelp canopies, and artificial structures. Larval rockfish feed on diatoms, dinoflagellates, tintinnids, and cladocerans, and juveniles consume copepods and euphausiids that are commonly produced as fouling organisms on the substrate provided by net pens. However, the net pens are a sufficiently small part of the available habitat that they are not likely to produce a detectable positive affect. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect China Rockfish.

COPPER ROCKFISH

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture[107]

Rockfish generally use habitat deeper than the areas over which net pens are located. Juvenile rockfish commonly inhabit nearshore areas with structures that offer protection. Young rockfish may include net pens as habitat they use during their transition from shorelines to deep-water habitats. Due to the deficiency of scientific evidence that existing salmon net pen facilities in Puget Sound harm rockfish species through escape, disease transfer, or other indirect effects; the overall lack of an overlap between existing net pen facilities and primary rockfish habitat; and the small quantity of net pen operations in Puget Sound, EPA has concluded that the existing net pen facilities carry an insignificant risk of negatively affecting rockfish. Therefore, Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Copper Rockfish.

Potential Effects from the Proposed Net Pen Species Conversion

Changing the existing net pen facilities operation of Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing is an action that will not change threats to Copper Rockfish or access to their marine spawning habitat. The action will occur in water less than 600 deep where Copper Rockfish commonly occur. Juveniles and sub-adults tend to be more common than adults in shallow water where they are commonly associated with rocky reefs, kelp canopies, and artificial structures. Larval rockfish feed on diatoms, dinoflagellates, tintinnids, and cladocerans, and juveniles consume copepods and euphausiids that are commonly produced as fouling organisms on the substrate provided by net pens. However, the net pens are a sufficiently small part of the available habitat that they are not likely to produce a detectable positive affect. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Copper Rockfish.

QUILLBACK ROCKFISH

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture[108]

Rockfish generally use habitat deeper than the areas over which net pens are located. Juvenile rockfish commonly inhabit nearshore areas with structures that offer protection. Young rockfish may include net pens as habitat they use during their transition from shorelines to deep-water habitats. Due to the deficiency of scientific evidence that existing salmon net pen facilities in Puget Sound harm rockfish species through escape, disease transfer, or other indirect effects; the overall lack of an overlap between existing net pen facilities and primary rockfish habitat; and the small quantity of net pen operations in Puget Sound, EPA has concluded that the existing net pen facilities carry an insignificant risk of negatively affecting rockfish. Therefore, Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Quillback Rockfish.

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[107] USEPA 2010.

[108] USEPA 2010.

Potential Effects from the Proposed Net Pen Species Conversion

Changing the existing net pen facilities operation of Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing is an action that will not change threats to Quillback Rockfish or access to their marine spawning habitat. The action will occur in water less than 900 ft deep where Quillback Rockfish commonly occur. Juveniles and sub-adults tend to be more common than adults in shallow water where they are commonly associated with rocky reefs, kelp canopies, and artificial structures. Larval rockfish feed on diatoms, dinoflagellates, tintinnids, and cladocerans, and juveniles consume copepods and euphausiids that are commonly produced as fouling organisms on the substrate provided by net pens. However, the net pens are a sufficiently small part of the available habitat that they are not likely to produce a detectable positive affect. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Quillback Rockfish.

REDSTRIPE ROCKFISH

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture[109]

Rockfish generally use habitat deeper than the areas over which net pens are located. Juvenile rockfish commonly inhabit nearshore areas with structures that offer protection. Young rockfish may include net pens as habitat they use during their transition from shorelines to deep-water habitats. Due to the deficiency of scientific evidence that existing salmon net pen facilities in Puget Sound harm rockfish species through escape, disease transfer, or other indirect effects; the overall lack of an overlap between existing net pen facilities and primary rockfish habitat; and the small quantity of net pen operations in Puget Sound, EPA has concluded that the existing net pen facilities carry an insignificant risk of negatively affecting rockfish. Therefore, Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Redstripe Rockfish.

Potential Effects from the Proposed Net Pen Species Conversion

Changing the existing net pen facilities operation of Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing is an action that will not change threats to Redstripe Rockfish or access to their marine spawning habitat. The action will not occur in water 500 to 900 ft deep where Quillback Rockfish commonly occur. Juveniles and sub-adults tend to be more common than adults in shallow water where they are commonly associated with rocky reefs, kelp canopies, and artificial structures. Larval rockfish feed on diatoms, dinoflagellates, tintinnids, and cladocerans, and juveniles consume copepods and euphausiids that are commonly produced as fouling organisms on the substrate provided by net pens. However, the net pens are a sufficiently small part of the available habitat that they are not likely to produce a detectable positive affect. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Redstripe Rockfish.

TIGER ROCKFISH

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture[110]

Rockfish generally use habitat deeper than the areas over which net pens are located. Juvenile rockfish commonly inhabit nearshore areas with structures that offer protection. Young rockfish may include net pens as habitat they use during their transition from shorelines to deep-water habitats. Due to the deficiency of scientific evidence that existing salmon net pen facilities in Puget Sound harm rockfish species through escape, disease transfer, or other indirect effects; the overall lack of an overlap between existing net pen facilities and primary rockfish habitat; and the small quantity of net pen operations in Puget Sound, EPA has concluded that the existing net pen facilities carry an insignificant risk of

[109] USEPA 2010.
[110] USEPA 2010.

negatively affecting rockfish. Therefore, Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Tiger Rockfish.

Potential Effects from the Proposed Net Pen Species Conversion

Habitat preferred by Tiger Rockfish does not occur at Cooke Aquaculture commercial net pen sites in Puget Sound. Further, the WDFW Priority Habitats and Species database mapping for the occurrence of this species excludes the marine waters of Island, King and Pierce Counties (WDFW 2008, updated January 2019).

YELLOWTAIL ROCKFISH

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture[111]

Rockfish generally use habitat deeper than the areas over which net pens are located. Juvenile rockfish commonly inhabit nearshore areas with structures that offer protection. Young rockfish may include net pens as habitat they use during their transition from shorelines to deep-water habitats. Due to the deficiency of scientific evidence that existing salmon net pen facilities in Puget Sound harm rockfish species through escape, disease transfer, or other indirect effects; the overall lack of an overlap between existing net pen facilities and primary rockfish habitat; and the small quantity of net pen operations in Puget Sound, EPA has concluded that the existing net pen facilities carry an insignificant risk of negatively affecting rockfish. Therefore, Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Yellowtail Rockfish.

Potential Effects from the Proposed Net Pen Species Conversion

Changing the existing net pen facilities operation of Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing is an action that will not change threats to Yellowtail Rockfish or access to their marine spawning habitat. The action will occur in relatively shallow water depths from the shoreline to about 1,800 ft deep where Yellowtail Rockfish commonly occur. Juveniles and sub-adults tend to be more common than adults in shallow water where they are commonly associated with rocky reefs, kelp canopies, and artificial structures. Larval rockfish feed on diatoms, dinoflagellates, tintinnids, and cladocerans, and juveniles consume copepods and euphausiids that are commonly produced as fouling organisms on the substrate provided by net pens. However, the net pens are a sufficiently small part of the available habitat that they are not likely to produce a detectable positive affect. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Yellowtail Rockfish.

WALLEYE POLLOCK (South Puget Sound)

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture[112]

The pelagic prey of Walleye Pollock (zooplankton like copepod nauplii, euphausiids, copepods, decapod larvae, and larvaceans) could be affected by changes in water quality within the area of affect around an Atlantic Salmon net pen; however, these pens are sited in areas with good circulation, and it is a requirement of the NPDES permit for each marine net pen site that water quality monitoring be performed. Therefore, water quality impacts are unlikely to be an issue. Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Walleye Pollock.

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111 USEPA 2010.

112 RPS ASA 2016.

Potential Effects from the Proposed Net Pen Species Conversion

Changing the existing net pen facilities operation of Atlantic Salmon to triploid all-female Rainbow Trout/steelhead rearing is an action that will not change threats to Walleye Pollock or access to their marine spawning habitat. The action will occur in relatively shallow water depths above the 330 to 980 ft depth range preferred by most Walleye Pollock. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Walleye Pollock.

OLYMPIA OYSTER

Potential Effects from Atlantic Salmon Floating Net Pen Aquaculture

The potential effects of floating net pen culture on Olympia Oyster are based on features and operations of Cooke Aquaculture existing marine net pens in Puget Sound in which primarily Atlantic Salmon have been raised since the 1990s. These effects have been described in hundreds of technical studies performed by Federal agencies and independent researchers, some of which are cited in this document. The existing marine net pens are not located in shallow nearshore waters where Olympia Oysters once commonly occurred. Atlantic Salmon culture in floating marine net pens May Affect but is Not Likely to Adversely Affect Olympia Oyster.

Potential Effects from the Proposed Net Pen Species Conversion

Other than transitioning to the commercial cultivation of a different species of fish, Cooke Aquaculture is not planning to alter the existing fish pen physical structures, site locations, supporting equipment, or general current practices, methods and cultivation techniques used for growing Atlantic Salmon in net pens. The action to convert from Atlantic Salmon to Rainbow Trout/steelhead rearing is Not Likely to Adversely Affect Olympia Oysters.

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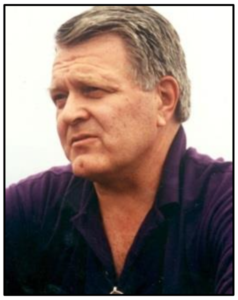
Attachment E

Curriculum Vitae:

Don Weitkamp. Ph.D., and Walton Dickhoff, Ph.D.

DON WEITKAMP, PH.D.



EXPERIENCE: 45+ years

RELEVANT EXPERIENCE:

- Net-Pen & Salmon Hatcheries
- ESA Analysis
- Estuarine Habitat Assessment
- Permitting
- Mitigation
- Habitat Monitoring
- Literature Reviews to Assess Effects

EDUCATION:

Ph.D., Fisheries, 1977
University of Washington

M.S., Invertebrate Pathology, 1971
University of Washington

B.S., Zoology, 1966
Washington State University

SUMMARY OF EXPERTISE

Don is an aquatic biologist and water quality expert who has been working with coastal and inland ports since the 1970s. Don began working with shoreline redevelopment issues and special studies for Puget Sound areas in the 1970s & 80s. His work includes considerable experience with aquaculture projects. He began his scientific carrier working with the shellfish industry issues as a research associate with the Fisheries Research Institute of the University of Washington. His master's thesis was prepared on research dealing with a parasite of cultured mussels and oysters. His initial research on fin fish dealt with dissolved gas supersaturation and its effects on riverine and hatchery fish throughout the Columbia R. basin. This work investigated the prevalence of supersaturation, it's causes and control measures, as well as the biological effects. He designed and directed research sampling in river populations, controlled populations in live cages, and conducted a mobile live cage experiment. He has provided services in program management, permitting, ESA, NEPA/SEPA, water quality, habitat restoration, regulatory compliance, and interagency coordination for ports, irrigation districts, the Corps of Engineers and dam operators.

Don has conducted numerous projects dealing with both hatchery and net pen techniques of aquaculture. His hatchery experience has dealt with the genetics of hatchery populations, the strategic program issues of accelerated rearing, and hatchery effluents. It also includes a 15-year study of fall chinook spawning in the Hanford Reach and its relation to hatchery practices, for an area strongly influenced by dam operation. He has directed studies of migration timing and survival of hatchery population of salmonids in the Columbia River. Other research has included invertebrate organisms that inhabit freshwater and marine environments. Topics of this research include: habitat restoration, contaminated sediments, assessments of invertebrate populations, and evaluation of invertebrate habitats

REPRESENTATIVE PROJECTS

Net-Pen Aquaculture Programmatic EIS: Don oversaw and participated in preparation of the SEPA programmatic EIS for net-pen rearing of salmon in Puget Sound. This project, for the State of Washington, identified alternatives, a vision of how aquaculture could be implemented, and assessed impacts and mitigation potentials for this action.

Harding Creek Aquaculture Project: Don directed a two-year effort to assess the environmental impacts of a proposed hatchery and net pen facility in a remote area of Puget Sound in Washington State. The project had the potential to impact both commercial fisheries and recreational use of the local area. The project conducted water quality monitoring and computational modeling to identify impacts and prepared environmental documentation. Don provided expert testimony at regulatory hearings on the resulted in authorization of the controversial project.

Sea Farms Environmental Assessment: An operational net-pen farm in southern Puget Sound was accused of producing unfavorable environmental impacts, and thus prevented from expansion. Don guided environmental monitoring and computational modeling to assess these impacts. He then provided expert testimony at regulatory hearings to obtain permits necessary for expansion of the net pen operation.

Skagit Net Pen Siting: A private entity and a native American tribe proposed to construct a large net pen operation in northern Puget Sound in an area commonly used for recreational purposes. Don directed an evaluation and computational modeling effort to identify a favorable site within the proposed area. A site was identified that would have no environmental impacts on the benthic habitat.

Net Pen Deposition Modeling: Directed a project to allow siting of a salmon net-pen operation in a bay adjacent to the Strait of Juan de Fuca. Recreational and commercial fishing interests necessitated a thorough examination of potential impacts to sediment and water quality. Computation modeling was used to predict impacts showing that waste accumulation would be limited to a thin layer within 100 m of the pens.

Steelhead Genetics Characterization: Don directed a study to genetically identify the portions of a returning steelhead run that originated from hatchery and wild fish production. Samples were collected from adults trapped at Wells Dam for hatchery production. Electrophoresis examination of these samples identified the portions of these fish produced by the hatchery and by wild fish spawning.

Aquaculture Monitoring Program, Maine: Assigned Principal for development of an environmental monitoring program for the State of Maine related to marine aquaculture. This project included evaluating state and federal water quality regulations, reviewing regulations in other countries, and developing a water quality and sediment monitoring plan for incorporation into state regulations. He also presented the findings and recommendations before a state legislative committee.

Hatchery Production Environmental Assessment: Oversaw a NEPA environmental assessment of a large salmon and steelhead hatchery program for a major Columbia River tributary, the Yakima River. Analysis of multiple proposed sites included potential effects on existing fisheries populations, water quality and quantity, land use and recreation, and wildlife. This hatchery system incorporates adaptive management strategies for program development and is being used as a prototype for the entire Columbia Basin.

Hatchery Effectiveness Survey: Assigned Principal for a comprehensive survey to identify non-published research projects conducted in the last 10 to 15 years on all aspects of salmon, trout, and sturgeon culture. The project developed a computerized database that summarizes this information and makes it readily available.

Cedar River Sockeye Project: Assigned Principal and technical expert for the site selection of a sockeye spawning channel or hatchery on the Cedar River in Washington State. Duties include the identification and evaluation of impacts to natural spawning populations in the river in general and at specific proposed sites and preparation of the fisheries section of the EIS.

Hatchery Accelerated Smoltification: Don directed a three-year study to evaluate the feasibility of accelerating the growth of spring chinook at the Leavenworth Natural Fish Hatchery using warm well water. This project evaluated the potential benefits of establishing an age-0 Chinook program in the Columbia River to increase survival and reduce rearing time.

Juvenile Salmon Studies, Elliott Bay and Duwamish River. Port of Seattle: Don conducted a number of studies in the Duwamish estuary and Elliott Bay to monitor juvenile salmon and resident fish populations. These investigations evaluated the effects of dredging, filling, and other shoreline modifications on juvenile salmon, prey organisms, and fish populations. These studies of the invertebrates and fish involved sampling to establish population densities and habitat types, measuring effects of habitat alterations and enhancement, and determining fish behavior to evaluate the impacts of dredging, filling and pier construction.

Columbia River Channel Deepening ESA Reconsultation: Don prepared the ESA evaluation of the Corps-proposed channel deepening action to address concerns the Federal Services had for the initial consultation evaluation. This evaluation met Services concerns for ESA listed species occurring in the Columbia R. allowing the action to be approved.

Salmon Spawning Assessment Vernita Bar: He helped design and conducted extensive studies of fall Chinook spawning for over 15 years at the largest natural spawning site in the U.S. (Hanford Reach). This FERC license study evaluated all factors potentially affecting spawning success with special emphasis on spawning habitat and flow fluctuations. It included development of an artificial spawning area to mitigate possible impacts due to flow regulation. These efforts resulted in operating criteria for Priest Rapids Dam, during the spawning period, that minimize the upper elevations at which the Chinook spawn, resulting in lower required flows during crucial spring periods.

Pier Removal, WA Dept. Natural Resources: Don led a project to remove three large derelict piers with over 2,500 creosote treated piles. This included coordination with agencies to permit cutting the piles, developing and conducting contaminant investigations and monitoring of water, sediment, and biota for contamination.

PUBLICATIONS

Weitkamp, D.E. 2009. Creosote release analysis ASARCO Docks removal. Report to Washington Department of Natural Resources and U.S. Environmental Protection Agency, Seattle, Washington. 8 p.

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Weitkamp, D.E. 2008. Report on potential effects of Post Falls Hydroelectric Project on fish resources. Report to Avista Corporation, Spokane, Washington. 69 p. + appendix.

Weitkamp, D.E. 2007. Fisheries resources additional analysis and supplemental information on bull trout and bull trout critical habitat in the Coeur d'Alene Lake Basin. Report to U.S. Fish and Wildlife and Avista Corporation, Spokane, Washington. 89 p.

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Sullivan, B.D., and D.E. Weitkamp. 2000. Total dissolved gas monitoring Cabinet Gorge And Noxon Rapids hydroelectric projects 2000. Unpublished report to Avista Corp. Spokane, Washington. 37 p. + Appendices.

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Jones, E.J., D.E. Weitkamp, and A.K. Sparks. 1969. Oyster mortality investigations. Research in Fisheries, University of Washington, Contribution No. 300.

Curriculum Vitae
Walton W. Dickhoff

Education
A.B. Biological Sciences, 1970, University of California, Berkeley.
Ph.D. Physiology, 1976, University of California, Berkeley.

Honors/Awards
U.S. Public Health Service Trainee, 1970-1974.
National Institutes of Health Fellowship, 1976-1977.
Tashiro Fellowship, Kitasato University, Japan, 1987.
Research Faculty Fellowship, University of Washington, College of Ocean and Fishery Sciences, 1987-1988.
Distinguished Research Award, University of Washington, College of Ocean and Fishery Sciences, 1991.
NOAA Distinguished Career Award, 2004
NOAA Fisheries Employee of the Year 2007
NOAA Administrators Award 2011

Employment

2018	Retired
2016-2017	Senior Scientist, National Marine Fisheries Service, Northwest Fisheries Science Center, Seattle
2003-2016	Division Director, Environmental and Fisheries Sciences Division, National Marine Fisheries Service, Northwest Fisheries Science Center, Seattle
1987-2012	Professor (WOT), School of Fisheries, University of Washington.
1986-2003	Physiologist, National Marine Fisheries Service, Northwest Fisheries Research Center, Seattle.
1993-2002	Director, Cooperative Education and Research Program(UW/NMFS)
1988-1989	Assistant Director, Aquaculture Division, School of Fisheries, UW.
1984-1987	Research Associate, School of Fisheries, University of Washington.
1977, '79, '80, '82	Acting Assistant Professor, Dept. Zoology, University Washington.
1975-1984	Research Associate, Dept. Zoology, University of Washington.
1974-1975	Research Assistant, Dept. Physiology/Anatomy, University of California, Berkeley.

Professional Activities
Professional Memberships:
American Association for the Advancement of Science, American Fisheries Society, Society of Integrative and Comparative Biology (formerly American Society of Zoologists; Chair, Division of Comparative Endocrinology 1993-96), Endocrine Society, Society for the Protection of Old Fishes; U.S. Representative on International Federation of Comparative Endocrinological Societies 1996-2001.

Editorships:
Member of Editorial Board of General and Comparative Endocrinology 1978-1991.

Served as Acting Editor-in-Chief of General and Comparative Endocrinology in 1979, 1981, 1983 and 1984.

Editor of General and Comparative Endocrinology 2001-2004

Invited to present papers at the following international meetings:

North Pacific Aquaculture Symposium, Anchorage Alaska, Newport Oregon, August, 1980.

Smoltification Symposium, La Jolla, California, June, 1981.

IX International Symposium on Comparative Endocrinology, Hong Kong, December, 1981.

Symposium on Fish Migration and Reproduction, Tokyo, Japan, December, 1981.

19th Gunma Symposium on Endocrinology, Maebashi, Japan, December, 1981.

U.S.-Japan Cooperative Program, Natural Resources-Aquaculture,Tokyo, Japan, October, 1982.

X International Symposium on Comparative Endocrinology, Copper Mountain, Colorado, June, 1985.

U.S.-Japan Cooperative Program on Natural Resources-AquacultureWoods Hole, Massachusetts, October, 1985.

International Symposium on Common Strategies of Anadromous and Catadromous Fishes, Boston, Massachusetts, March, 1986.

First International Symposium on Fish Endocrinology, Edmonton, Canada, June 12-17, 1988.

Third International Smoltification Workshop, Trondheim, Norway, June 27-July 3, 1988.

U.S.-Japan Cooperative Program on Natural Resources-Aquaculture, Ise City, Japan, October, 1988.

Symposium on Unconventional Vertebrates as Models in Endocrine Research, NIH, Bethesda, MD, December 5–6, 1988.

XI International Symposium on Comparative Endocrinology, Malaga, Spain, May 14–20, 1989.

Applications of Comparative Endocrinology to Aquaculture Symposium, Almuñecar, Spain, May 22-23, 1989.

Symposium on Advances in Fish Pituitary Hormones, Tokyo, Japan, Sept. 21-22, 1992.

Symposium on Applications of Endocrinology to Aquaculture in the Pacific Rim, Bodega Bay, California, September 1994.

Symposium on Biotechnology in Aquacutlure, AAAS Annual Meeting, Baltimore, Maryland, February 1996.

3rd International Symposium on Fish Endocrinology, Hakodate, Japan, June 1996.

Satellite Symposium on fish Migration, Lake Toya, Japan, June 1996.

International Symposium on Insulin Family Peptides, Barcelona, Spain, August 1997.

XII International Symposium on Comparative Endocrinology, Yokohama, Japan, Nov., 1997.

"Molecular Ancestry of Vertebrate Polypeptide Hormones and Neuropeptides", Waseda University, Tokyo, Japan, Nov. 1997.

International Congress on Fish Biology, Vancouver B.C. Canada, July 2002.

Served on the following committees/professional organizations:

Program Officer (elected), Division of Comparative Endocrinology, American Society of Zoologists, 1987- 1989.

Co-organizer, Western Regional Conference on Comparative Endocrinology, Seattle, 1988.

Organizer of XII Annual Smoltification Workshop, Seattle, October, 1990.

Division Chair (elected), Division of Comparative Endocrinology, American Society of Zoologists, 1993-95.

U.S. Representative (elected), International Federation of Comparative Endocrinological Societies, 1996-1999.

Organizer, Fourth International Symposium on Fish Endocrinology, Seattle, WA, July 31-August 3, 2000.

Instructional Activities

Taught undergraduate courses in Dept. Physiology/Anatomy, University of California, Berkeley, 1971 to 1975.

Taught undergraduate and graduate courses in Dept. Zoology, University of Washington, 1977 – 1985, and in School of Fisheries and School of Aquatic and Fishery Sciences, University of Washington 1985 – 2000.

Graduate Advising

Graduate Faculty Member University of Washington and Oregon State University and mentored over 100 graduate students and post-docs.

Publications
Walton W. Dickhoff

1976

1. Matty, A.J., K. Tsuneki, W.W. Dickhoff and A. Gorbman. 1976. Thyroid and gonadal function in hypophysectomized hagfish, *Eptatretus stouti.* Gen. Comp. Endocrinol. 30:500-516.

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2. Dickhoff, W.W. and A. Gorbman. 1977. *In vitro* thyrotropic effect of the pituitary of the Pacific hagfish, *Eptatretus stouti* . Gen. Comp. Endocrinol. 31:75-79.

3. Dickhoff, W.W. 1977. A rapid, high-efficiency bioassay of melanocyte-stimulating hormone. Gen. Comp. Endocrinol. 33:304-306.

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4. Dickhoff, W.W., J.W. Crim and A. Gorbman. 1978. Lack of effect of synthetic thyrotropin releasing hormone on Pacific hagfish (*Eptatretus stouti*) pituitary-thyroid tissue *in vitro* . Gen. Comp. Endocrinol. 35:96-98.

5. Crim, J.W., W.W. Dickhoff and A. Gorbman. 1978. Comparative endocrinology of piscine hypothalamic hypophysiotropic peptides: distribution and activity. Amer. Zool. 18:411-424.

6. Dickhoff, W.W., L.C. Folmar and A. Gorbman. 1978. Changes in plasma thyroxine during smoltification of coho salmon, *Oncorhynchus kisutch* . Gen. Comp. Endocrinol. 36:229-232.

7. Gorbman, A. and W.W. Dickhoff. 1978. Endocrine control of reproduction in hagfish. In: Comparative Endocrinology (P.J. Gaillard and H.H. Boer, eds.) Elsevier/North Holland Biomedical Press, Amsterdam, pp. 49-54.

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