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Guidance for Stressor Identification of Biologically Impaired Aquatic Resources in Washington State

by

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Abstract

This guidance document describes the Stressor Identification method for addressing biological impairment (e.g., a noticeable fish kill or change in invertebrate communities) in waterbodies.

Criteria for biological impairment can be critical tools in detecting degradation of water quality. These criteria have been included in the Washington State Department of Ecology (Ecology) Water Quality Policy (WQP 1-11) since 2006.

Biotic communities may reflect degraded conditions even when common water quality parameters (e.g., fecal coliform, dissolved oxygen, pH) meet standards. This may indicate that other pollutants or activities are degrading water quality or that available water quality data are not strong enough to detect the impairment. Biological impairment can be difficult to address when there is no clear cause of degradation.

This document provides a formal and rigorous process for identifying stressors when a biological impairment has been identified. It provides structure for organizing scientific evidence to support assessment conclusions. The Stressor Identification process involves three basic steps:

- A review of critical information related to the possible causes of degradation.
- Data collection to address information gaps.
- Identification of the cause of impairment based on the strength of evidence collected.

Stressor Identification may be iterative depending on the data available and the complexity of the impairment. There is no regulatory component inherent in this process. However, conclusions drawn from a Stressor Identification assessment can help prioritize the Total Maximum Daily Load or Water Quality Improvement Plan processes.

This guidance document is intended to assist anyone (government or private) involved in managing Washington's aquatic resources.

A case study at the Touchet River in 2006 serves as an example of how Stressor Identification has been carried out in Washington State. Other resources to help guide investigations include web links throughout the document and example data sheets and conceptual models in the appendices.

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Executive Summary

Section 303 of the federal Clean Water Act provides (1) guidance for setting water quality standards and (2) a framework to guide the remediation of waterbodies that do not comply with those guidelines. The Clean Water Act mandates that states and tribes sustain water quality at a level that maintains the integrity of its physical, chemical, and biological resources. The Clean Water Act also directs states to improve water quality at impaired sites.

Traditionally, water quality managers emphasized monitoring for physical and chemical pollutants for which numeric criteria have been adopted. When a water chemistry parameter such as dissolved oxygen, pH, turbidity, or temperature demonstrates impairment, managers take action to remedy the issue through the process of determining a Total Maximum Daily Load (TMDL). A TMDL sets the daily maximum amount of a pollutant that a waterbody can receive from all sources and still meet water quality standards. The goal of the Clean Water Act is not only to address the chemical integrity of the waterbody, but the physical and biological integrity as well. Over the past decade, in recognition of this issue, resource managers have begun to use biological data to make decisions.

Biological criteria can be a critical tool in detecting degradation of water quality. Biotic communities may exhibit the impacts of degradation even when common water quality parameters meet standards. This may indicate that other anthropogenic pollutants or activities are degrading water quality or that available water quality data are not robust enough to detect the impairment.

Organisms may respond to a vast number of complex factors and cumulative impacts for which current water quality standards are inadequate in detecting. For example, there are many chemicals making their way into waterways for which numeric water quality criteria have not been adopted. Likewise, there is no mechanism to measure impairments due to declines in habitat quality, alteration of flow, or changes in sedimentation patterns. Therefore, the United States Environmental Protection Agency (EPA) has provided guidance on the process of identifying stressors responsible for biotic impairment (EPA 2000).

The Stressor Identification (SI) process or Causal Analysis/Diagnosis Decision Information System (CADDIS) leads resource managers through a formal and rigorous process that identifies stressors causing biological impairment in aquatic systems. It provides structure for organizing scientific evidence that supports assessment conclusions (EPA 2000). Stressor Identification also has the benefit of being understandable to a wide variety of stakeholders, allowing them to address and improve water quality issues in their jurisdiction. The ability for people invested in the management and protection of natural resources to participate in identifying causes of impairment is a vital step in improving water quality more quickly.

The SI process involves three basic steps:

- Once a biological impairment has been identified and the case has been defined, a review of critical information should lead to identification of possible candidate causes of degradation.
- All pertinent evidence supporting or refuting each possible cause is collected to determine the most probable causes of impairment.
- Weigh the strength of evidence for each probable cause and identify which candidate is primarily responsible for biotic stress.

This process may be iterative depending on the data available and the complexity of the impairment. It may be necessary to collect more data before managers can make conclusive decisions about the cause(s) of the biological impairment. The final product of an SI assessment is not to develop a TMDL and set load allocations. There is no regulatory component to an SI assessment. Rather, the purpose of the SI assessment is to identify the most likely causes of impairment and provide evidence to support the findings.

Ultimately, the conclusions drawn from the SI process help prioritize the TMDL or Water Quality Improvement Plan process (Figure 1). The accuracy of these conclusions depends on the quality of the data and information used in the SI process. For that reason, the SI process is most effective when it draws on the expertise of professionals in relevant disciplines, such as aquatic ecology, biology, geomorphology, chemistry, toxicology, environmental risk assessment, and statistics. While this analysis can be accomplished with very basic tools, the inclusion of wider ecological expertise increases the number of tools available to investigators, and can result in more precise assessments.

The Washington State Department of Ecology (Ecology) intends this guidance document to assist anyone involved in managing Washington's aquatic resources. This includes tribes, land use planners, industrial and municipal dischargers, reclamation companies, state and local regulators, and volunteer organizations involved in activities that directly or indirectly affect water quality or aquatic habitats.

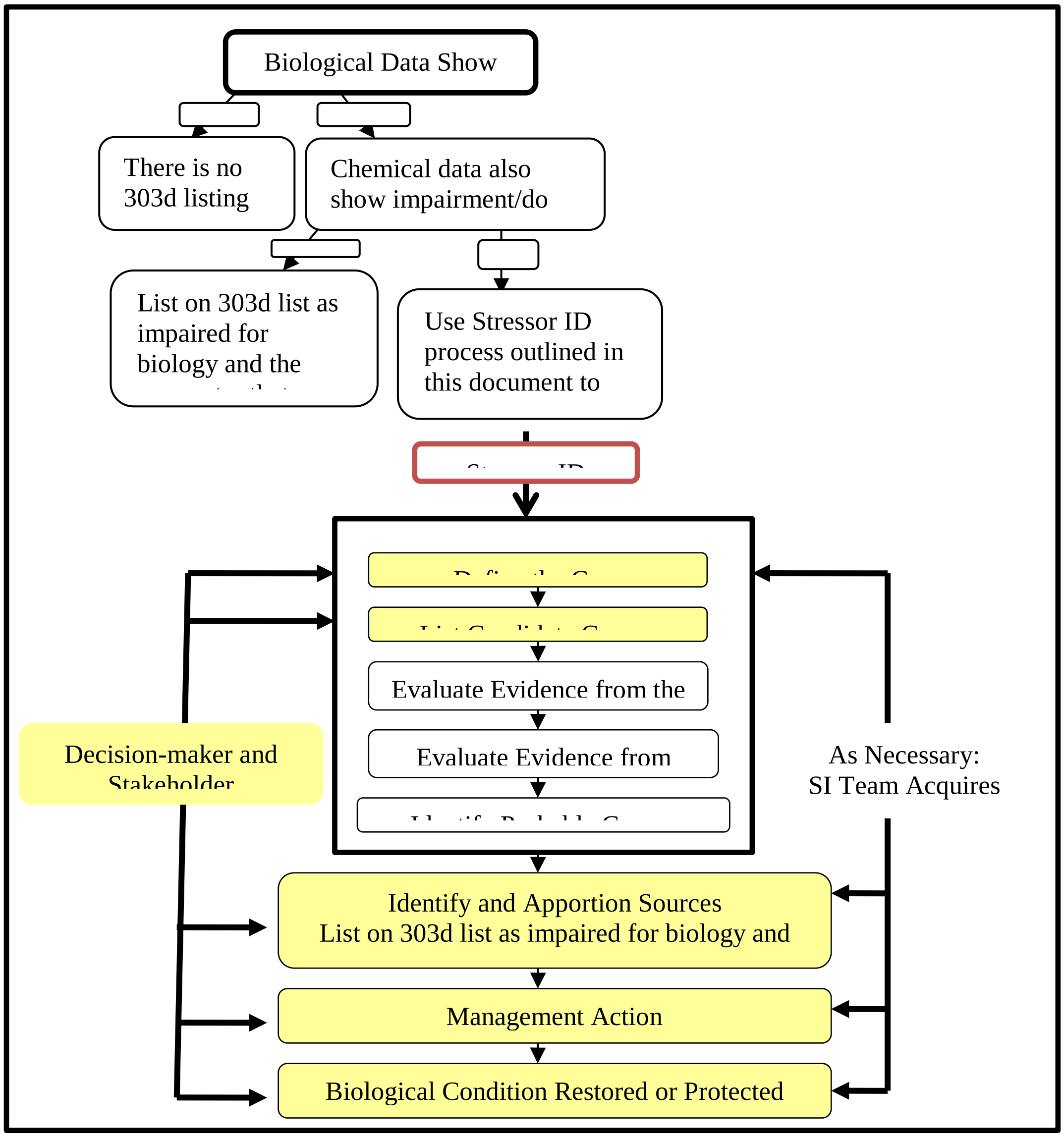


Figure 1. Flow chart depicting how the Stressor Identification process can be used to support regulatory decision making.

The SI process is a useful tool that can help prioritize many water resource management programs (Table 1). In Washington State, the SI process will be informative to many programs in addition to TMDLs and other water cleanup plans. For example, the SI process can be used in a predictive manner to inform the State Environmental Policy Act (SEPA) review process when examining the environmental impacts of a proposal. Stressor Identification can be instrumental for informing Ecology's Watershed Planning Program and the Urban Waters Program to help identify areas of concern and provide evidence of the demands on the water resources in a watershed. The knowledge gained during an SI/Causal Analysis can also help prioritize the disbursement of funds through water quality grant and loan programs. The SI process may also help identify pollutant sources within a watershed and determine what remedial efforts would be the most effective.

Table 1. The role of Stressor Identification (SI) in various water management programs.

Program Type/Name	Purpose	Role of Stressor Identification (SI)
305(b) Characterizing the Quality of the Nation's Waters	Under section 305(b) of the Clean Water Act (CWA), states and tribes are required to assess the general status of their waterbodies and identify, in general terms, known or suspected causes of water quality impairments, including biological impairments.	SI procedures assist states and tribes in accurately identifying the causes of biological impairment. This is a non-regulatory, information reporting effort. A high degree of certainty in identifying the causes of impairment is not always needed for 305(b) reports.
303(d) Listings and TMDLs Identifying Waterbodies and Wetlands that Exceed Water Quality Standards	Under section 303(d) of the CWA, states and tribes are required to prepare and submit to EPA lists of specific waterbodies that currently violate, or have the potential to violate, water quality standards, including designated uses and numeric or narrative criteria such as biocriteria. Wetlands assessment programs are also being developed, and wetlands may be listed on 303(d) lists.	Accurate, reliable stressor identification procedures are necessary for EPA and the states and tribes to accurately identify the cause(s) of water quality standards violations. A high degree of accuracy and reliability in the SI process is necessary, and sources will need to be identified.
State/Local Watershed Management Programs	Managing water resources on a watershed basis involves examining the quality of a waterbody relative to all the stressors within its watershed. Stressors, once identified, are prioritized and controlled through a combination of voluntary and mandatory programs, possibly employing the CWA 402, 319, 404, 401, and other programs.	Stressor Identification procedures will help to identify the different types of stressors within a watershed that may be contributing to biological impairment. A high degree of certainty in identifying the causes of impairment is needed.
319 Nonpoint Source Control Program	The 319 program is a voluntary, advisory program under which the states develop plans for controlling the impacts of nonpoint source runoff using guidance and information about different types of nonpoint source pollution.	Stressor Identification procedures will help to identify the different types of nonpoint sources within a watershed that may be contributing to biological impairment. A high degree of certainty in identifying the causes of impairment is not always needed.
NPDES Permit Program	Under section 402 of the CWA, it is illegal to discharge pollutants to waters of the United States from any "point source" (a discrete conveyance) unless authorized by a National Pollutant Discharge Elimination System (NPDES) permit issued by either the states or EPA. NPDES permits are required whenever a discharge is found to be causing a violation of water quality, including biological impairment.	Accurate SI can be very critical in NPDES permitting cases, both for fairness and success in stressor control. The SI process can help to determine whether the discharge is the cause of biological impairment. This is especially important when site-specific modifications of state standards or national criteria are used. A high degree of accuracy and reliability in the SI process is necessary, and sources will need to be identified. The SI process is not designed to allocate the amount of responsibility for an impact when multiple sources for a stressor are present.
316(b) Cooling Water Intake Program	Under section 316(b) of the CWA, any NPDES permitted discharger that also intakes cooling water must not cause an adverse environmental impact on the waterbody.	To determine whether a cooling water intake structure is causing adverse environmental impacts on the waterbody, the overall health of the waterbody should be known. Where biological impairments are found, SI procedures should be used to identify the different stressors causing the waterbody to be impaired, including the intake structure. A high degree of certainty is needed.

Program Type/Name	Purpose	Role of Stressor Identification (SI)
401 Water Quality Certifications	Under section 401 of the CWA, different types of federal permitting activities (such as wetlands dredge and fill permitting) require a state's certification that there will be no adverse impact on water quality as a result of the activity. This certification process is the 401 Water Quality Certification.	SI procedures will help to identify the different types of stress an activity might place on water quality that can then be addressed through conditions in the 401 Certification.
Wetlands Permitting	Under section 404 of the CWA, the discharge of dredge and fill materials into a wetland is illegal unless authorized by a 404 Permit. The 404 Permit must receive a 401 Water Quality Certification.	SI procedures may help to identify unanticipated stress from a dredge and fill activity on water quality or the biological community after the activity is underway. SI procedures will also help in pre-permitting evaluations of the potential impacts of 404 permitting by assessing different potential stressors on the wetland in advance.
Compliance and Enforcement	Whenever an enforcement action is taken by a regulatory authority, the type of pollution, the source, and other stressors that play a role in causing the violation need to be clearly identified and related to the violating source.	SI procedures must be able to clearly identify the different types of pollution causing the violation with a high degree of confidence. Legal defensibility is required. Identifying the source with a high degree of confidence is also needed, though the current SI process does not provide that guidance.
Risk Assessments	Results of bioassessment studies can be used in watershed ecological risk assessments to predict risk from specific stressors and anticipate the success of management actions.	Accurate SI is an integral part of this process and can help ensure that management actions are properly targeted and efficient in producing the desired results.
Wetlands Assessments	States are beginning to develop wetlands assessment procedures. In the future, wetlands protection is expected to be increasingly incorporated into state water quality standards.	SI procedures, as well as future tools specific to wetland investigations, are very much needed by wetlands managers. The biological assessment methods will allow resource managers to evaluate the condition of wetlands and may provide some indication of the type of stressor damaging a wetland. Once bioassessment methods are completed and incorporated into monitoring programs, wetlands may be listed on 305(b) lists as impaired due to biological impairment. The SI process should help identify stressors causing biological impairment so resource managers can better remedy the problems.
Preservation Programs	The National Estuary Program (NEP) was established in 1987 by amendments to the CWA to identify, restore, and protect nationally significant estuaries of the United States. The program focuses on improving water quality in estuaries, and on maintaining the integrity of the whole system, its chemical, physical, and biological properties as well as its economic, recreational, and aesthetic values.	SI procedures should be useful to the NEP, and other preservation programs, by helping stakeholders identify causes of impairments. This information would feed into the development of a management plan.

Program Type/Name	Purpose	Role of Stressor Identification (SI)
Restoration Programs	The Comprehensive Environmental Response, Compensation, and Liability Act (CERCLA), commonly known as Superfund, was enacted in 1980 (and amended in 1986) for hazardous waste cleanup.	As in enforcement and compliance programs, SI procedures must be able to clearly identify the different types of pollution causing the impairment with a high degree of confidence. Legal defensibility is required. Identifying the source with a high degree of confidence is also needed, though the current SI process does not provide that guidance.
Pollution Control Effectiveness	A key component of any pollution control program or watershed management effort is the ability to ascertain (or predict) the likely effectiveness of pollution control measures or management strategies.	SI procedures will help to identify the different types of pollution a control measure needs to reduce and the different types of stressors a management strategy needs to address.

This document provides an overview of the SI process. Several tools are available in the EPA guidance document on SI at www.epa.gov/waterscience/biocriteria/stressors/stressorid.pdf.

As an extension of the EPA guidance document, EPA has developed an online application called the Causal Analysis/Diagnosis Decision Information System (CADDIS) (http://cfpub.epa.gov/caddis/) that helps users access, organize, use, and share information to conduct the SI process. In this guidance document the emphasis is on listing the most likely or relevant candidate stressors in Washington State, as well as listing symptoms, sources, and other regionally specific tools to analyze data.

The Stressor Identification (SI) Process

The federal Clean Water Act requires states to identify and remediate impaired bodies of water in order to maintain the chemical, physical, and biological integrity of the resource. This has traditionally been accomplished using water chemistry measurements and physical data to some extent. When a measurement is outside of a healthy range, the waterbody is considered impaired and the stressor is addressed through Total Maximum Daily Load (TMDL) allocations. Rather than determine biological integrity through a surrogate parameter, biological measurements of ecological assessment provide a direct link to resources that the Clean Water Act was designed to protect.

However, while biological data can help determine the condition of the resource, the data do not identify the cause of the impairment, nor do the data illustrate the most effective management plan to remediate the problem. For example, a fish kill or a degraded invertebrate community may indicate the impairment of a waterbody, but there may be no indication of the cause. It is in such cases that the use of the SI process is necessary to develop a water cleanup plan.

While a single individual can complete the SI process alone, it is greatly expedited when conducted by a team. The collective knowledge base is expanded by including those who have expertise in a wide variety of environmental fields, local perspectives, and knowledge of the watershed. The more input received, the better the list of possible causes of impairment. Also, data may need to be collected at different times and locations, and a team of people may be able to accomplish this more quickly.

There is no minimum data set required to conduct a SI. Existing data are often enough to make a determination on the cause of an impairment. However, if the available data set is not sufficient to support a robust determination of causation, it may still be useful to go through the SI process at a screening level. A screening level SI analysis will identify the data that would be the most effective to collect when a full SI analysis can be conducted. Visit the "Fundamentals of Data Analysis" website within CADDIS (http://cfpub.epa.gov/caddis/analytical_tools.cfm?Section=174) to help ensure a sound data set for conducting a full SI analysis. If available data do not meet these requirements, an investigator may opt for a less intense SI process, with the intention of iterative analyses as data become available.

It is important to remove as much bias from the study as possible to accurately assess each site. Therefore, review and question reasons for including or excluding data and look for other relevant data sources throughout the process.

The EPA guidance (2000) and the CADDIS website provide a step-by-step process to determine probable cause once an impairment has been identified. A graphical representation of the SI process (Figure 2) demonstrates the flow of activity and information that ultimately links symptoms of impairment (which prompted the investigation) with a cause.

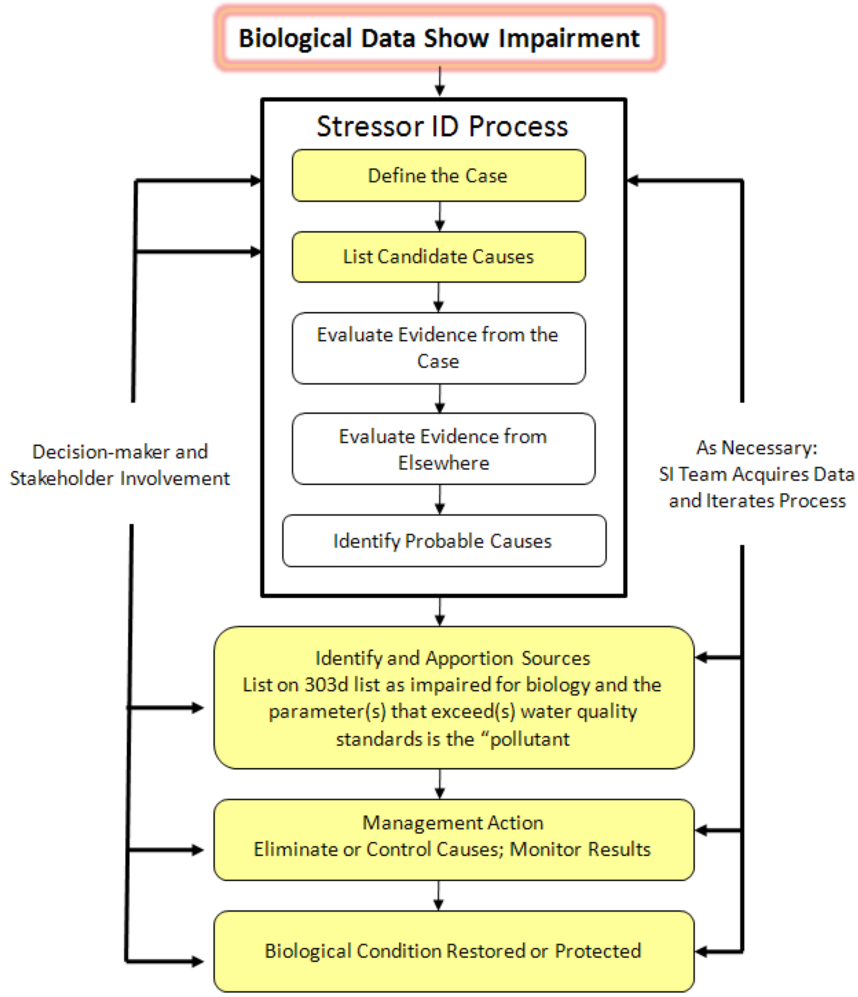


Figure 2. Graphical representation of the Stressor Identification (SI) process (taken from EPA, 2000).

Step 1: Define the Case and Reconnaissance

Verify the Impairment

Stressor Identification is useful when a biological impairment is discovered for which the cause is not evident. Indicators of biological impairment include, but are not limited to:

- A noticeable kill of fish, invertebrates, plants, domestic animals, or wildlife.
- Anomalies in life form such as lesions, parasites, and disease.
- Altered community structure in a biotic community.
- Alteration of extent and processes in an ecosystem.
- Response of indicators that monitor biological or ecological condition of a habitat.

These indicators may or may not coincide with scheduled monitoring or assessment of the waterbody.

Washington State's water quality assessment meets requirements under Sections 303(d) and 305(b) of the federal Clean Water Act. The assessment evaluates waterbody use attainment. Waterbodies are placed in one of five categories for each pollutant or source of degradation for which they have been assessed (Ecology, 2006).

- Category 1 describes a stream that meets the tested criteria for a specific parameter.
- Category 2 describes waters of concern, which may be impaired but not enough information is available to make an impairment determination.
- Category 3 describes a situation when there is little or no data to make a determination.
- Category 4 represents sites that are listed as impaired but do not require a TMDL, because either a TMDL has already been developed, there is another pollution control plan to address the impairment, or it is impaired by a nonpollutant. Examples of nonpollutant impairments are low-flow conditions and the introduction of invasive exotic species.
- Category 5 describes a waterbody that is critically impaired but a cleanup plan has not yet been developed. Streams in this category are listed on the Clean Water Act Section 303(d) list, mandating that action be taken to remedy the problem. 303(d) listings often lead to TMDL development and water cleanup actions.

The Washington State Department of Ecology (Ecology) uses two primary mechanisms of biotic assessment, both based on benthic macroinvertebrate communities. These include

- A multivariate index developed from the Riverine Invertebrate Prediction and Classification System (RIVPACS) and based on the O/E (Observed/Expected) models developed by Hawkins et al. (2000).
- A multi-metric index developed from the Benthic Index of Biotic Integrity (BIBI) (Karr, 1981; Kerans and Karr, 1994).

According to an Ecology policy, the level of confidence afforded by using rigorous assessment tools allows a waterbody segment to be placed in Category 5 when scores from the most recent year of available invertebrate data result in a score representing a degraded condition (Ecology, 2006). Data based on other assessment methods may be used, but these methods require three years of monitoring data to demonstrate consistently degraded results.

The RIVPACS type (O/E) method determines biological health of a site by comparing the observed (O) presence of invertebrate taxa at a study site with the taxa expected (E) to be found at reference sites. This yields an observed versus expected ratio (O/E). These models are based on predictor variables (e.g., elevation, slope, rainfall, latitude, longitude) that are not subject to human influence, describing the variation in the community as a result of natural conditions. Based on Ecology's previous model development work, an O/E score of less than 0.73 is sufficient to demonstrate an impairment for a 303(d) listing in Washington State (Water Quality Program Policy 1-11).

Multi-metric indices (MMIs), such as the BIBI, are another common method used for biotic assessment. The MMI determines the health of a site by scoring metrics representing community diversity, composition, life history components, and the presence of indicator species adapted to tolerate varying degrees of water quality degradation. The MMI is calculated based on models developed for level 3 ecoregions (Omernick, 1995) that were built using reference data. Currently Washington State has developed two region-specific MMI models including one for the Puget Lowlands and one for the Cascades ecoregions (Figure 3; Wiseman, 2003).

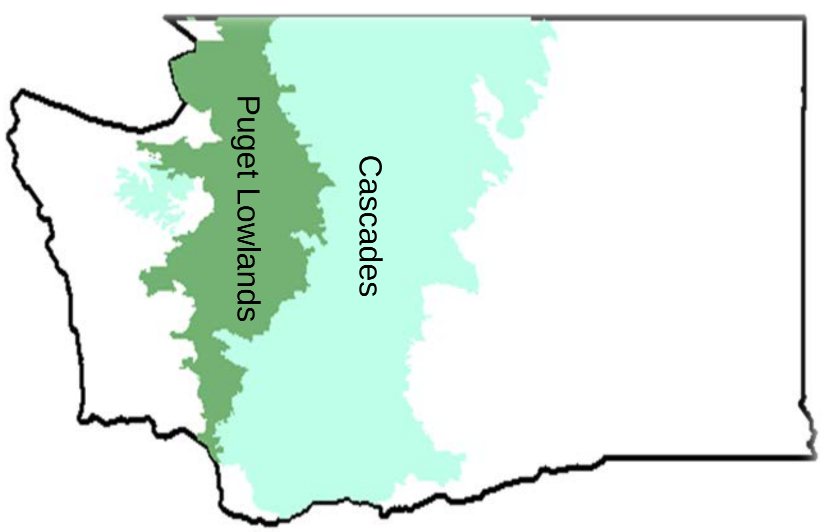


Figure 3. Ecoregions for which Ecology has developed MMI models in Washington State.

Current 303(d) listing criteria in Washington State require that the most recent year of data reflects impairment using the MMI method based on a regionally specific reference model. For the Puget Lowland model, impairment is described by a score of less than 20; for the Cascades model, impairment is described by a score of less than 23 (Wiseman, 2003). Washington State's models will be recalibrated over time to capture the way organisms respond to long-term changes in their environment, including anthropogenic disturbance and climate change. Any updates will be published on the biological monitoring program website: www.ecy.wa.gov/programs/eap/fw_benth/index.htm.

Currently these are the only two calibrated MMI models available for Washington State. While Ecology staff is working to develop models to cover the state, other indices may be used to demonstrate the biological health status of a site. Other index scores from three of the most recent five years of data may be submitted for the water quality assessment to determine impairment of the designated uses.

Natural vs. Anthropogenic Causes

Impaired biological communities that result from natural conditions do occur. For example, a stream reach downstream of a wetland may exhibit low dissolved oxygen levels due to anaerobic soil conditions combined with biological and chemical oxygen demand within the wetland. Likewise, streams fed by groundwater springs may also demonstrate low dissolved oxygen due to a lack of exposure to oxygen as the water filters up through the aquifer. In such cases, there are no anthropogenic sources contributing to the impairment, and no remediation is required (Ecology, 2006). Therefore, after additional monitoring to establish a causal relationship, the investigator can suggest that the waterbody be delisted according to Ecology policy (Ecology, 2006). These waterbodies that do not meet water quality criteria are deemed to be water quality limited based on natural conditions and not by anthropogenic actions.

Define the Impairment

The impairment is generally defined by its nature, magnitude, frequency, and duration. Frequency and duration are often more applicable to water quality impairments. The response of biological communities to stressful events often tends to be longer in duration and to oscillate over time.

The nature of an impairment refers to the parameters that are impaired. One should discuss the designated use classes (www.ecy.wa.gov/programs/wq/swqs/desig_uses.html) of the impaired reach, and the water quality standards used to assess them. This information will help explain how the impaired parameters impact the designated use of the stream.

Magnitude of impairment is measured by the degree to which a water quality standard has been violated and the spatial extent of the waterbody that has been impacted. Describing what biological indicator shows the strongest response, and how it responds, would be important to include to lend inference to candidate stressors. For example, the O/E or MMI scores can be further analyzed to demonstrate which community component is responding to the disturbance in order to help describe the type and degree of the impairment.

Defining impairment of a waterbody is facilitated by mapping and conducting reconnaissance of the affected watershed. Both desktop and field reconnaissance help to develop (1) a list of candidate causes of the event that led to biological impairment, and (2) a map of the impacted watershed, including all significant reaches upstream of the impacted reach.

Defining the Extent of the Impairment: Establishing a Study Area

Washington State currently lists impaired waterbodies by segments or reaches. Currently the water quality assessment stream reaches are based on their location within a section of township and range. Beginning in 2012 these segments will be based on the U.S. Environmental Protection Agency (EPA) National Hydrography Dataset (NHD). The NHD defines a reach as a "significant piece of surface water" (www.epa.gov/owow/monitoring/georef/rf3defined.html) defined by the stretch of river between two confluences or the entry and exit point into and out of a lake on a 1:24,000 scale map. If a sample location is found to be impaired, the entire reach of the stream is listed. The NHD-based segmentation system provides a more hydrologically-based estimation of the extent of the impairment found at a sample site.

Once a waterbody is listed as impaired, it is necessary to determine the true extent of impairment. Biological and water chemistry samples should be taken upstream and downstream of the detection site to (1) determine the location or point of entry of any source of pollution, and (2) establish the attenuation curve as the impact of the stressor decreases downstream. These data may also be reviewed and collected over time. Archival data, when present, can indicate past conditions at a site. Data collected in different seasons or under different weather conditions help to determine the critical period for variable pollutant and impairment conditions. This information provides evidence of co-occurrence and a biological gradient, lending strength to the case later in the process. A worksheet is included in Appendix A as a guide for defining the extent of the impairment.

Defining the Extent of the Impairment: Establishing Reference Sites

Defining the extent of impairment is impossible without defining a reference (or unimpaired) condition. It is best to find unimpaired sites upstream of the impairment. It is generally accepted that "pristine" reference conditions will be rare. The investigator may decide to use a site that is in a "least impacted" state.

It is important to define reference conditions, and use that same definition when choosing all of your reference sites. This provides a frame of reference to determine the extent of the impairment. Also, expectations for what should be achieved from restoration are described based on these reference conditions. At least three reference sites are used for comparison in the study. Whether unimpaired reaches are available, or the use of a least impacted site is unavoidable, describe how the reference condition compares to the impaired site. State the range of conditions of the sites (high quality, reference condition vs. least impaired) as well as the condition of the channel and riparian zone. Considerations when choosing a site are described in Table 2.

Table 2. Considerations when choosing a reference site for study.

Parameter	Consideration
Stream Order	Is the reference site the same order stream as the impaired site?
Valley Type	Is the reference site in the same valley type as the impaired site?
Slope	Is the reference site of a similar slope as the impaired site?
Disturbance	Is the type of disturbance similar in type and amount between the sites?
Elevation	Is the reference site within 1000 feet of the elevation at the impaired site?

Define the Objectives of the Project

Set the expectations for the outcomes, scope, and ramifications of the project by defining the objectives in the context of the management or regulatory purposes, as well as subsequent applications. Stressor investigations can be limited or broad in scope. The general purpose of this analysis is to tease out the leading cause(s) that are responsible for observed effects from among many possibilities. The stated purpose of the project will affect the types of causal candidates considered, the outcome of the project, and data usage later. The objectives also affect the extent and types of data used, the geographic area that is covered, and the time-frame under which this project will be completed.

Tools for Step 1

Listed below are tools to assist in the collection and display of information to determine candidate causes of impairment. These tools will help identify data gaps, which need to be filled to support or refute each cause. These gaps should be explored early in the SI process. The information compiled from this step should point out several candidate causes of impairment for which evidence is gathered in Step 3. Much of the information gathered here is applicable to subsequent remediation programs such as TMDLs and WQIPs.

Reconnaissance

Level One: Desktop

The first practical step to conducting Stressor Identification is to conduct some basic research about the area of interest. Important elements to include might be historical and contemporary aerial photography, topographic maps, pertinent historical data, GIS coverages, land use information, construction permit review, NPDES dischargers, and points of water removal from and returning to the stream (Table 2). Once all the data resources are together, create a map and locate points of interest or any “red flags” that were noticed and warrant verification through a field visit. These “flags” and “points of interest” include any land use anomalies, point source dischargers, or instream alterations that may represent a source of stress to the stream.

For example, there seems to be no riparian buffer along a reach of the Cowlitz River (Figure 4). This particular site appears to be agricultural, suggesting that sediment and agrochemicals, otherwise slowed or prevented from entering the river by riparian vegetation, may reach the river faster and impact the health of the biological community. It may be necessary to visit this site to determine the extent of the riparian buffer that currently exists along the river.

Figure 4. Image of the Cowlitz River generated using EPA's Enviromapper, demonstrating the use of aerial photos to flag areas of potential concern.

The mapping process guides field reconnaissance.

The items in Table 3 can provide helpful information for a desktop reconnaissance, though the investigator should not feel limited to this list. The more information that is collected about a particular case, the stronger the argument is built for or against each candidate cause.

Table 3. Desktop reconnaissance tools for Stressor Identification.

General Information (Ecology Websites)	
Water Quality Program	www.ecy.wa.gov/programs/wq/wqhome.html
Environmental Assessment Program	www.ecy.wa.gov/programs/eap/fwintro.htm
Flow Monitoring Network	https://fortress.wa.gov/ecy/wrx/wrx/flows/regions/state.asp
Biological Information	
Puget Sound Stream Benthos	http://pugetsoundstreambenthos.org//Default.aspx
National Biological Information Structure	www.nbii.gov/portal/server.pt?open=512&objID=236&mode=2&cached=true
Fish Data for the Northwest	www.streamnet.org/
Bonneville Power Admin. Library – Habitat and Fish surveys	www.bpa.gov/corporate/pubs/s_moment.cfm
Washington Dept. of Fish and Wildlife Salmon Scape	http://wdfw.wa.gov/mapping/salmonscape/
Aerial Photography	
Univ. of Washington Map Collection	www.wsulibs.wsu.edu/holland/masc/xmaps.html
Google Maps	http://maps.google.com/
Washington State Department of Transportation	www.wsdot.wa.gov/mapsdata/aerial/
Maps	
Enviromapper for Water	www.epa.gov/waters/enviromapper/
State Soils Map	http://remotesens.css.wsu.edu/washingtonsoil/
Washington State Department of Transportation	www.wsdot.wa.gov/mapsdata/aerial/
Univ. of Washington Map Collection	www.wsulibs.wsu.edu/holland/masc/xmaps.html
National Atlas	http://nationalatlas.gov/natlas/Natlasstart.asp
United States Geological Survey	www.usgs.gov/pubprod/
Geographic InformationSystem Coverages	
Ecology	www.ecy.wa.gov/services/gis/data/data.htm
US Fish and Wildlife Service Data and Systems Service	www.fws.gov/data/
USGS data for Washington State	http://geo-nsdi.er.usgs.gov/catalog/place.php?g=fUS53
GIS Data Depot	http://data.geocomm.com/
Dischargers	
NPDES Dischargers permitted in Washington State	www.ecy.wa.gov/programs/wq/permits/wwdischargepermits.html
Combined Sewer Overflows Metadata and Info	http://aww.ecology/services/gis/gis_meta/facsite/themes.htm
Facilities/Sites that may Impact the Environment	www.ecy.wa.gov/fs/index.html/services/as/iss/fsweb/fshome.html
Archival Water Quality Data (Ecology Websites)	
River and Stream Water Quality Monitoring Program	www.ecy.wa.gov/programs/eap/fw_riv/rv_main.html
Ambient Stream Biological Monitoring Program	www.ecy.wa.gov/programs/eap/fw_benth/ambient.html
Environmental Information Management System	www.ecy.wa.gov/eim/index.htm

Possible Pollutants	
Fertilizer Database	http://apps.ecy.wa.gov/fertilizer/index.html
Water Quality Assessment Tool	http://apps.ecy.wa.gov/wats08/Default.aspx
Miscellaneous Tools	
Western Waters Digital Library Resources	www.westernwater.org/index.php/browse/resources/links/

Interpretation of High Level Indicators

Mathematical models, such as O/E or multi-metric, are composed of many parts. By examining each component, the indicators (O/E or multi-metric indices) used to verify a biological impairment can help the investigator understand the nature of the impairment and indicate possible candidate stressors and exposure pathways affecting biotic integrity. This section will review some of the metrics used in calculating Washington's multi-metric index (MMI) score and describe how they reflect the biological and ecological health of the stream. Some metrics are affected by multiple stressors, and their cumulative effects may not be diagnostic of any one stressor.

Species Richness and Composition Metrics

These metrics demonstrate the diversity of a community, which is directly correlated to the health of the system. A highly diverse community represents long-standing healthy conditions that allow the establishment of a multitude of species with a range of environmental tolerances.

Total Taxa Richness: A species richness metric is common to nearly all MMI indices in the U.S. It is well documented that species richness declines as environmental quality declines (Karr et al., 1986); however, numerous types of environmental degradation influence this index. Therefore, total taxa richness is not diagnostic of any one stressor.

Number of Ephemeroptera, Plecoptera, and Tricoptera (EPT) Taxa: Aquatic invertebrates of these orders are excellent indicators of environmental disturbance. They inhabit a wide variety of habitats including riffles, runs, and pools. A high diversity of these taxa indicate high quality habitat or water quality. The first sign of degradation may be indicated by the absence of stoneflies (Plecoptera) and mayflies (Ephemeroptera) while many caddis fly species (Trichoptera) die out entirely with increasing impairment.

Percent Dominant Taxa: Healthy aquatic systems generally have a diverse community with a relatively low percentage of the community made up of dominant taxa. As disturbance increases, the least tolerant taxa are negatively impacted. The community structure begins to skew towards fewer taxa, which become more abundant because of decreasing competitive inhibition. An increase in dominant taxa indicates disturbance. The particular disturbance may be identified based on the tolerances of the dominant species and stream conditions.

Long-lived Taxa Richness: Long-lived taxa exist where suitable conditions for growth persist over time. A high diversity of long-lived taxa indicates an extensive period of good environmental quality.

Percent Filterers: Filter feeders generally have a mechanism to sieve through the water column to capture particulate matter for food. A high number of filter feeders indicates that there is an abundance of particulate matter suspended in the water column to support such a community. This may be due to a natural seasonal effect occurring when leaf litter falls into the creeks or may reflect higher nutrient conditions. The interpretation for habitat quality will depend on the conditions at the site.

Tolerance Measures

Number of Intolerant Taxa: The presence of a community of intolerant taxa represents good aquatic health; however, a community composed primarily of intolerant taxa may indicate a nutrient poor system. This situation mostly occurs from natural causes (e.g., high numbers of intolerant taxa are often seen in high gradient streams).

Number of Tolerant Taxa: The presence of a high number of tolerant taxa indicates that some form of disturbance, particularly stressors leading to low dissolved oxygen, high turbidity and heavy siltation, or excess nutrients, is impacting the system. Although tolerant organisms are found in both disturbed and natural sites, their abundance increases as conditions deteriorate.

Habitat Measures

Number of Clinger Taxa: Clinger taxa require exposed rocks and plants as substrates on which to attach in fast flowing water. Consequently a lack of clinger taxa indicates that sedimentation may be an issue.

O/E (observed/expected)

A multivariate model such as RIVPACS and other O/E models is based on making predictions about how the community at a study site compares to the community expected given certain site characteristics using the ratio of observed: expected. Therefore if O/E <1, fewer species were encountered than expected, and impairment is suspected. However, when O/E >1, more species were encountered than expected, also indicating impaired conditions, generally stemming from enriched conditions in the stream. Analysis of the actual species composition of a site compared to what was expected, as well as what species were absent or added, may be able to suggest candidate stressors. Conclusions of this comparison would be based on the sensitivity of the species that were either added or taken away from the expected species list.

Sensitivity is measured based on the probability of capturing an invertebrate at any given site based on conditions at that site. This probability is calculated in the taxa tolerance and O/E models. The sensitivity of a species in O/E models is calculated by the following formula:

$$\sum(\text{sites}_{i \ldots n}) \text{ probability of capture for species 1}$$

In other words, the sensitivity of a species is equal to the sum of the capture probability of that species across all sites in the sample. When this sum of probability is greater than 1, it means that their tolerance of parameters found at those sites is high. Those species are called “increasers,” as these species generally replace less tolerant taxa in stressful sites where they are observed when they were not expected. If the sum of probability of capture is less than 1, that species is known as a “decreaser”. If their tolerance for pollution is lower and conditions degrade, decreaser species will disappear from the community where they would otherwise be seen.

Level Two: Field Reconnaissance

While remote sensing, aerial photos, and GIS data layers are increasingly helpful tools, they cannot replace actually visiting a site. Although the investigator may be able to identify points of erosion, discharge, or collection of pollutants from photographs, their current degree of impact and the details necessary to address them cannot be determined without seeing the site. Field reconnaissance allows the investigator to verify and elaborate on information collected from the desktop effort. This section presents additional categories of information necessary (Table 4) to construct a complete sense of the processes that would lead to the conditions in the aquatic habitat under study.

Table 4. Summary checklist of tasks completed in reconnaissance.

Desktop Reconnaissance
Collect high-resolution aerial photography. This should be considered if it is feasible given the scale of the project. Historic aerial photos should be considered for documenting land use history, changes in stream geomorphology, etc.
Obtain and plot applicable GIS layers for the project. Establish watershed sub-catchments within the project area to help identify potential stressor sources and pathways.
Develop a project map using all available GIS layers, aerial photos, and other spatial information. Use this map to plan monitoring activities and field reconnaissance efforts. Existing and proposed monitoring stations should be included on this map.
Field Reconnaissance
Document physical integrity of the stream channel and riparian conditions. See Table 5 for a list of some of the available methodologies. The goal of this assessment should be a general characterization of the stream reaches that will be addressed during the investigation.
Document hydrological features and pathways within the watershed. Collect reconnaissance data at hydrologically significant locations under different flow regimes (snowmelt, baseflow, storm events).
Document impacted areas and potential reference reaches. Within the study area there will likely be areas of varying degrees of habitat degradation. Identify potential stressor sources such as stream impoundments, point source discharges, areas of severe bank erosion, cattle pastures, etc.
Identify areas for future biological, water chemistry, flow, and physical monitoring. These sites should be located in reaches that will provide the evidence needed to evaluate the relationships between potential stressors and biological assemblages.

Document Physical Integrity of Stream Channel and Riparian Conditions

Structural integrity and stability largely determine the quality and type of habitat in a stream. Stream stability is influenced by a limited number of conditions, including flow regime, sediment regime, channel dimensions, and riparian conditions. These conditions can change based on both natural conditions such as stream order, slope, and geology, and anthropogenic disturbances such as land use. During field reconnaissance, it is important to assess the physical integrity of the stream segment and the watershed as a whole. Physical integrity may be naturally unstable. Involving a geomorphologist to help make the determination of natural versus anthropogenic causes for instability may be useful.

Describing channel and riparian conditions may increase understanding of the biological response that brought attention to the physical conditions within and around the stream. Field reconnaissance may identify multiple sources of stress within the system.

Assessing stream channel integrity can be accomplished in a number of ways. During the reconnaissance phase, the investigator should obtain broad descriptions of channel and riparian conditions. This process will ultimately help prioritize reaches that require more detailed assessment later. A list of tools for basic stream assessment is provided in Table 5 to facilitate this process.

Table 5. Tools used by resource managers to assess stream stability and the condition of the riparian corridor.

Tools	Assessment Type	Link
Pfankuch Channel Stability Rating Procedure	Qualitative	www.epa.gov/warsss/pla/pdf/7st9tabV10.pdf
Rapid Geomorphic Assessment		www.maine.gov/dep/blwq/docstream/team/stream_survey_manual/vol_1/appJ-1_scs-instructions.pdf
Rapid Geomorphic Assessment Field Sheets		www.maine.gov/dep/blwq/docstream/team/stream_survey_manual/vol_1/appJ-2_scs-datasheets.pdf
Rapid Geomorphic Assessment Picture Key		www.maine.gov/dep/blwq/docstream/team/stream_survey_manual/vol_1/appJ-3.pdf
NRCS Riparian and Stream Channel Stability Assessment		ftp://ftp-fc.sc.egov.usda.gov/MT/www/technical/environment/envtechnoteMT2.pdf
NRCS Stream Visual Assessment		www.nrcs.usda.gov/technical/ECS/aquatic/svapfnl.pdf
Rosgen/EPA WARSSS	Quantitative	www.epa.gov/warsss/

NRCS - Natural Resources Conservation Service
WARSSS – Watershed Assessment of River Stability and Sediment Supply.

In the interest of saving time and money, a watershed-wide assessment, even on a basic scale, may not be practical. For this reason, the focus is to establish strategic assessment sites to capture the most information about the watershed for the least amount of effort. For example, there are sites where previous data about the aquatic environment have been collected, extending the amount of data for the sites over time.

Take site photographs and conduct stream channel assessments in the following types of locations:

- The site where the impairment was first identified.
- A suite of at least three reference sites that represent a stream of comparable size and hydrogeomorphic setting (Rosgen, 1996) within the same ecoregion.
- Sites corresponding with changes in geology within the drainage.
- Change in land use.
- Upstream and downstream of tributaries.
- Upstream and downstream of impoundments.
- Upstream and downstream of incisions.

Document Hydrologic Features and Pathways

It is possible that the source of impairment is not in the stream under study, but occurs on one of the tributary water sources, including creeks, wetlands, lakes, road-side or agricultural ditches, or groundwater springs. Photographs and basic water chemistry measurements from just upstream of the confluence of each type of inflow should be made, including dissolved oxygen, pH, temperature, and conductivity. It may also be necessary to measure a representative sample of each of the water sources that contribute to the stream under study and to evaluate their impacts. If possible, make these measurements from a range of hydrological conditions such as just after or during a rain event, during the spring snowmelt, and during baseflow (low-flow) periods in late summer.

Level Three: Data Gaps Analysis

The purpose of data gaps analysis is to determine if the investigator has have collected adequate quality data for the physical, chemical, and biological attributes of the stream to address the possible stressors in the system. At this point the investigator should have a good sense of the setting in which the impairment takes place. This includes an understanding of the nature, magnitude, and extent of the impairment. It is important not to make any foregone conclusions about the cause of a stressor. A study can introduce bias toward only one stressor in the watershed, when there may be multiple factors operating on the biological community. Try to capture these factors (Table 6) by continuously reviewing and questioning motives for including or not including data, and always look for other possible data sources relevant to the study.

Table 6. Summary of data gaps analysis.

Hydrology
Document hydrological pathways and processes for the impaired reach and watershed, including historic/current flow regime (intermittent/perennial; flashy hydrology vs. watershed storage).
Document the water sources and pathways that drive the local hydrology (i.e. groundwater, rainfall, and snowmelt). • Approximate bankfull discharge and recurrence interval. • Alterations to the natural hydrology of the watershed from land uses or climatic events.
Plan flow monitoring approach for the project. Consider: • Continuous stream gauging station and development of rating curves. • Obtaining and analyzing historic flow records.
Water Chemistry
Organize and evaluate existing water chemistry data, including grab samples, continuous monitoring data, watershed reconnaissance data.
A complete suite of water chemistry measurements should include: • Dissolved Oxygen • Temperature • Conductivity • pH • Alkalinity • Turbidity • Ammonia • Total Phosphates • Chloride • Chlorophyll a
Decide on the objectives for additional water chemistry monitoring. • Targeted design – Sites located strategically around sources, and impaired and reference sites. • Key parameters – Other possible causal parameters (e.g., organic chemicals, metals). • Timing (e.g., baseflow, rain event). • Frequency.
Establish locations and protocol for continuous monitoring of temperature, conductivity, dissolved oxygen.
Consider using geochemistry tracing techniques to better understand hydrological pathways/processes within the watershed and how they react with the underlying bedrock and sediments to impact water chemistry as described in Panno et al., 1994. Hydrochemistry of Mahomet Bedrock Valley Aquifer, East-Central Illinois: Indicators of recharge and ground-water flow.
Physical Habitat
Organize existing habitat data for the impaired reach and reference areas.
Collect quantitative habitat data at all biological monitoring stations.
Collect additional habitat data if necessary.
Fluvial Geomorphology
Determine objectives for geomorphological data collection. • Watershed vs. stream reach scale. • Rapid/qualitative vs. quantitative. • Rosgen/EPA Watershed Assessment of River Stability and Sediment Supply (WARSSS).

Stream and Watershed Connectivity
Plan and implement an assessment strategy for watershed connectivity. • Barriers to fish migration (perched undersized culverts, dams, waterfalls, intermittent streams) • Assess watershed connectivity in the study area • Evaluate biological dataset upstream and downstream of suspected barriers to determine effects
Biology
Organize existing biological data • Adequate spatial and temporal coverage? • Coverage of various habitat types, disturbance gradients, etc.? • Evaluate existing biological data for trends/relationships with the location of candidate causes for impairments • Identify locations for further biological assessment, if necessary. • Probabilistic or targeted monitoring?

The investigator should be aware of any holes in the dataset that require continued investigation. For example, perhaps the dataset is old or is missing essential components. New information needs to be collected to verify and completely define the current conditions of the impairment. The stronger the initial list the dataset is based on, the less data will need to be collected after this step. Below are several major areas to check for gaps when assembling a dataset.

Flow Monitoring

Improving biological conditions is highly dependent on understanding flow regimes. For example, knowledge of the following characteristics will help determine the type and intensity of monitoring that should occur in a stream:

- The permanence of a stream (intermittent, ephemeral, perennial flow).
- Whether the hydrology is driven by snowmelt, rainfall, or groundwater.
- Whether the stream tends to be flashy or consistent.
- Timing and duration of peak flows and low flows.

Streamflows can provide information about the seasonality, sensitivity, and bankfull discharge of the stream. Bankfull discharge is the discharge when the channel is full, just before flooding begins. At least one continuous flow gage should be identified in the watershed under study to help describe flow patterns of the stream in question. There are several stream gages established:

- United States Geological Survey (http://waterwatch.usgs.gov/?m=real&r=wa&w=map).
- National Oceanic and Atmospheric Administration (www.weather.gov/ahps/).
- Washington State (https://fortress.wa.gov/ecy/wrx/wrx/flows/regions/state.asp).
- Local governments also support gages in some cases (e.g. King County: http://green.kingcounty.gov/wlr/waterres/hydrology/).

Coverage may not extend to the stream of interest. This would require the installation of a gage at the study site. The frequency of recording depends on the site characteristics. For example, in urban settings with a high percent impervious surface, it is necessary to record flow frequently (every 10 to 15 minutes) to catch the details of the hydrograph during a storm event. However, if gaging a stream section fed by wetlands, the investigator can set the recording frequency to a more extended period, such as once an hour, and still get the full hydrograph.

Water Chemistry

Location, sampling time, and frequency of water chemistry measurements will be dependent on whether the investigator is targeting areas or has a particular stressor (some point source location) that seems a likely candidate. Often, repeated measurements of chemical parameters are needed to differentiate between ambient and incident conditions (those that caused the biological response) in a stream. Table 7 lists chemical parameters that are commonly monitored in SI analyses. This list is not comprehensive. The investigator may identify potentially problematic additional items specific to each case.

Table 7. Commonly monitored water quality parameters.

Parameter	Life Use Designation	Washington Water Quality Criteria	Monitoring Frequency	Location of Station
Dissolved Oxygen (DO)	Char Spawning and Rearing	9.5 mg/L	Diurnal sampling (pre-dawn and late afternoon) during irregular flow periods and critical seasons (late summer) makes a strong dataset. Depth profiling in waterbodies > 4 feet is also advised.	At the site of biological impairment, as well as upstream and downstream of any feature that might lead to fluctuating DO levels.
	Core Summer Salmonid Habitat	9.5 mg/L		
	Salmonid Spawning, Rearing, and Migration	8.0 mg/L		
	Salmonid Rearing and Migration Only	6.5 mg/L		
	Non-anadromous Interior Redband Trout	8.0 mg/L		
	Indigenous Warm Water Species	6.5 mg/L		
Biological Oxygen Demand (BOD)	Freshwater Systems	Not addressed by Ecology's Water Quality Program.	May require diurnal sampling as described above and a depth profile in reservoir settings.	At the site of impairment, in incremental steps away from the site in upstream and downstream directions until you find the range of BOD levels or determine if it is problematic.
Ammonia	Freshwater Systems	Not addressed by Ecology's Water Quality Program.	Routine monitoring. If a stream demonstrates an ammonia problem, it may require temporary continuous monitoring.	At the site of impairment, as well as upstream and downstream in incremental steps to document source region and extent of impact.

Parameter	Life Use Designation	Washington Water Quality Criteria	Monitoring Frequency	Location of Station
Temperature	Char Spawning	9°C (48.2°F)	Routine monitoring. If cool water stream has a temperature problem, it may require temporary continuous monitoring.	At the site of the impairment.
	Char Spawning and Rearing	12°C (53.6°F)		
	Salmon and Trout Spawning	13°C (55.4°F)		
	Core Summer Salmonid Habitat	16°C (60.8°F)		
	Salmonid Spawning, Rearing, and Migration	17.5°C (63.5°F)		
	Salmonid Rearing and Migration Only	17.5°C (63.5°F)		
	Non-anadromous Interior Redband Trout	18°C (64.4°F)		
	Indigenous Warm Water Species	20°C (68°F)		
pH	Char Spawning and Rearing	6.5 to 8.5, including a human-caused variation of less than 0.2 units.	Routine monitoring	At the site of the impairment.
	Core Summer Salmonid Habitat	Same as above.		
	Salmonid Spawning, Rearing, and Migration	6.5 to 8.5, including a human-caused variation of less than 0.5 units.		
	Salmonid Rearing and Migration Only	Same as above.		
	Non-anadromous Interior Redband Trout	Same as above.		
	Indigenous Warm Water Species	Same as above.		
Conductivity	Freshwater Systems	Not addressed by Ecology's Water Quality Program. May be able to address using turbidity if there is a point source.	If chloride is a potential stressor, may require continuous monitoring.	At the site of the impairment.

Parameter	Life Use Designation	Washington Water Quality Criteria	Monitoring Frequency	Location of Station
Phosphates	Freshwater Systems	Not addressed by Ecology’s Water Quality Program. Addressed through pH and DO.	Snow melt/Baseflow/ Storm flow	At the site of the impairment
Nitrogen	Freshwater Systems	Not addressed by Ecology’s Water Quality Program. Addressed through pH and DO.	Snow melt/Baseflow/ Storm flow	At the site of the impairment
Chloride	Freshwater Systems	860.0 mg/L over 1 hour average concentration not to be exceeded >1 time every 3 years 230.0 mg/L over 4-day average not to be exceeded >1 time every 3 years	Snow melt/Baseflow/ Storm flow	At the site of the impairment
TSS	Freshwater Systems	Not addressed by Ecology’s Water Quality Program.	Snow melt/Baseflow/ Storm flow	At the site of the impairment
Turbidity	Char Spawning and Rearing	Turbidity shall not exceed: 5 NTU over background when the background is ≤50 NTU; or a 10 percent increase in turbidity when the background is > 50 NTU.	Snow melt/Baseflow /Storm flow	At the site of the impairment
	Core Summer Salmonid Habitat	Same as Above.		
	Salmonid Spawning, Rearing, and Migration	Same as Above.		
	Salmonid Rearing and Migration Only	Turbidity shall not exceed: 10 NTU over background when the background is ≤50 NTU or a 20 percent increase when the turbidity is > 50 NTU.		
	Non-anadromous Interior Redband Trout	Turbidity shall not exceed: 5 NTU over background when the background is ≤50 NTU or a 10 percent increase in turbidity when the background is > 50 NTU.		

Parameter	Life Use Designation	Washington Water Quality Criteria	Monitoring Frequency	Location of Station
	Indigenous Warm Water Species	Turbidity shall not exceed: 10 NTU over background when the background is ≤50 NTU or a 20 percent increase when the background is > 50 NTU.		
Pesticides	Freshwater Systems	Varies by chemical, not all are addressed. Visit our website.	Daily samples during application season. Check with the county extension agent.	At the site of the impairment and targeted at any suspected source locations.
Metals (Both dissolved and total)	Freshwater Systems	Varies by metal, not all are addressed. Visit our website	Depending on source. May require routine or targeted (after storm event) sampling.	At the site of the impairment and targeted at any suspected source locations.

Physical Habitat Surveys

Instream and riparian environments have a significant influence on the structure and function of the benthic community. As a result, biological assessments should be accompanied by some form of habitat assessment. In the context of Stressor Identification, an understanding of the physical habitat surrounding a stream can inform analysis about possible pathways of transport for some stressors to the study area. There are a number of methods to conduct habitat assessment, ranging from qualitative (less rigorous) to quantitative (more rigorous) assessments (Table 8). Assessments that use quantitative data are recommended to compare the quality of sites and provide a stronger, more defensible argument for a case where habitat plays a role as a source of stress on the biological community.

Table 8. Guidance options for conducting the physical habitat assessment of an impaired stream.

Assessment Method	Reference	Qualitative vs. Quantitative
Rapid Bioassessment	EPA	Largely Qualitative
Status and Trends Monitoring for Watershed Health & Salmon Recovery: Appendices H-S	Washington State Department of Ecology	Largely Quantitative
Qualitative Habitat Evaluation Index	Ohio EPA QHEI Manual Ohio EPA QHEI Field Sheets	Qualitative
Natural Resource Conservation Service (NRCS) Stream Visual Assessment Protocol	Natural Resource Conservation Service	
Stream Walk	EPA Stream Walk Manual EPA Stream Walk Data Sheets	Qualitative and Quantitative

Suspended Sediment and Fluvial Geomorphology

Sediment pollution, suspended or bedded, is one of the leading causes of water quality and biological impairments in the U.S. In Washington State, there are currently no criteria directly addressing suspended or bedded sediment. If the investigator has identified a potential source of sediment to the stream during the desktop reconnaissance (e.g., through land use mapping or reviewing NPDES or construction permits), the investigator may be able to use turbidity criterion to demonstrate that sediments are a cause of impairments (Washington State Water Quality Policy 1-11). Instantaneous turbidity measurements are not very informative. However, if the investigator can document conditions across space and/or time, and in combination with measurements of embeddedness, this information may help support a case for or against sedimentation effects.

Turbidity criteria are related to sediment pollution and suspended sediments. The criterion for turbidity (Policy 1-11, WAC-173-201A-200 (1)(e)) states that for water use and criteria class AA (extraordinary) and A (excellent) waters, turbidity should not exceed 5 NTUs over background turbidity if background is less than 50 NTUs and should not exceed 20% above background turbidity if the background levels are greater than 50 NTUs (Ecology, 2006, Water Quality Policy 1-11, WAC-173-201A 200, WAC-173-201A 070). Turbidity and embeddedness observations should be made at the site of impairment. Many of these measurements are captured using the methods in the physical habitat surveys in the section above.

Stream and Watershed Connectivity

Among the items that may be important to map are connectivity issues. Connectivity is defined as the longitudinal, vertical, lateral, and temporal pathways across which physical, chemical, and biological processes occur within aquatic habitats (Annear, 2004). It is important to maintain these connections for healthy stream and biological communities. Therefore, a survey and documentation of potential barriers to connectivity should be conducted, and the results should be added to the site map. These barriers may be caused by road crossings over poorly designed culverts, diversions, impoundments, and other obstacles to migration of aquatic organisms and hydrologic processes.

Biological Data

Although biological data usually initiate a study, there are many ways to collect supplementary data. Additional sampling may be carried out to provide a status update at the site. There are several reasons that the number of samples at a site may need to be increased. This would allow a solid estimate variability. Larger sample size allows better definition of the extent and magnitude of the problem in a spatial and temporal context. Sampling should occur in multiple sites along the stream, during different seasons if possible. Biological sampling may include multiple life forms, such as sampling benthic macroinvertebrates and periphyton.

If additional sampling is necessary, it should be carried out based on the site selection and protocols provided by the Status and Trends Monitoring protocols for Washington State (www.ecy.wa.gov/programs/eap/stsmf/docs/01SnTWadeableManA-Vv3bhfl.pdf). Biological monitoring stations should be located within the watershed of interest based on well defined objectives. For example, if looking at a more broad scale question such as habitat degradation, a randomized probabilistic sampling design may be best. However, if looking at a particular point source stressor, a targeted study design is most effective.

Meteorological and Climatic Data

Climatological conditions are capable of driving physical and chemical conditions in a stream system. An extensive record of the weather patterns and responding flow patterns for the years during and previous to biological sampling is sometimes helpful to identify weather-related responses. This record also may play a role in a conceptual model pathway.

Summary of Step 1

Table 4 provides a list of items to be addressed and included. If not, document why they were not included in the dataset. At the end of the first step, the following will be accomplished:

- Defined the impairment(s) that are the focus of the investigation.
- Defined the specific biological impairments that triggered the analysis.
- Gathered existing data.
- Identified and addressed data gaps.
- Described the geographic extent and the impact of the impairment.

At this point, move to the next step to identify possible causes of impairment.

Step 2: List Candidate Causes

Make an Initial List

The initial list of candidate causes of biological impairment should be all-inclusive. Even potential causes that seem illogical should be included.

Primarily the list is compiled by brainstorming based on the knowledge of ecological processes and interactions between biological, chemical, and physical factors in the environment. However, the list could potentially get unwieldy. Use the following guidelines to help streamline the list (taken from CADDIS Website http://cfpub.epa.gov/caddis/candidate.cfm).

Do:

- Include all causes (even unlikely ones) suggested by landowners or other stakeholders.
- Include common causes of the observed biological effects.
- Include any possible cause.
- Consider what is unique about the impaired site.
- Consider the natural history of the impaired organisms.
- Exclude anything you can confidently eliminate without quantitative analysis.
- Exclude causes related to natural background conditions.
- Exclude causes that are at an inappropriate scale to the case at hand.

Do not:

- Exclude a stressor based on its concentration in, or absence from, a sample.
- Exclude a stressor for a lack of data.
- Exclude a stressor because there is no apparent source or a link in the causal chain is missing.
- Exclude a stressor because it cannot be managed.

There is a suite of candidate causes (including metals, sediments, nutrients, dissolved oxygen, temperature, ionic strength, flow alteration, and unspecified toxic chemicals) that lead to biological impairment that are commonly referenced across the U.S. The CADDIS website provides an excellent overview of each.

The overview discusses how each cause leads to biological stress, what sources to consider, ways to measure the candidate stressor, evidence to look for if you suspect a particular kind of stress, and the biological effects you might expect to see in response to a particular stressor. The website also discusses other causes to consider, which produce similar evidence and symptoms in the system.

The overview includes literature reviews of the stressor and its effects. Literature reviews also provide useful information such as the dose response relationship for certain biota that should be considered during the "Evaluation of Evidence – Step 3."

It is important to consider possible synergistic effects of multiple stressors. A single element may not have an impact strong enough to cause impairment on its own; however, the interaction of two elements may lead to impaired conditions. For example, low levels of elevated nutrients may not cause conditions that lead to biological impairment alone. However, if riparian vegetation is removed, allowing water temperature to increase and more sunlight to enter the water column, an algae bloom may occur that leads to impairment.

Map the Locations of Presumed Origins of Candidate Stressors

Mapping out the locations of candidate stressors can provide spatial perspective and a visual tool for demonstrating causation or linkage. If the exact locations are not known, consider blocking out an area surrounding the point where the stressor enters or impacts the stream. There may be more than one location for any given stressor, and all locations should be identified.

Construct Conceptual Models

Conceptual models are visual tools that organize information into diagrams. These models should be created for each candidate stressor on the list (Appendix A). The models show the relationship between the potential sources of a stressor, the interactions and pathways they work through, and the particular biological response that describes how the biological community was impaired. The models can also reveal areas that require more data to refute or support a case for a candidate stressor.

EPA is developing interactive conceptual models for their CADDIS website. These models allow the investigator to highlight pathways particular to a candidate cause. The models can guide collection of information to build the case for or against each candidate. The diagrams also demonstrate how a particular stressor would impact a system by suggesting possible pathways. To date, EPA has completed several models, including an interactive model for phosphorus. Soon they will add models for pH, ammonia, pesticides, and physical habitat. These models can be found at http://cfpub.epa.gov/caddis/icm/ICM.htm.

The EPA models are not presented as comprehensive. Users are encouraged to let these models guide them in developing additional pathways through which their candidate stressor may be producing the impairment in their particular case. Each model should be fine-tuned to reflect hypotheses about how and why conditions in the watershed exist.

It is important to include in each model all reasonable pathways that lead to a candidate stressor. Some pathways can be eliminated later, as analysis of the information supports refutation of the candidate stressor as a cause of impairment. It is also helpful to map out the potential point and nonpoint sources of the stressors. For example, point sources such as sewage outfalls and permitted discharges are easy to identify and are often available as GIS coverages. Construction of pathways may be needed to identify less straightforward links such as how smoke stacks affect stream water chemistry by producing substances that can cause acid rain.

Engage Stakeholders

It is important to the success of a Stressor Identification analysis to engage a broad range of stakeholders, including landowners, managers, local citizens, and scientists. Their input will help (1) ensure that the investigation includes as much information as is known about the watershed, and (2) reduce bias by including multiple perspectives. This "outside" input may contribute ideas that investigators had not considered. The maps, models, and data compiled thus far can serve as useful visual aids to explain the current work and the intended outcome of their cooperation.

Organizing and directing such a diverse group can be a difficult task. To lead useful discussions, include questions such as:

- What information is missing?
- Is additional information available that would be helpful to consider in the analysis?
- Are there other potential causes of biological impairment that should be added to the list for analysis?

Any pertinent information gathered from these groups should be included on the candidate stressor list and in the conceptual models. Even if the investigator feels the information is not relevant, it is important to be able to demonstrate to the stakeholders how the evidence does not support the candidate stressor as a cause of the problem.

Finalize the List

Upon consideration of all available evidence and the cataloging of all possible stressors, the investigator may be faced with an extensive list. Some stressors may be eliminated based on evidence gathered that clearly removes them from possibility. However, this should be done with caution to avoid overlooking possible stressors before proper consideration is taken.

The decision to remove a candidate stressor from the list is often based on the history of the waterbody and its watershed, or on the biology and mechanisms identified as a response to a stressor. There are five reasons for defensibly removing candidates from the list:

- Evidence that the candidate cause is absent based on rigorous quantitative field measurements.
- Indisputable evidence that a candidate cause would not occur at a site or that the effect is never caused by the candidate.
- Lack of evidence or observations that are seen in conjunction with a particular cause.
- Insufficient data or lack of confidence in data. This may lead investigators to defer exploration of these causes until data become available.
- Implication with other stressors as a "proximate stressor". Other candidate causes may be found to be proximate stressors, or part of the pathway through which an ultimate stressor must pass to result in biological impairment. In either case, it is strongly recommended that

the reasons for removal from the initial candidate list be documented in case the decision is called into question later.

Multiple Stressors

Lists of candidate causes of biological impairment can grow quickly and become difficult to work through. CADDIS recommends several strategies for combining stressors that are strongly correlated by causal pathways or that may induce an effect through interaction with one another. It is important to avoid broad definitions of candidate causes and to maintain the independent effects of each cause. Clearly defined causes and detailed mechanistic models will help build support, while vague definitions and weakly related aggregations of causes weaken the case. The final list will include stressors identified from the following sources:

- Causes based on knowledge of the scientific/mechanistic processes that could lead to the biological impairment.
- Causes suggested by reconnaissance.
- EPA's common stressors and their indicators.

Summary of Step 2

At the end of this step, the investigator will have created the following:

- A list of candidate causes of biological impairments.
- A map of the possible sources of stressors and their spatial relationship to the site.
- Conceptual models that describe possible pathways leading from pre-existing conditions in the watershed to the proximate and ultimate causes of impairment.

These items will help to focus and direct the collection and analysis of data in the next step.

Step 3: Evaluate the Data

There are two primary objectives to achieve in this step:

- Evaluate the data and evidence to diagnose or refute as many candidate causes of biological impairment as possible.
- For those candidates that are not diagnosed or eliminated from consideration, build a case for each candidate that will be compared in Step 4 to determine likely causes.

In Stressor Identification analysis, investigators evaluate two basic types of data:

- Data collected directly from the case at hand (and allows refutation and diagnosis of causes).
- Data collected from outside the case.

Data from outside the case cannot refute or diagnose but can lend strong support for or against a candidate cause. The strongest support for or against a case comes from evidence that is case specific, such as data that originates from within the watershed for both reference and impaired sites. It is helpful to have both types of data support.

The degree to which each type of evidence supports or weakens a case will be useful in describing the probability of causation. Ultimately, this exercise helps to eliminate possible causes based on credible evidence and to build support for those causes that are not eliminated. Based on the strength of the association between measures of candidate stressors and measures of biological response, a final estimation of the ultimate stressor(s) can be made.

The following tables provide lists of useful lines of evidence directly related to the case (Table 9) and evidence external to the case (Table 10). These evidence lines will be discussed in detail later in this chapter.

Assemble the Data

Case Specific

Below are three questions to answer about the case specific data.

1. Do the candidate cause and biological effect occur in the same location?
2. Does the magnitude of effect increase with the magnitude of exposure?
3. Does a series of events along a causal pathway link the source to the stressor?

Evidence may logically establish that a candidate cause is highly unlikely, or even impossible, as a source of the biological response, thus allowing for elimination of a candidate. The strength of evidence and confidence in the data supporting that evidence may affect whether a candidate cause remains under investigation and needs further data collection.

Table 9. Types of evidence to be gathered directly from the case. (http://cfpub.epa.gov/caddis/step.cfm?step=3&Section=8)

Type of evidence	Description
Spatial and Temporal Co-occurrence	The biological effect is observed where and when the candidate cause is observed and is not observed in the absence of the agent.
Evidence of Exposure to a Biological Mechanism	Measurements of the biota show that relevant exposure has occurred or that other biological processes linking the candidate cause with the effect have occurred.
Causal Pathway	Precursors of a candidate cause (components of the causal pathway) provide supplementary or surrogate evidence that the biological effect and candidate cause are likely to have co-occurred.
Stressor-Response Relationship from the Field	The intensity or frequency of biological effects at the site increases with increasing levels of exposure to the candidate cause or decreases with decreasing levels of exposure.
Manipulation of Exposure	Field experiments or management actions that decrease or increase exposure to a candidate cause decrease or increase the biological effect.
Lab Tests of Site Media	Laboratory tests of site media can provide evidence of toxicity, and Toxicity Identification Evaluation (TIE) methods can provide evidence of specific toxic chemicals, chemical classes, or non-chemical agents.
Temporal Sequence	The cause must precede the biological effect.
Verified Prediction	Knowledge of the candidate cause's mode of action permits prediction of unobserved effects that can be subsequently confirmed.
Symptoms	Biological measurements (often at lower levels of biological organization than the effect) can be characteristic of one or a few specific candidate causes. A set of symptoms may be diagnostic of a particular cause if the symptoms are unique to that cause.

External to the Case

Information collected from outside the study site cannot be used to demonstrate direct causation. Rather, it is used to improve the case for the candidate causes that were not refuted after directly related evidence was evaluated. Relevant information from outside data are as follows:

1. Is it plausible that the candidate cause resulted in the observed biological effect given stressor-response relationships described from lab experiments?
2. Is it plausible that the candidate cause resulted in the observed biological effect given stressor-response relationships derived from other field studies?
3. Is the pathway linking the candidate cause to the observed effect mechanistically plausible?
4. Are there other cases where the biological effect in question responded to manipulation of the candidate cause?
5. Is it plausible that the candidate cause resulted in the observed biological effect given stressor-response relationships derived from simulation models?
6. Do analogous stressors cause similar effects?

These external data (Table 10) may include information from other sites within the same region, stressor-response relationships demonstrated in other field or lab studies, and findings of studies of similar situations. Once this information is collected, tie it back to the case at hand. This is done by describing the similarities between the stress caused by the impairment and the results of other lab and field studies. A strong case builds when the logic of the causal pathway in the conceptual model is sound and the relationship is mechanistically plausible. The inclusion of supporting evidence from simulation models also lends strength to the argument for or against causation.

Table 10. Types of evidence collected from outside the study site.

Type of Evidence	Definition
Stressor-Response Relationships from Other Field Studies	The candidate cause in the case is at levels that are associated with similar biological effects in other field studies.
Stressor-Response Relationships from Laboratory Studies	The candidate cause in the case is at levels that are associated with related effects in laboratory studies. These studies may test chemicals, materials, or contaminated media from sites contaminated by the same chemical, mixture, or other agent as the case. If the effects or conditions in the laboratory and field are dissimilar, extrapolation models may improve the correspondence.
Stressor-Response Relationships from Ecological Simulation Models	The candidate cause in the case is at levels that are associated with similar effects in mathematical models that simulate ecological processes.
Manipulation of Exposure at Other Sites	At similarly affected sites, field experiments or management actions that alter exposure to a candidate cause also alter the biological effects.
Analogous Stressors	Evidence that analogous stressors similar to the candidate cause lead to effects comparable to those observed in the case.

Data Organization

It is helpful to organize the data in such a way that the specific measurements of key parameters are clearly linked to each candidate cause. One way to start is to list each candidate cause and then list all the sources available to analyze each cause. Then the data can be organized for each candidate cause in a way that clearly shows comparisons of impacted and reference sites. EPA recommends developing tables (e.g., Table 11) for each candidate cause for this specific purpose. The more types of evidence gathered to support a case, the stronger and more defensible the determination of causation will be.

Table 11. Data analysis table for a hypothetical Stressor Identification (SI) study system running through agricultural fields.

Candidate Cause	Parameter	Concentration or level at reference site	Concentration or level at impaired site	Consistent with pathway?	Strength of support
Habitat Alteration – Lack of large woody debris	Biomass of large woody debris (volume m^3)	25 m^3	20 m^3	Yes	Weak
Excess Nutrients –	ppm nitrates and phosphates	Low	Low	Neither supports nor refutes	No evidence
Excess Sediment – Channel modification	Embeddedness	Low	High	Yes	Strong

Evaluate the Data

Statistical Considerations

Cases should be logical and statistically valid since the decisions based on them are often contentious and under scrutiny. Traditional statistical tests are based on controlled experiments that are designed to meet key assumptions, such as adequate replication, randomized samples, independent data points, and normal variation in the data. Field data collected for SI rarely meet these assumptions. There are many confounding variables in the field (e.g., flow augmentation, stream gradient, weather and climatic differences) and SI is based more on observations than experimental process.

Data analysis for SI does not support traditional hypothesis testing. Instead of testing hypotheses in which a candidate is the likely cause, statistical analyses in the SI process can be helpful in describing data (e.g., mean, variance, range), exploring datasets (describing trends), providing evidence for the best scenario (goodness of fit tests), and modeling exposure-response relationships.

Statistical significance does not equal biological significance. Likewise, a significant correlation does not equal causation. Consider if and how each result fits into a specific causal pathway and let logic dictate interpretation of the results, as opposed to simply accepting that a significant correlation represents a significant result in the ecological system. Statistics used for SI help identify and quantify associations, magnitudes of change, and gradients and patterns in the data. The direction of these associations, changes, and patterns (positive or negative) describe how data relates to the candidate cause. The strength of each piece of evidence builds a more robust case for or against a candidate cause.

To answer questions about the strength of supporting evidence for or against a particular candidate cause's effect, the investigator must analyze and present the data in an organized fashion. Data analysis in SI is primarily done using descriptive statistics, correlation analysis, and quantitative modeling if appropriate. The CADDIS website has conveniently broken down the different applicable methods of data analysis and provided some statistical tools that can be downloaded for this purpose: CADDIS data analysis page.

Ranking Relative Strength of Evidence

To compare different types of evidence, EPA recommends standardizing the scoring of evidence based on the degree to which it supports or weakens a case for a candidate cause. One suggested method for determining relative strength of evidence is to use a system of plusses and minuses (Appendix B). For example, a plus sign demonstrates support for a case, a minus sign demonstrates that the evidence weakens a case, and a zero demonstrates that the evidence has no impact on the biological community. The number of +/- symbols (up to three) indicates the relative strength of a piece of evidence. The highest scores are given to the strongest evidence (evidence collected from the study site), and is based on one or more associations, to link the proximate cause with the effect.

If a type of evidence does not make logical sense, then ranking of "Not Applicable", or "NA." would apply. If the data cannot be analyzed to address a particular type of evidence, rank it "No Evidence", or "NE." If a type of evidence conclusively supports or refutes a candidate cause, it may be marked with a "D" for diagnose or an "R" for refutes.

Types of Evidence

Spatial and Temporal Co-Occurrence

To help prove that a candidate cause actually caused the stressor response, the investigator must demonstrate that the response coincides with when and where the candidate occurs. In addition, the investigator must demonstrate that the response is not seen when or where the candidate is absent. Upstream and downstream investigations from the source should demonstrate this pattern. However investigating a nonpoint source candidate makes locating "upstream" more difficult. This type of evidence is used only for data concerning the proximate stressor and the response.

There are other types of evidence that consider surrogate parameters and measures of indirect steps. For some types of data, the investigator will be able to simply say that a surrogate is either present or absent. However, for stressors that are measured with continuous data, a gradient of exposure and response will need to be established. Co-occurrence can be confounded by time lags between exposure and response, and by episodic exposure. It is, therefore, necessary to (1) determine the likelihood that these factors are involved in the relationship between the candidate cause and the response, and (2) collect samples over time, especially at key periods when exposure is likely.

Evidence of Exposure or Biological Mechanism

Measurements should show that the exposure of the biota to the candidate cause has occurred, or that some mechanism linking the cause to the effect exists. This type of evidence considers steps between the candidate and the effect. While spatial and temporal co-occurrence only establishes that the candidate and the response were seen together, this line of evidence demonstrates that exposure occurred. For example, look for the co-occurrence of characteristic biological symptoms of parasites or toxins, behavioral characteristics (such as fish gulping for air in situations of low dissolved oxygen), and responses of organisms that have different life history strategies.

Time lags and episodic exposure events confound evidence of exposure. The investigator should make the same types of data collection and analysis considerations for co-occurrence in these cases.

Causal Pathway

Causal pathways demonstrate the links between the candidate causes, proximate stressors, and biological responses. Data collected from this line of evidence serve as surrogates by describing the proximate stressor rather than the candidate cause. It is also valuable supporting information when measurements of the stressor are available. There may be many pathways leading to a candidate cause, but only pathways that demonstrate logical links between the source and the cause strengthen the case for a candidate cause. If only a few of the steps are present in a pathway, the investigator may be able to fill in the blanks with further data collection. If this is not within the scope of the study, simply acknowledge that there are other steps missing for which data were unable to be collected, and discuss the implications to the study.

Where there may be some missing steps, data analysis for this line of evidence cannot refute a candidate cause. The possibility of unknown and uninvestigated steps weakens the case.

Stressor-Response Relationships from the Field

Use stressor-response relationships to establish a correlated gradient of exposure and response. The impairment should be greatest where the exposure to the candidate cause is the highest, and where impairment decreases with lower exposure. Likewise, the response should occur within the same period as exposure. The strongest evidence comes from the same time period, showing changes in both stressor and biological response across a set of sites where levels of other candidate causes are constant over time.

Generally, some type of regression or correlation analysis is used to demonstrate the direction of response. Use caution when interpreting results. Keep in mind that these type of analyses are sensitive to sample size, and there is often no replication or randomization involved in these types of field studies. There also may be a danger of confounding candidates. The use of surrogate measurements can amplify this effect, as a single surrogate could be used for multiple stressors. For example, scientists often use conductivity as a surrogate for increased nutrient levels, although conductivity may also be used to describe the underlying lithology of a stream as well. Conductivity may not be a useful measurement without other evidence to support it. This demonstrates the importance of collecting multiple lines of evidence in support of a candidate cause.

Manipulation of Exposure

The ability to manipulate exposure in the field to demonstrate the biological response to the magnitude, duration, and frequency of exposure to a particular stressor could lend important evidence to the case for a candidate cause. Likewise, the lack of the proper response to a stressor because of manipulation will weaken or even refute the case for a candidate cause. Although difficult to come by, this line of evidence helps reduce confounding factors due to manipulation of a single stressor.

Data are analyzed before and after manipulation using a Before/After/Control/Impact (BACI) study design. The analysis may include time series analysis and simple statistical tests (t-tests) if replication and randomization of manipulation are achieved in the study design, though this is a rare case. This type of evidence does not necessarily demonstrate the response to a single manipulated pollutant alone. A biological response may represent a synergistic interaction of factors if multiple pollutants occur at the same site.

Laboratory Tests of Site Media

Material transported from the site can be subjected to lab analysis and controlled experiments. For example, water or sediment samples analyzed for toxins also can be analyzed for the same biological effects as those observed in the field. The strength of this line of evidence lies in the ability to replicate and control the experiment: however, it often requires some extrapolation of biological response between the lab and the field. This line of evidence *cannot be used* to refute a candidate cause due to differences between field and lab conditions, which may confound the effects of toxins in the sample. However, it can lend strong evidence to a water chemistry related cause when toxins in the water are identified and demonstrated to have the same biological effects in the lab as in the field.

Temporal Sequence

In this line of evidence, the cause *must* precede the biological effect. If the effect was seen before a candidate cause occurred, then that candidate is refuted. Generally, this evidence is uncommon, since it relies on data from long-term monitoring to show “before” conditions. This only occurs at a few sites across the state.

Data collected for this line of evidence should be interpreted with caution. Only measurements of the candidate cause should be used to evaluate the temporal sequence. Surrogates, measurements of other steps in the causal pathway that link the stressor to the response, measurements of co-occurrence, and the magnitude of impact do not demonstrate temporal sequence, and are considered in other lines of evidence. Consider that where multiple stressors occur, the stressor that occurs first may mask the impact of subsequent stressors. This may confound the correct identification of a candidate cause if another unknown candidate cause occurred earlier.

Verified Predictions

Knowing how a stressor may affect biological systems allows predictions and measurements of the response in other organisms in the system. This line of evidence highlights the importance of involving a diverse team of investigators to conduct SI analysis. For example, a person with the knowledge of how a toxicant affects biological systems (e.g., digestion, breathing, movement) and a person with the knowledge of the biological systems in different organisms can make educated predictions together. They can combine their knowledge about the response of the community to a given pollutant.

For instance, in one case, cholinesterase-inhibiting pesticides were listed as a candidate cause of biotic stress in an agriculturally influenced waterbody. Investigators predicted that they would not find organisms that had cholinergic systems in a waterbody impacted by these chemicals. This cause was refuted when species with (fish and crawfish) and without (rotifers) systems sensitive to these chemicals were found alive in a waterbody adjacent to an agricultural field (CADDIS Website, 2009).

Predictions may involve data collected to address other lines of evidence, such as temporal sequence, co-occurrence, and causal pathways. Consequently, care should be taken to avoid biasing the analysis by using the same evidence more than once. If a candidate cause truly results from a prediction, and there is no question of the integrity of the data, this line of evidence strengthens the case. Making and validating predictions along several lines of evidence can greatly strengthen the case for a candidate cause.

Symptoms

The presence or absence of characteristic symptoms may be associated with one or only a few specific causes. The larger the number of characteristic symptoms identified, the stronger the case for a candidate cause. The more specifically defined an impairment is, the easier it is to screen for specific symptoms among the affected biota. For example, if endocrine disrupting chemicals are identified as a candidate cause of impairment leading to reduced abundance of fish, necropsies should demonstrate related symptoms such as the presence of ovotestes or vitellogenin in male fish. If these symptoms are not found, but instead the stomachs of many appear blue, the investigator may refute the original candidate and attribute the new symptom as diagnostic of molybdenum toxicity.

Although “symptom” has a medical association, identified symptoms may not be on a medical level. For example, the investigator may find that a particular taxon is affected more than or exclusive of other taxa. Diagnostic symptoms have been well established for many vertebrates, in particular for fish kills. However symptoms are less well defined for invertebrates or for phytoplankton. Nevertheless, this should not limit the inclusion of symptoms that have been defined.

Stressor-Response from Other Field and Laboratory Studies

The purpose of analyzing this type of evidence is to provide documentation that the impaired sites have been exposed to the candidate stressor at quantities, durations, or frequencies sufficient to induce observed biological effects. In this line of evidence, the investigator must demonstrate that the impaired site under study has been impacted by a stressor at or above levels demonstrated to cause similar effects in other field and lab studies. The more studies documented, the stronger the argument for a candidate cause becomes. Studies that are conducted in similar ecosystems and during the same season also strengthen the case. While the most compelling studies are those that document a stressor-response curve, the investigator may also use presence-absence data, which establish the impact of exposure frequency on the stressor-response relationship.

This type of evidence is analyzed using regression analysis to determine the strength of association through the slope of the regression line. The steeper the regression slope, the more highly correlated the stressor and response may be. This type of data should be interpreted with caution, checking that assumptions of randomness and replication are met. Also, co-occurring stressors may require the use of multivariate analysis to tease out the strength of impact of the stressor of interest. When scoring the data, keep in mind that both the direction and magnitude of the relationship describe support or weakness for a candidate.

It is not possible to refute a case using evidence from other studies. This line of evidence must be used with other lines of evidence from the impaired site to confidently describe the strength of evidence for or against a candidate cause.

Stressor-Response Relationships from Ecological Simulation Models

This line of evidence is similar to relationships established in other studies in that the candidate cause must occur at levels identified by mathematical models that lead to the impairment. The development or application of a model often requires specialized knowledge and skills; therefore, this line of evidence may be rarely used. Ecological models are most helpful when modelers are able to manipulate one factor at a time, while holding the other potentially confounding stressors constant. Models alone cannot refute a cause, but they can strengthen the argument when used with other lines of evidence.

Mechanistically Plausible Cause

This line of evidence highlights the difference between a lack of information about a mechanism and evidence that a mechanism simply is not plausible. Evaluation of data from this line of evidence requires a thorough review of whether the proposed causal relationships make mechanistic sense based on the observed impairment, the pathways of exposure, and the known impact of each stressor on biological systems. To conduct this analysis, a mechanism through which the candidate cause could cause the specific biological effects is identified and supported with compelling logic and relevant literature citations. A candidate cause is only refuted if there is no mechanistically plausible cause.

Manipulation at Other Sites

This line of evidence provides a degree of replication to the investigation when conducted under the same manipulative experiments at other sites similar to the site under study. When conducting these types of experiments, the investigator should choose sites that are similarly impacted by the candidate stressor, and include at least some sites in the same ecoregion as the impaired site. The investigator is looking for a correlated biological response to changes in exposure. The investigator can use the same techniques as the “manipulation onsite” line of evidence, including time series analysis and statistical tests, when the number of sites allows adequate replication and randomization. Caution should be used in interpretation of this evidence due to inherent differences between the sites. The use of Before-After-Control-Impact (BACI) (Schmitt and Osenberg, 1996) study design can help to improve these issues. However, differences in recovery rates and treatment effectiveness may not be controllable.

Analogous Stressors

This line of evidence is generally used when investigating organic chemical candidate causes. Justification is necessary to defend the choice of analogous stressors, since the definition of “analogous” is often a matter of professional judgment. With this line of evidence, the aim is to demonstrate that similar responses are seen as a result of the impact of analogous stressors. The example provided by the CADDIS website describes how hormones with similar chemical structures should show a similar response to the stressor.

This kind of evidence is usually less available and often gathered from the literature or from other case studies. If the mechanism through which a particular chemical acts is unknown, there are software packages available to identify analogues that may have known effects.

Multiple Stressors

The investigator may frequently suspect that there are multiple stressors involved in a case. It is necessary to collect multiple lines of evidence for each one. Although the investigator will analyze and score the data for each separately, it is important to consider the consistency, amount, and strength of each line of evidence. The strongest case comes from abundant evidence pointing toward the same conclusion. Too few data weakens a case for or against a stressor. Inconsistencies between lines of evidence do not necessarily weaken the case as long as other evidence backs up explanations for them.

Summary of Step 3

At the end of this step, the investigator should have the following information:

- A summary worksheet (from CADDIS) of the data and analyses (see Appendix A).
- A table of scores demonstrating the relative strength of support for each candidate cause, including information from onsite and offsite.
- A list of candidate causes either eliminated or diagnosed as causes of impairment.

The compilation and relative strength of support for causation are reviewed in Step 4 to determine the likely candidate cause for the biological impairment at the study site.

Step 4: Identify the Probable Cause

The analyses in this step leads to a final statement of causal identification. Aim to provide clear, consistent, and logical rationale for identifying one cause to justify future management activities that address the impairment. Avoid bias to ensure that all candidate causes are equally considered and not overlooked.

The final determination of probable cause(s) is conducted in two parts:

- Weigh the evidence.
- Rank lines of evidence between candidate causes from most to least compelling.

Each originally listed candidate can be considered only if there is a consistent and credible argument in support of its plausibility. After dropping those with a lack of consistent and credible evidence, each of the remaining causes must be compared to determine the most probable cause(s) that led to the specific effects observed at the site of the impairment. If there does not appear to be a readily identified candidate cause, identify what information is known and what is unknown. A list of additional information that could significantly strengthen a determination for a cause is useful.

There are a number of possible outcomes of this process, ranging from straightforward identification of a stressor, to not identifying a stressor and needing to reiterate portions of the process. The desired product is the identification of a cause of biological impairment and a presentation of the evidence supporting that decision. In the final report, the investigator should be able to:

- Report the scores for each type of evidence.
- Evaluate the relative consistency and credibility of each cause based on the scores.
- Classify each candidate cause as diagnosed, likely, unlikely, or uncertain.
- Discuss the way the final conclusions were drawn including the most compelling lines of evidence.

Note: It may be better to make a preliminary report and then reiterate, than to force the identification of a cause that may not be responsible. It is better to defer a determination of an unlikely cause of impairment than to risk an incorrect decision.

Weigh the Body of Evidence for Each Candidate Cause

Weighing the body of evidence determines the relative quality, quantity, consistency, and credibility of each line of support and results in a determination of causation with high confidence. While the investigator cannot prove that a candidate caused the impairment, they can provide strong evidence that it did. They can only refute a cause with confidence when credible evidence demonstrates that a candidate could not have been the cause. Finding the most probable cause involves identifying both probable and uncertain or unlikely causes. This section will help identify both.

Quality and Quantity of Evidence

Recall that the quality and quantity of the data and evidence have influenced the scores that were assigned to each piece of evidence in the last step. It might be helpful to review the evidence evaluation to be sure that all information about the quality of the evidence was considered.

Remember:

- High quality data are always superior to data of uncertain origin or questionable quality.
- Increasing the types or number of pieces of high quality evidence increases confidence in the conclusions about a particular cause, which increases the quality of the evidence.

If a decision is made to drop evidence of questionable quality from the investigation, it is wise to first determine if that data can help establish the type and quality of new data needed to improve the assessment. The more pieces of evidence collected, the less likely that a single faulty study will mislead the investigator.

When determining the quality and quantity of data and evidence, consider the following characteristics:

- The number of pieces of evidence.
- The number of types of evidence.
- Quality of the data.
- Quantity of the data.
- Proper sampling design.
- Relevance of data collected from outside the case to the case at hand.
- Distribution of the data across causes.

Confirm that statistical evaluations are valid by reviewing the sampling design. This will help ensure the study meets the assumptions of the analysis. Also, data most relevant to the case will be of higher quality and value to the study. Finally, make an equal effort to investigate all causes. Uneven distribution of data across potential causes may indicate bias, which must be avoided if possible.

Evaluating Consistency and Credibility

Consistency and credibility of evidence is just as important as the quality and quantity of evidence gathered to support a case. When looking at the evidence as a whole for a particular candidate causes, does it tell a consistent story or is the evidence conflicting? Consistent support for or against a candidate helps ensure that a candidate cause led to the impairment. If a reasonable explanation for an inconsistency exists, and a conceptual or mathematical model can demonstrate the plausibility of that explanation, the determination of the strength of the body of evidence can change. The agreement between a large number of types of evidence gathered for a cause should help to bolster confidence in a determination of probability, as it would be unlikely that several types of evidence lead to the same conclusion by chance.

To evaluate consistency, consider the summary tables produced in the last step. Evaluate each candidate cause individually. Do not add up the scores. Rather than a total score or a total number of supporting pieces of evidence, the investigator is looking for those that consistently support or weaken a candidate cause, and those that may have mixed scores indicating a weaker case. Use the scoring criteria in Appendix B to help describe how the evidence supports or weakens the case for each candidate cause.

From this process, identify the most compelling lines of evidence and determine if there is adequate support for or against a cause. The strongest case generally demonstrates that the candidate meets the characteristics of causal relationships (Table 12) using many lines of evidence. Use the most compelling evidence to convince stakeholders and decision makers that the assessment findings are sound.

Table 12. The six characteristics of causal relationship.*

Characteristics of Causal Relationship	Principle
Time Order	The cause precedes the effect.
Co-Occurrence	An effect consistently occurs where and when its cause occurs and does not occur in its absence.
Sufficiency	The intensity or frequency of a cause is adequate to produce the observed magnitude of effect.
Preceding Causation	Each causal relationship is a result of a larger web of cause-and-effect relationships.
Alteration	The entity is changed by the interaction with the cause.
Interaction	The cause physically interacts with the entity in a way that induces the effect.

*Although there are 15 types of evidence, they generally support six characteristics of causal relationships. The more of these characteristics that are addressed, the stronger the case (Cormier et al., 2010).

The diversity of the above characteristics that are addressed by the evidence as well as data for a candidate cause help describe the strength of the body of evidence. Table 13 lists the potential outcomes based on the strength of the body of evidence for a cause. The weaker candidates are refuted or dropped from consideration in the investigation. The stronger, more compelling evidence will be considered once more for the possibility of multiple causality.

Table 13. Determination of probability of causation for each candidate cause based on strength of evidence (in descending order).

Direction of Support of Evidence	Probability of Causation
Cause refuted by indisputable evidence.	Refuted
Cause of impairment identified by diagnostic symptoms.	Diagnosed
Cause of impairment refuted by diagnostic symptoms.	Refuted
All evidence supports the case for a cause; evidence is of high quality and diverse.	Likely
All evidence weakens the case for a cause; evidence is of high quality and diverse.	Unlikely
All evidence supports the case for a cause, but only a few types of evidence are available.	Probable with low confidence
All evidence weakens the case for a cause, but only a few types of evidence are available.	Unlikely with low confidence
Some evidence supports and some weakens the case for a cause.	Unlikely with low confidence
Insufficient evidence to make a determination.	Additional information required

Compare the Evidence among Causes

After data organization and scoring, the investigator can begin to make a final determination by comparing the strength of evidence across causes. Comparing the evidence allows the investigator to treat each candidate cause fairly and without bias in the data collection and analysis. Any identified bias should be addressed, or at least acknowledged in the report. Furthermore, comparison allows the investigator to determine the cause with the strongest support when evidence is sparse, and to identify the information needed to improve confidence in the assessment conclusions. However, if several impacts or impairments were analyzed (e.g., impairments in the fish, benthic, and algal community were identified), compare data separately for each effect. There may be more than one cause involved, or one cause may be responsible for all of the effects. Because of this, there are many possible outcomes of a SI analysis, as described below.

Typical Outcomes of Stressor Identification (SI) Analysis

There are several outcomes for SI ranging from identifying a single cause, to multiple causes, to no cause identified (Table 14). Depending on the goal(s) of the project, a potential outcome may be reiterating portions of this process if no conclusive determination can be made. Look for unexplored possibilities to find some causal mechanism. Stay objective and consider every option. This will help to determine what the cause is and perhaps lead to suggestions for how to address it.

Table 14. Typical outcomes for Stressor Identification (SI) analysis.

Outcome	Result
1. One candidate diagnosed	Write conclusions and rationale to complete this step.
2. Different impacts due to different causes	Write conclusions and rationale to complete this step.
3. Multiple causes are identified for a single impairment	While it is likely that this could be possible, review the effort to be sure that the impairment was properly defined, and perhaps break the impairment down to component parts. If there are multiple causes, it may be useful to prioritize remediation options.
4. Sparse support for causes	Determine the cause with the strongest support relative to other candidates, while describing uncertainty about others. If there is not one, determine what actions may be taken to improve conditions at the site while continuing with SI.
5. Uneven evidence	Determine if there is a lack of data or if there is some unrecognized bias. If possible, pull more data from outside the study to provide equal investigation of each cause.
6. Insufficient evidence	Consider if the original survey may have been erroneous and mischaracterized the impairment. Consider if there are additional candidate causes, previously unrecognized. Determine whether the event that led to impairment is episodic and if data collection coincided with the events. Possibly resort to best professional judgment, recognizing that the case is not as strong as the investigator would like it to be.
7. No evidence	Use the basic information from the desktop reconnaissance as a "screening level" analysis to suggest what data to collect, and engage stakeholders to see if data collection is needed.

1. One candidate cause is diagnosed or probable, while the other causes considered are unlikely or refuted.

In this case, the conclusion is easy to draw. The investigator writes the conclusions and rationale, resulting in the end of the project. The investigator describes how the conclusions are made regarding both the supported or refuted candidate cause.

2. There is compelling evidence that different impairments were due to different candidate causes while other causes are unlikely or refuted.

The evidence gathered should facilitate documentation and justification of the assessment conclusions. The report should explain how each candidate cause relates to the impairment under investigation. It is also helpful to recommend the best management option(s) for improving site conditions or for reducing the adverse effects of the identified stressor.

3. The evidence suggests that there are multiple causes.

Although it is possible that multiple causes are responsible for the impairment, consider whether the impairment was properly defined. It may be necessary to break the "biological impairment" down into multiple effects. For example, the biological impairment may include fish lesions and changes in invertebrate and periphyton community performance and concentrations (Table 15). These are multiple effects and each may have a different cause.

Table 15. Metrics used to describe biological community performance as a reflection of the environment (Wiseman, 2003; Rhithron, 2009).

Aquatic Macroinvertebrate Metrics	Periphyton Metrics
Species Richness	Disturbance Taxa %
Species Abundance	Dominant Taxon %
Ephemeroptera, Plecoptera, and Trichoptera (EPT) Richness	Eutraphentic Taxa %
% EPT	Low Dissolved Oxygen Taxa %
Ephemeroptera Richness	Metals Tolerant Taxa %
% Ephemeroptera	Motile Taxa %
Plecoptera Richness	Rare Taxa %
% Plecoptera	Native Taxa %
Trichoptera Richness	Nitrogen Autotroph
% Trichoptera	Nitrogen Heterotroph
Clinger Richness	Pollution Index
% Clingers	Siltation Taxa Percent
% Tolerant	Species Richness
% Top 3 Most Abundant	
Long Lived Taxa Richness	
Intolerant Richness	
Total Richness	
% Predators	
Hilsenhoff Biotic Index	
% Filterers	
% Scrapers	
% Chironomidae	
% Predators	

If multiple causes are determined, recommend a remedial strategy based on addressing one of the following:

- The dominant cause: This cause may be sufficient to cause impairment alone, or it may be masking the subtle effects of other causes.
- A necessary cause: If only one of the causes is necessary to lead to the impaired condition, remediate that cause.
- A feasible cause: If it is not clear how multiple causes interact, address the cause whose remediation is easiest to achieve and monitor the results.
- Address all causes if possible.

4. There is sparse evidence across all candidate causes.

If the investigator has collected only sparse evidence for each candidate cause, the investigator may still be able to identify the cause with the strongest support. The candidate that has the highest magnitude, intensity, and duration of exposure relative to effective exposure levels is likely to have caused the observed impact. Discuss the candidate with the strongest support, and describe the uncertainty about the others. If finding a cause is unattainable, at least point out the strongest cause and suggest some remedial action that can improve conditions at the site. In the meantime, the investigator may decide to terminate the study or continue to investigate the possible causes.

5. There is uneven evidence across causes.

Occasionally data supporting or weakening a particular cause is more abundant or easily obtained for some lines of evidence than others. While there may simply be a lack of information for some causes at the study site, stay unbiased and continue to look for adequate evidence, even from outside the site. This is a good opportunity to make the right measurements and use data from historical studies to level out the effort across causes.

6. There is insufficient evidence across all candidate causes.

The investigator may come to the end of this process and find that not enough strong evidence exists to suggest a candidate cause. This may result in a range of responses to this issue depending on time and budgetary resources. This range spans from reiterating the whole process, starting with the listing of new candidate causes, to simply collecting more data for each identified cause. In determining the path forward, there are special considerations:

- Consider whether errors in the biological survey may have resulted in the mischaracterization of the biological effect. For example, the use of the Puget Lowlands multi-metric index in the Columbia plateau may have led to the inaccurate identification of the effect. The investigator should specifically define the biological effect and if possible define more than one effect to make it easier to find relevant evidence.
- Determine if there are other candidate causes that have not yet been considered.
- Consider the possibility of synergistic (jointly acting) events.
- Determine if the data have been collected at the proper times that coincide with the stressing event. Narrow the geographic scope of the assessment, and reassess the types of sources and land uses that might lead to episodic events.
- Resort to best professional judgment to determine the most likely cause, and indicate what new data are needed to make a more confident determination.

In any of these cases, the investigator should document the reasons for the decision and the follow-up activity in the final report.

7. There are insufficient data or no data.

If there are insufficient data to determine a probable cause, the investigator may decide to collect more data. This is particularly important if identification of a cause is imperative because costs of remediation are high or if the situation is contentious.

Depending on budgetary and time constraints or unforeseeable setbacks, the investigator may find that no data exists on which to make a determination. The investigator can still make use of the collected information. At a minimum, land use/land cover information and sources of the impairment within the watershed should be available. This information can be used in a "screening level" SI analysis to identify the most useful data to collect for a more detailed study later. The investigator can then consult with the decision makers to determine if data collection is feasible.

Summary of Step 4

At the end of this step, the investigator should have:

- Relative scores for each type of evidence.
- An evaluation of consistency and credibility of the scores for each candidate cause.
- A classification of each candidate cause as refuted, diagnosed, probable, uncertain, or unlikely.
- A discussion of the reasons for the final conclusion including the most compelling lines of evidence.
- A report describing the causal assessment.

The best strategy for communicating the results of SI analysis depends on the audience, the cost, and the level of contention surrounding the case. A complete report should include:

- The reason for the analysis.
- A list of the candidate causes and the information supporting their selection.
- The sources of the data used in the analysis.
- Tables of evidence derived from the data.
- Conceptual models of causal pathways.
- Key evidence that strengthens the probable cause and weakens other causes.
- Determination of the probable cause(s).
- Qualitative assessment of the overall confidence of the entire case.
- Next steps or recommendations.

A study that is more costly or involves controversial actions or skeptical stakeholders will require more complete documentation. Accurate and defensible identification of the cause of impairment may be important in future actions (e.g., legal, restorative, legislative activity) generated by this study. Investigate as many candidate causes as possible, be vigilant about excluding bias and paying attention to detail, and communicate the reasons behind identifying one probable cause over another. Tables and conceptual models can considerably help illustrate the findings in the final report.

Case Study: Stressor Identification on the Touchet River

Below is an example of how Stressor Identification (SI) was used in Washington State to provide a basic analysis with data that had already been collected. This study (Wiseman et al., 2006) was conducted at the landscape scale, although many other studies have been done for a more local scale where the lines of evidence have been limited to a stream reach or segment. Some of the lines of evidence would be strengthened by collection of additional data. This example shows what can be done with a standard data collection effort.

Step 1: Define the Impairment and Reconnaissance

The Touchet River watershed covers 470,000 acres in southeast Washington State (Figure 5). It is a tributary to the Walla Walla River in the Columbia River Basin. The Walla Walla River headwaters are in the Blue Mountains, which are primarily forested with some small farms. The mainstem of the Touchet River forms upstream of the small town (pop. 2655) of Dayton. The Touchet watershed has been modified by grazing and agriculture since the mid 1800s. The headwaters have been impacted by logging since the 1860s during a heavy period of settlement, affecting salmonid spawning grounds.

Today, logging, agriculture, and channel alterations that provide flood abatement for the towns of Dayton, Waitsburg, and Prescott still influence the river. Agriculture has the largest impact on the health of the river. These impacts take the form of riparian vegetation removal, filling channels, livestock accessing the river, withdrawal and return flows for irrigation, and conversion of native vegetation and habitat to annual cropland.

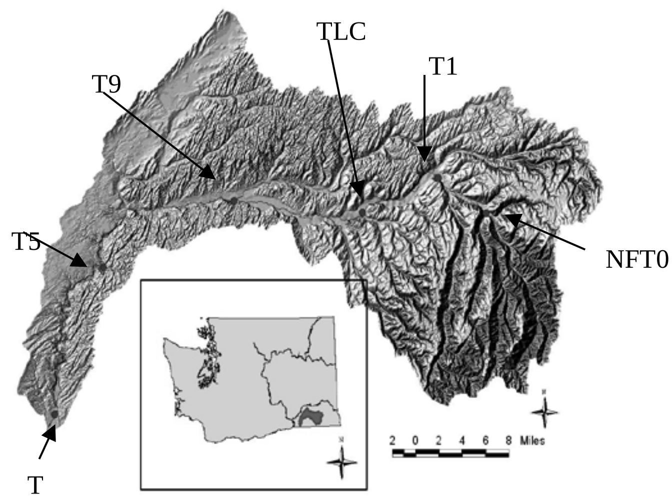
Define the Purpose of the Stressor Identification (SI) Study

In 1998, the Walla Walla River and its tributaries, including the Touchet River, were listed on the 303(d) list for criterion violations in fecal coliform bacteria, temperature, pH, and several pesticide-related chemicals. In addition, declining fish stocks were noted on the Touchet River. The Walla Walla River and its tributaries were scheduled for a “single entry” TMDL, where multiple impairments are addressed by a single TMDL. The SI process was used at the Touchet River to develop effective restoration plans for the river by identifying and prioritizing factors in the Touchet River that cause biological impairment.

Specific Biological Impairment: Degraded Invertebrate Community and Decreased Salmonid Abundance

Two types of biological impairment were identified in the Touchet River. The macroinvertebrate community had been degraded, and there were large decreases or absences of cold water fish species throughout the creek. All biological indicators demonstrated increased impairment from upstream to downstream. Because salmonids were absent from much of the river, benthic macroinvertebrates were also used as an assessment criteria. In particular, Ephemeroptera, Plecoptera, and Trichoptera richness was selected as it provided a strong signal. Ephemeroptera, Plecoptera, and Trichoptera taxa diversity is directly correlated with salmonid abundance; therefore, Ephemeroptera, Plecoptera, and Trichoptera diversity is often used as a surrogate for salmon population condition. The Touchet supports summer steelhead, (*Oncorhynchus mykiss*), and the endangered bull trout, (*Salvelinus confluentus*). Populations of spring chinook, (*Oncorhynchus tshawytscha*) have been extirpated from the Touchet basin since the 1950s.

(a)



T0 – Touchet River Mile (RM) 2.0; T5 – Touchet RM 17.8; T9 Touchet RM 34.2; TLCS – Touchet Lewis and Clark State Park Site RM 47.3; T11 – Touchet RM 53.9; and NFT0 – North Fork of the Touchet River at mouth – RM 58.8.

(b)

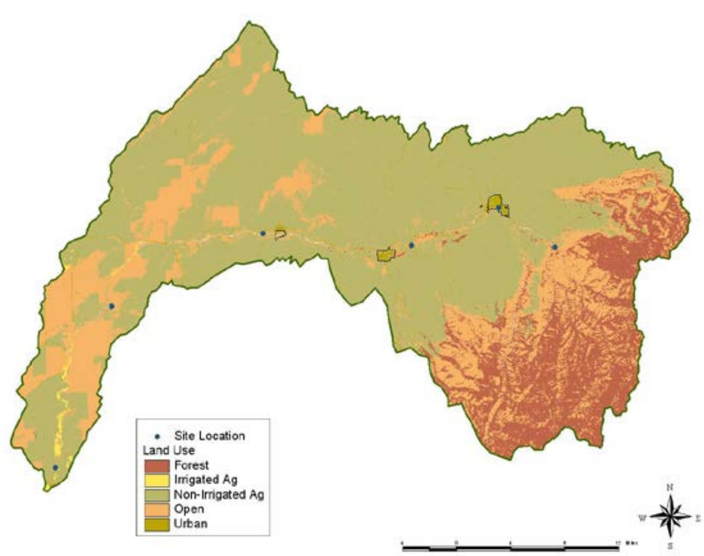


Figure 5. Relief (a) and land use (b) of the Touchet Watershed in Southeast Washington.

Step 2: List Candidate Stressors

Following the SI guidance (EPA, 2000), a list of all possible candidate stressors was compiled (Table 16). A conceptual model (Figure 6) was developed to help guide the formation of hypotheses about the cause of the impairment and guide data analysis. To be included in the analysis, the candidate cause must have been present in the watershed and be known to cause a biological response. Six sources of stressors were identified in the watershed including:

1. Urban development.
2. Wastewater treatment.
3. Forestry practices.
4. Livestock use.
5. Dry land agriculture.
6. Irrigated agriculture.

Urban development often leads to channel alteration and may reduce habitat complexity. Runoff carrying sediments and chemicals also increased with urbanization. Wastewater treatment is another byproduct of urbanization. The only town with a surface connection to the Touchet River is Dayton. Waitsburg and Prescott use an infiltration pond, creating a hyporheic connection. These two towns have not implemented channel alteration, and so they presumably have less impact on habitat.

In more rural areas, livestock are allowed direct access to the river channel. Although this occurs in low densities, the activity leads to the introduction of sediments and nutrients. While historic logging may have long-term effects on habitat and biological communities, the more current source of stress is both dry-land and irrigated agriculture. These sources of stress could lead to the proximate causes listed in Table 16.

Table 16. Table of all candidate stressors identified in the Touchet watershed.

Candidate Cause	Reasoning
Increased Toxic Chemicals	Can result in reduced aquatic species richness.
Reduced Habitat Complexity	Can result in decreased species richness via channel confinement, filling, and reduced amount of detritus.
Increased Sedimentation	Can smother existing aquatic plants, reduce intergravel habitat, and homogenize habitat reducing species richness.
Reduced Detrital Food	Can reduce available food and decrease species richness.
High Temperatures	Can reduce or eliminate cold-water aquatic life.
Increased pH	Can cause issues with ion balance and ammonia excretion resulting in adverse effects to macroinvertebrates.
Low Dissolved Oxygen	Can adversely affect the health of aquatic species.

Although the stressors in Table 16 are focused on effects to macroinvertebrates, they are also detrimental to select species of fish and other aquatic biota. All the connections between the sources of stress and the biological response through the proximate stressors are depicted in the conceptual model below (Figure. 6).

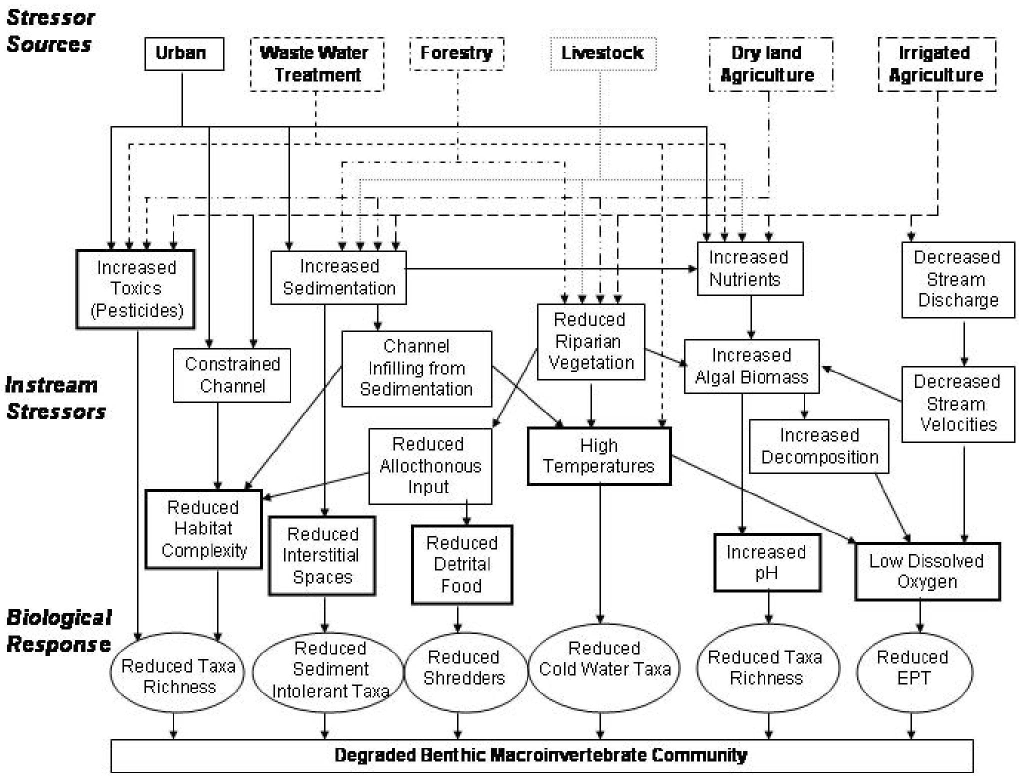


Figure 6. Conceptual model showing the candidate causes for the Touchet River Stressor Identification (SI).

Step 3: Evaluate Data from Within and Outside of the Case

Assemble the Data from Within and Outside of the Case

The Stressor Identification study on the Touchet River was conducted over a period of two years. Six sampling sites were established on the mainstem of the river (Figure 5b). The uppermost stream location on the Touchet River was used to compare with more impaired downstream reaches. Also, data from the Touchet River were compared with data collected from six similar streams that exhibited better biological condition in the Columbia Plateau ecoregion: Quilomene Creek, Oak Creek, Entiat River, North Fork Asotin Creek, Cummings Creek, and Tucannon River.

At each site, data (Table 17) were collected to address the benthic community, water chemistry, physical habitat characteristics, and toxic chemicals.

Table 17. Data parameters measured for the Touchet River Stressor Identification (SI) study in 1998 to capture information about the benthic community, water chemistry, toxic chemicals, and physical habitat characteristics.

Parameter	Method
Temperature, continuous	Onset Temperature Probe
pH	150.1/4500H
Dissolved Oxygen	360.2/4500-OC
Chlorophyll a	/10200H(3)
Ammonia	350.1/4500-NH3D
Nitrate + Nitrate	353.2/4500-NO3F
Orthophosphorus	365.4/4500PF
Total Organic Carbon	415.1/5310B
Total Phosphorus	365.3/4500PF
Total Persulfate Nitrogen	/4500-NC
Total Suspended Solids	160.2/2540D
River Mile	Distance from the mouth
Elevation	ArcView, using Digital Elevation Model
Sinuosity	ArcView, using Rosgen (1996)
Slope	ArcView, using Digital Elevation Model
Median Substrate Size (pebble counts)	Wolman Pebble Counts (Wolman 1954)
Dominant Substrate Size (pebble counts)	Wolman Pebble Counts (Wolman 1954)
Percent Fine Sediment	Wolman Pebble Counts (Wolman 1954)
Direct Sunlight	Solar Pathfinder, Center of Stream
Canopy Cover	Concave Densiometer, Center of Stream
Wetted Width	Tape Measure
Bankfull Width	Tape Measure
Wetted Depth	Average Depth, Stadia Rod
Bankfull Depth	Average Depth, Stadia Rod
Hilsenhoff Biotic Index	Benthic Macroinvertebrate Metric

Parameter	Method
Percent Tolerant	Benthic Macroinvertebrate Metric
Percent Chironomidae	Benthic Macroinvertebrate Metric
Percent Clingers	Benthic Macroinvertebrate Metric
Percent Ephemeroptera	Benthic Macroinvertebrate Metric
Percent Ephemeroptera + Plecoptera + Trichoptera	Benthic Macroinvertebrate Metric
Percent Filterers	Benthic Macroinvertebrate Metric
Percent Predators	Benthic Macroinvertebrate Metric
Percent Scrapers	Benthic Macroinvertebrate Metric
Percent Top 3 Abundant	Benthic Macroinvertebrate Metric
Percent Tolerant	Benthic Macroinvertebrate Metric
Intolerant Richness	Benthic Macroinvertebrate Metric
Clinger Richness	Benthic Macroinvertebrate Metric
Ephemeroptera Richness	Benthic Macroinvertebrate Metric
Ephemeroptera + Plecoptera + Trichoptera Richness	Benthic Macroinvertebrate Metric
Plecoptera Richness	Benthic Macroinvertebrate Metric
Long-Lived Richness	Benthic Macroinvertebrate Metric
Total Richness	Benthic Macroinvertebrate Metric
Trichoptera Richness	Benthic Macroinvertebrate Metric
Percent Tolerant	Benthic Macroinvertebrate Metric
Multi-metric Index	Benthic Macroinvertebrate Metric
Cold-water Taxa Richness	Benthic Macroinvertebrate Metric
Sediment Intolerant Richness	Benthic Macroinvertebrate Metric
Percent Sediment Tolerant	Benthic Macroinvertebrate Metric

Initial Screening of Candidate Causes

When the candidate causes were selected for analysis, some were deferred. The candidates of low dissolved oxygen and toxic chemicals were deferred from consideration because measurements of these parameters in the Touchet River met Washington's water quality criteria. These two candidates were not eliminated because measurements were not continuous, and acute episodic events could not be ruled out (Table 18).

Table 18. Results of data evaluation.

Candidate Cause	Reasoning	Relevance to Touchet
Increased Toxic Chemicals (Deferred from Consideration)	Can result in reduced aquatic species richness.	Toxic chemicals measured met aquatic life criteria, and the Touchet River was noted to have low levels of pesticides and not considered a stressor by toxicologists.
Reduced Habitat Complexity	Can reduce species richness via channel confinement, filling, and reduced amount of detritus.	Sedimentation was occurring, large woody debris counts were depressed, and diking was constraining some reaches. Flow volume was reduced due to water withdrawal for irrigation.
Increased Sedimentation	Can smother existing aquatic plants and homogenize habitat reducing species richness.	Decrease in substrate size class and increase in total suspended solids suggested the potential for sedimentation that may result in reduced interstitial spaces.
Reduced Detrital Food	Can reduce available food and decrease species richness.	Riparian canopy was reduced downstream possibly decreasing allochthonous inputs, resulting in reduced shredder community.
High Temperatures	Can reduce or eliminate cold-water aquatic life.	Temperatures increase to very high levels and a decrease in cold-water obligate benthic macroinvertebrates and fish may occur.
Increased pH	Can cause issues with ion balance and ammonia excretion resulting in adverse effects to macroinvertebrates.	pH levels come close to not meeting aquatic life criteria and may affect the biological community.
Low Dissolved Oxygen (Deferred from Consideration)	Can adversely affect the health of aquatic species.	Available data indicated that dissolved oxygen met criteria for aquatic life use and are not at levels harmful to aquatic organisms in all places where the impairment occurred.

In-depth Analysis

Investigators evaluated the data for each of the remaining candidate causes in the context of evidence:

1. Of a preceding cause that could lead to a candidate cause at the impaired sites.
2. Of an occurrence of the candidate cause where the impairment occurred.
3. Of an alteration in the make-up of the invertebrate assemblage consistent with a candidate cause.
4. That the candidate cause occurred at levels sufficient to cause the observed biological effects to fish or invertebrates.

Evidence of Co-occurrence: Spatial and Temporal Co-occurrence for Invertebrates in the Touchet River

Investigators evaluated three candidate causes for evidence of co-occurrence: increased sediment, high temperature, and pH. One of these, pH, was correlated with biological degradation at Touchet River, but it did not have a noticeable impact on taxa diversity at the test sites compared to reference sites (Table 19). Habitat complexity and reduced detrital food could not be assessed for co-occurrence because there were no available data from the reference sites or previous time periods. For the purposes of this project, data were judged to be adequate for a screening assessment.

Table 19. Summary of co-occurrence analysis.

Candidate Cause	Score*	Reasoning
Reduced Habitat Complexity*	0	No evidence available at reference sites for this stressor.
Increased Sedimentation	+	Perhaps seasonal in nature, but increased sedimentation (as expressed by % fine sediments) co-occurred with a decrease in fine sediment tolerant taxa when compared to 6 regional reference sites.
Reduced Detrital Food	+	General evidence of co-occurrence of detrital material (as expressed by canopy cover) and the percentage of shredders where shredder taxa in the Touchet may be due to lack of riparian cover.
High Temperatures	++	Higher temperatures co-occurred with biological decrease at 5 of 6 sites, compared to reference sites. Cold- water stenothermic fishes also were absent at sites with temperatures higher than reference.
Increased pH	+	Steady increase in maximum pH measured related to biological degradation and co-occurred at 5 of 6 sites when compared to regional reference sites.

*Reduced habitat complexity could not be analyzed as a prospective stressor due to lack of appropriate data from the reference sites.

Evidence of Co-occurrence: Spatial/Temporal Co-Occurrence with Salmonids in Other Rivers.

The presence of salmonids and temperatures in other rivers in the Walla Walla drainage were compared to the Touchet River (Figure 2). Salmonids in other rivers were present in upstream reaches and absent in warmer downstream reaches (Table 20).

Table 20. Evidence of co-occurrence in three streams in the Walla-Walla region similar in size to the Touchet River: presence of salmonids and average and maximum temperatures.

Stream	Average summer temperature (◦C)	Maximum summer temperature (◦C)	Salmonids present	Salmonids in upstream reaches[a]	Summer temperature <18◦C upstream[b]
Lower Coppei Creek	>18	>21	No	Yes	Yes
Dry Creek	>18	>29	No	Yes	Yes
Cottonwood Creek	>21	Not reported	No	NA	NA
Touchet River	>22	>30	No	Yes	Yes
Score[a] for co-occurrence compared to other impaired streams and unimpaired upstream reaches.					+ [c]

[a]Mendel et al. (1999); [b]Mendel et al. (1999, 2002); Kuttell (2001);
[c] This finding somewhat supports the case for the candidate cause, but is not strongly supportive because the association could be coincidental and the conditions in other watersheds may be different from the Touchet River.

Evidence of Alteration (macroinvertebrate symptomatology)

Specific groups of invertebrate taxa were affected that have been shown to be affected by a candidate cause (Table 21). Five of the seven candidate causes were analyzed by comparing relevant biological metrics and stressor indicators of each Touchet River site with the unimpaired upstream site (NFTO) and corresponding ranges of values at the six regional reference sites from a range of elevations.

Macroinvertebrates symptomatic of temperature, sedimentation, and lack of detrital food were simultaneously collected with spatial/temporal co-occurrence of the relevant candidate cause. Temperature intolerant macroinvertebrate taxa were based on descriptions from Brandt (2001). Sediment intolerant macroinvertebrate taxa were developed by Relyea et al. (2000) using biological and physical information for Idaho, Oregon, and Washington streams. For a specific alteration of lack of detrital food, percent shredder taxa were taken from WSDE (Plotnikoff and Wiseman 2001).

Information was not available for dissolved oxygen and pH tolerance for benthic invertebrates; therefore, Plecoptera richness, a taxonomic group particularly intolerant to low dissolved oxygen, was compared with dissolved oxygen measurements. Total taxon richness was compared with pH because alkaline conditions would affect most taxonomic groups. Habitat complexity was not analyzed due to a lack of appropriate data from ecoregional reference sites.

The five stressors—temperature, sediment, pH, low dissolved oxygen, and lack of detritus—were greater at impaired sites than at unimpaired sites and related to specific benthic macroinvertebrate assemblage symptomatology (Table 21). One of these, pH, was elevated only slightly by 0.2–0.4 standard units on average relative to reference sites. Low dissolved oxygen levels were clearly linked with the two furthest downstream sites, T5 and T0.

Table 21. Specific alteration of the macroinvertebrate assemblage and proximate stressors in the Touchet River.[a]

Values in bold differ from reference conditions: specific taxon alteration for temperature, sediment, dissolved oxygen, and lack of detrital food.

Locations	Candidate cause	Specific alteration	Co-occurs	Score[b]
Temperature				++
	◦C	Cold water richness		
Reference[c]	10.5–16.5	5.9		
NFTO	17.7	**4**	no	
T11	**20.6**	6	no	
TLCS	**22.8**	**2**	yes	
T9	**24.5**	**1**	yes	
T5	**25.4**	**2**	yes	
T0	**26.4**	**2**	Yes	
Sedimentation				++
	Substrate class[d]	Sediment intolerant richness		
Reference	9	6–10		
NFTO	11	9	no	
T11	11	8	no	
TLCS	9	6	no	
T9	**3**	**4**	yes	
T5	**1**	**3**	yes	
T0	**7**	**3**	Yes	
Dissolved oxygen(DO) range(mg/l)				+
	mg/l DO *diel* change	Plecoptera richness		
Reference	1.7	1–9		
NFTO	**3.47**	7	no	
T11	**3.73**	6	no	
TLCS	**3.40**	4	no	
T9	**3.66**	1	no	
T5	**4.24**	**0**	yes	
T0	**4.90**	**0**	Yes	
Alkaline pH				+
	pH	Total richness		
Reference	8.0–8.4	30–46		
NFTO	7.8	44	no	
T11	8.3	36	no	
TLCS	**8.5**	**27**	yes	
T9	**8.7**	**27**	yes	
T5	**8.8**	**33**	no	
T0	**8.6**	**28**	Yes	
Lack of detrital food				++
	%canopy cover[e]	% shredders		
Reference	45.5–92.5	1–8		
NFTO	**18.7**	1	no	
T11	**13.8**	**0**	yes	
TLCS	**17**	**0**	yes	
T9	**4**	**0**	yes	
T5	**12.7**	**0**	yes	
T0	**12.2**	**0**	Yes	

(See explanations for this table on the next page.)

Explanations for Table 21:

[a]Locations of the Touchet River sites are shown in Figure 5(a).
[b]Co-occurrence at each site was noted by a "yes" if stressor is greater than regional reference and symptomatic taxa were indicative of the cause.
++ This biological alteration is somewhat specific of the candidate cause.
+ This finding somewhat supports the case for the candidate cause, but is not strongly supportive because symptoms or species are indicative of multiple possible causes.
[c]Ranges for reference are based on data from regional reference sites.
[d]Sedimentation represents the dominant substrate particle size class (larger values represents less fine sediment).
[e] Percent canopy cover over the stream is used as a surrogate indicator for detrital food abundance.

Evidence of Preceding Causation: Causal Pathway Analysis

Five of the six candidate causes were analyzed for evidence of preceding causation by identifying one or more associations with potential sources with the candidate cause. Due to lack of measurements for the intermediate steps, investigators were unable to assess habitat complexity as a potential stressor (Table 22). However, increased sedimentation, reduced detrital food, high temperature, and increased pH were all supported with good confidence by this line of evidence. Several previous studies verified that sediment erosion from croplands was delivering excess fine sediments at a level that was detrimental to aquatic life. Sediment is delivered by both sheet flow and rill erosion.

Two factors support high temperatures as a stress to aquatic life. First, there is no canopy cover to provide shade on the lower reaches where agriculture is practiced right up to the river's edge. Second, there are few groundwater inputs and no major tributaries to the mainstem Touchet River after it reaches the valley from the Blue Mountains. Therefore, there is no way to ameliorate the effects of the hot summer climate on the river environment.

Orthophosphates and pH increase moving downstream. Chlorophyll-a data were taken from the water column, with an expectation that there might be an increase in algal production leading to elevated pH. There was no clear correlation of algal production increases and orthophosphate concentration. While there was no chlorophyll data taken from the benthic samples, it is possible that benthic algae production could drive the pH up as well. Although the algal mechanism for increased pH was shown, the pathway has not been disproved and future measurements of benthic algae production could close this data gap. The trend of increasing pH and decreasing invertebrate diversity is supported by this line of evidence (Table 22). Where trees were absent, the percent shredders were fewer and algal scrapers were greater; however, the evidence was not strong.

Table 22. Summary of complete pathway analysis.

Candidate Cause	Score	Reasoning
Reduced Habitat Complexity	+	Measurements of intermediate steps were not taken.
Increased Sedimentation	++	Total suspended solids increases with flow. Flow is high during winter rain events and spring runoff.
Reduced Detrital Food	+	Canopy cover and the percentage of shredders were somewhat negatively associated in the watershed but measures of detritus not available. Percent algal scrapers is somewhat positively associated with reduced canopy, where increased sunlight reaches the stream bottom. Algal data are not complete.
High Temperatures	++	Riparian canopy is reduced and climate is arid and hot. The stream has little groundwater inputs, resulting in high summer temperatures. Also, higher temperatures were associated with more direct radiation.
Increased pH	+	pH increases downstream. So does orthophosphorus. There is some increase in scrapers; algal data are not complete.

Evidence of Sufficiency: stressor-response relationship from the Touchet River

Within the Touchet watershed, biological indicators responded most strongly to temperature and pH (Table 21). Due to the presence of rip-rap in the lower reaches, sediment conditions were hard to asses. While there was a noticeable trend in decreasing numbers of intolerant taxa with decreasing sediment size, the correlation was not strong. There was no relationship seen between the invertebrate fauna and measures of habitat complexity, and an inverse association between canopy and percent shredders compared to what one would expect. These two stressors (reduced habitat complexity and reduced food resources) were not supported by this line of evidence.

Stressor– response relationships observed in the Touchet River were evaluated by inspecting longitudinal plots of individual stressors and biological effects for each candidate cause (Wiseman et al. 2009) in addition to considering results from rank correlation analysis (Kendall's Tau) (Table 23). The same metrics were used for evaluating macroinvertebrate symptomatology (Table 21) were used in rank correlation analyses. Habitat complexity was evaluated using total richness. The relative strength and the direction of the associations were particularly strong for temperature and somewhat weaker for pH, dissolved oxygen, and sediment. Reduced detritus and habitat complexity were not supported. The results for reduced detritus were probably an artifact of zero values for shredder taxa and therefore were scored as ambiguous.

Table 23. Evidence of sufficiency: stressor response from the Touchet River rank correlations between proximate stressors and biological effects.*

Proximate cause	Measure or surrogate	Biological effect	Kendall's Tau	Stressor-response[a]
Temperature	°C	% Temperature intolerant	−0.85	++
Increased Sedimentation	Dominant size class	Sediment intolerant richness	0.87	+
Dissolved oxygen	Synoptic diel changes (mg/l)	Plecoptera richness	−0.55	+
pH	pH	Total richness	−0.64	+
Detrital food	Percent canopy cover	Percent shredders	−0.54	0[b]
Habitat complexity	Bankfull width/depth	Total richness	−0.33	−
Habitat complexity	Sinuosity	Total richness	−0.25	−

*Score for a ++, $\tau > 0.7$, $p < .05$; This finding strongly supports the case for the candidate cause, but is not convincing due to potential confounding. A strong effect gradient is observed relative to exposure to the candidate cause, at spatially linked sites, and the gradient is in the expected direction;
+, $\tau < 0.7$, $p > .05$ or $\tau > 0.7$, $p < .05$; This finding somewhat supports the case for the candidate cause, but is not strongly supportive due to potential confounding or random error reflected in the weaker gradient;
–, $\tau < 0.5$, $p > .05$; This finding somewhat weakens the case for the candidate cause, but is not strongly weakening due to potential confounding or random error;
b Result inconclusive due to zero values at all sites except site NFTO (see Table 21).

Evidence of Sufficiency: stressor-response relationship from other studies

Stressor-response relationships from other field and laboratory studies were compared to salmonid effects and temperature and dissolved oxygen levels in the Touchet River. Sources of stressor effect levels are noted along with the evidence (Tables 24 and 25). Both temperature and dissolved oxygen levels were suboptimal for salmonids. Laboratory studies for Plecoptera or total taxon richness were not available from other studies**.**

Table 24. Evidence of sufficiency: stressor response from other studies of rainbow and bull trout compared with temperature range (°C) in the Touchet River.
(grab sample 17.7 - 26.4 °C, maximum 30.8 °C).

Species	Adult preference	Upper incipient lethal temperature	Spawning	Rearing	Reference
Rainbow trout	10–13	21–22	13.3	5.6–11.1	Hicks (1998); Bell(1986); Smith et al. (1983)
Bull trout	10–12	19	4–10	< 10	Spence et al. (1996); ODEQ (1995)
Score[a] for stressor response from other studies	+++	+++	+++	+++	

[a] +++ This finding convincingly supports the case for the candidate cause of temperature.
All criteria were not met including for adult incipient lethal temperature. It is not definitive because the correspondence could be coincidental due to confounding or conditions between the case and the laboratory.

Table 25. Evidence of sufficiency: stressor response from other studies of dissolved oxygen (mg/l) tolerance information for salmonids compared with minimum dissolved oxygen measurements in the Touchet River

(Grab samples and diel surveys, 6.45-7.76 mg/l) (Hicks 2002; Spence et al. 1996).

Metric	Salmonidae	Plausible stressor response score[a]
Adult preference	8–9	++
Upper incipient lethal DO	3.3–4.6	–
Migration	6.5–9.0	–
Spawning	8.0–11.5	+++
Rearing	8.0–8.5	+++

[a] On balance, this finding supports the case for the candidate cause of dissolved oxygen, but the upper incipient lethal limit was not observed. It is not definitive because the correspondence could be coincidental due to confounding or differences between conditions in the Touchet River and the laboratory.
+++ Convincingly supports.
++ Strongly supports.
– Somewhat weaken.

Evidence of Co-occurrence: Manipulation of Exposure

Coincidentally, there is a land trust that was taken out of agricultural production and allowed to return to natural vegetation about 90 years ago. The site is at river mile 5 (site T5 in Figure 5a) approximately midway down the mainstem of the Touchet River, with one comparable site upstream (T9) and one comparable site downstream (T0) at the mouth. This provided a “long-term” manipulation of exposure to agriculture and its associated sources of stress (sedimentation and decreased riparian cover).

Results for this line of evidence showed that there was no difference in habitat complexity, temperature, or pH conditions among the three sites (Table 26). This line of evidence provided no support for the influence of riparian cover manipulation over these factors. Sampling occurred in August before autumn leaf fall. Interestingly, though canopy cover was highest at the manipulation site, the % scrapers, (*not* % shredders), increased at this site over the others. This demonstrated that detritus did not have an effect on this particular community.

It may be that the detrital food produced at the site of manipulation traveled downstream before settling to the bottom where shredders could access it. Also, although there may have been more canopy to supply leaves at some sites, the detritus may not have been retained long enough to affect the invertebrate assemblage. This is likely because stream retention of coarse particulate matter increases as stream heterogeneity increases (Allen, 1995).

Perhaps a measurement of the ratio of shredders in the benthic community at an intermediate point downstream of the land trust would provide more insight to this issue. Until then, food resources are not supported as a stressor by this line of evidence. Sedimentation was a stressor that was strongly supported by this line of evidence. As erosion was reduced by a vegetated riparian buffer, sediment size and taxa richness increased (Table 26). This line of evidence was judged as supporting for sediment but weak, negative, or ambiguous for other candidate causes.

Table 26. Causal consideration for manipulation of exposure of site T5 compared to upstream site T9 and downstream site T0 in the Touchet River.

Candidate Cause	Score	Reasoning
Reduced Habitat Complexity	NE	No evidence of manipulation due to no change in proximate causes.
Sedimentation	+++	Increase sediment size and an increase in total taxa richness. All other parameters had no trend strong enough to alter the biological community.
Reduced Detrital Food	–	% canopy cover was highest at T5. However % scrapers, not shredders, increased at T5 suggesting detrital food did not have a substantial effect on the biological community.
High Temperatures	NE	No evidence of manipulation due to no change in proximate causes.
Increased pH	NE	No evidence of manipulation due to no change in proximate causes.

There have been a few regional studies about causes of salmonid decline in the Walla Walla watershed (Table 27). Temperature and pH were the only stressors that could be evaluated with confidence using evidence based on exposure manipulation. However, this evidence came from supporting data from other studies outside of the region. Other stressors considered in this study for comparison were either not commonly measured in other studies or not comparable with other sites due to differences in region specific characteristics. For example, detrital food resources are not commonly considered important in semi-arid stream systems, and so it is rarely measured for these habitats.

Table 27. Summary of regional information about proximate causes of salmonid decline in streams in the Walla Walla watershed including the Touchet River.

Study	Primary Proximate Causes	Secondary Proximate Causes
Current Study	Temperature, Sediment	pH, Habitat Complexity, Riparian/ Detrital
Kuttell (2001)	Screens and Diversions, Riparian Condition, Floodplain Connectivity, Substrate Embeddedness, LWD, Pool Frequency, Off-Channel Habitat, Water Quality/Temperature, Biological Processes	Fish Passage, Streambank Condition, Pool Frequency, Pool Quality, Water Quantity/ Dewatering, Change in Flow Regime
Mendel (2003, 2004)	Temperature	Sediment, Flow, Cover, Pools, etc.
WWWPU (2004)[a]	Sediment	Habitat Quantity, Flow, Predation, Temperature, Channel Stability, Obstructions
WWWPU (2004)[b]	Habitat Diversity, Flow	Temperature, Sediment, Predation, Channel Stability

LWD = Length to Width Ratio.
[a] Ecosystem diagnosis and treatment analysis for steelhead and spring chinook, mouth to Waitsburg.
[b] Ecosystem diagnosis and treatment analysis for steelhead and spring chinook, Waitsburg to Forks.

The local studies generally supported some hypotheses about the impact of sediment, flow rates, and habitat. Two studies (Table 27) conducted by the Walla Walla Watershed Planning Unit (2004) investigated the effects of sediment, water flow, and habitat diversity, while other studies documented the effects of temperature (Mendel, 2000) and various habitat characteristics (Kuttell, 2001).

Identify a Probable Cause

Scoring data for each candidate cause from multiple lines of evidence (Table 28-30) indicate that increased temperature and sediment input are the primary stressors impacting the biological communities of the Touchet River. Elevated pH was also a concern at some locations. These conclusions were based on the number and strength of supporting evidence. Although investigators did not eliminate pH as a primary factor, it seems unlikely to be a direct cause of biological impairment. Detrital food resources in the Touchet River do not appear to drive the biotic community.

There were many issues related to confounding effects of stressor-response gradients by natural instream longitudinal gradients such as elevation, temperature, stream size and land use. However, the use of reference data helped to determine the strength of the stressors over the influence of natural gradients found at the Touchet River. Combined with the results of all lines of evidence (Table 28-30), there is a strong indication that restoration of temperature and sediment regimes would improve biological condition in this river.

Table 28. Summary of strength of relationships between candidate causes and biological observations in the Touchet River using various lines of evidence: C*ase-Specific Considerations.*

Consideration	Evidence	Possible Score	Temperature	Sedimentation	Reduced Detrital Food	Habitat Complexity	pH
Spatial Co-occurrence	Compatible	+	+	+	+		+
	Uncertain	0				0	
	Incompatible	- - -					
Macroinvertebrate Symptomatology	This biological alteration is somewhat specific to the cause.	++				NE	NE
	Somewhat supports the case for the cause, but is not strongly supportive because symptoms or species are indicative of multiple possible causes.		++	+	++		
Biological Gradient	Strong and monotonic	+++	+++				
	Weak or other than monotonic	+					+
	Ambiguous	0		0			
	None or weak but wrong sign	-				-	
	Clear association but wrong sign	- - -			- - -		
Complete Exposure Pathway	Evidence for all steps	++	++	++			
	Incomplete evidence	+			+	+	
	Ambiguous	0					0
	Some steps missing or implausible	-					
Manipulation of Exposure	Removal of the candidate cause eliminated the effect OR the addition of the candidate cause induced the effect	+++		+++			
	Ambiguous experimental results	0	NE			NE	NE
	Removal of the candidate cause did not eliminate the effect OR the addition of the candidate cause did not induce the effect	---			---		

Table 29. Summary of strength of relationships between candidate causes and biological observations in the Touchet River using various lines of evidence: *Evidence from Other Situations or from Biological Knowledge*.

Consideration	Evidence	Score	Temperature	Sedimentation	Reduced Detrital Food	Habitat Complexity	pH
Mechanistically Plausible Causal Pathway	Actual evidence Plausible Not known Implausible	+++ + 0 -	+++	+++	NE	NE	NE
Plausible Effect Given Stressor-Response Relationships	Quantitatively consistent Comparable Ambiguous Not comparable	+++ + 0 -	+	+	NE	NE	NE
Consistency of Association (or: Dichotomous Association)	Invariant In most places In some places Ambiguous At background frequency or many exceptions	+++ ++ + 0 -	++	++	+	+	+
Specificity of Cause	Only possible cause One of a few One of many	+++ ++ 0	0	0	0	0	0

Table 30. Summary of strength, consistency, and coherence of the body of several lines of evidence for each candidate cause and biological condition in the Touchet River: C*onsiderations Based on Multiple Lines of Evidence.*

Consideration	Evidence	Score	Temperature	Sedimentation	Reduced Detrital Food	Habitat Complexity	pH
Total Scores	Positive	+	+	+			+
	Ambiguous/unknown	0			0	0	
	Negative	-					
	No evidence	NE					
Consistency	All consistent	+++					
	Most consistent (some 0, no -)	++	++	++			
	Most consistent (some -)	+					+
	Multiple inconsistencies	---			---	---	
Coherence	Inconsistency explained by a credible mechanism.	+			+	+	+
	No known explanation	0					
Overall Strength of Body of Evidence	High, Medium, Low, No evidence	H,M, L, 0	H	H	L	L	M

Consider Reiteration to Refine Results

Recall that dissolved oxygen (DO) was removed from consideration as a stressor during Step 3 after considering the evidence from the case. Conditions in the environment, including nutrient loading from agricultural run-off, full sun exposure, and low flows, often lead to algal growth and decay, which negatively affect DO levels in the stream. Investigators realized that only instantaneous measurements of DO were taken at the time of the site visit. These measurements did not capture the daily fluctuation in instream conditions.

These types of flags are a good indicator that reiteration may be necessary. After reiteration of the SI process, investigators discovered through continuous DO monitoring that there were sites with large fluctuations in DO. Collection and analysis of further evidence provided weak support for spatial and temporal co-occurrence, and ambiguous support for preceding causation and survival of any species besides salmonids. This evidence is enough to reintroduce DO as a probable cause for decreased salmonid density and survival, but less of a cause for altered invertebrate communities. It was recognized that further data collection was needed and DO was considered as problematic for the lower river (Wiseman et al., 2010)

Conclusions of Case Study

At the beginning of the process, there was an expectation that some of the candidate stressor data might have been limited to a particular year and season (habitat characteristics and pH) and therefore difficult to identify. However, the investigators felt confident at the end of the Touchet River project that they had identified the primary causes of biological impairment.

The investigators suggested that the findings from this study would be applicable to many related projects. For example, findings could be used with TMDL recommendations to reduce pollutant loads and in restoration strategies. Because we know the degree and location of elevated pH, we could include efforts to neutralize pH in restoration efforts to avoid unexpected interactions with other environmental factors. Elevated pH, low dissolved oxygen, high orthophosphate levels, and changes in sedimentation and temperature could interact and lead to otherwise unexpected responses to restoration (i.e. reduction in sediment delivery and decreased temperatures) such as algal blooms and related issues.

After restoration projects, the utility of SI data could continue to be relevant and useful. This work could also assist future effectiveness monitoring of TMDL implementation efforts. Information from an SI study could provide inexpensive and useful information for evaluating implementation effects and pollutant reductions.

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Appendices

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Appendix A. Worksheets to Facilitate Stressor Identification in Washington State

Table A-1. Define Extent of the Study Area

Data Sheets Step 1: DEFINE THE CASE/RECONNAISSANCE – Define Extent of the Study Area

Fill out one sheet for all sites used to define the extent of the impairment.
Identify the site on a map using the Site # assigned below.

Date:_______________ Investigators:_____________________________________

Name of Study:____

Site Type (circle one): **Reference** **Impaired** **Downstream**
Site #: River Mile: Weather at Time of Assessment:

Impairment Location (will be the same for all sites within a study)

County	WRIA	Watershed
River Mile:	GPS Coordinates:	Name of Waterbody:

Biology:
MMI/OE Scores:____________ MMI ____________ OE

Water Chemistry

Parameter	Amount	Notes
Dissolved Oxygen		
pH		
Conductivity		
Temperature		
Ammonia		
Total Phosphorus		
Chloride		
Turbidity		
Chlorophyll a		
Fecal Coliforms		

Channel Condition:

Riparian Condition:

Notes: How does the condition of this site compare with the Impaired site? Describe in terms of Reference or Degree of Disturbance, as well as in terms of the Impairment. The more details the better.

Data Sheets Step 1: DEFINE THE CASE/RECONNAISSANCE – Define Extent of the Study Area

Date:_______________ Investigators:____________________________________

Name of Study:____

Notes on Historical Impacts:

Include date, impact type, affect on stream (if known), and photos (if available).

303(d) impairment listings (if applicable):

Possible Pollution Affecting Site

Pollution Type Historical/Current	Pollution Source	Distance from Site	Map Identifier

Table A-2. Verify Impairment

Data Sheets Step 1: DEFINE THE CASE/RECONNAISSANCE – Verify Impairment

Date:_______________ Investigators:_____________________________________

Nature of the Impairment

Biological Indicators of Impairment At Site: (check all that apply)	
√	**Indicator:**
	Change in indices of biological criteria (MMI/O/E).
	Dead animals in or near water.
	Anomalies in life form (e.g., tumors, lesions, parasites, disease).
	Change in organism behavior.
	Change in area or pattern of ecosystem (e.g., shrinking wetlands).
	Change in Community or population structure: Specify affected metrics (e.g., Ephemeroptera, Plecoptera, and Tricoptera richness, % Chironomids).
	Change in ecosystem function.
Which indicators above are responding most strongly? :	
Magnitude of the Impairment	
Explain the magnitude of the impairment, based on Criteria scores (RIVPACS/MMI) and their component parts, percentage of impacted fish vs. healthy fish, or any visual extent you may see impacted	
Frequency of Impairment: Note what information is available about the frequency of impairment, if it is episodic or if it is persistent, as well as the source of your information:	
Duration of Impairment: Note how long this problem has been known, and the length of time each episode lasts:	
Season of occurrence:	Time of day of occurrence:

Suspected Natural or Anthropogenic Causes:	
Notes:	

Checklist of Documents to include in Reconnaissance:

List of dischargers in the watershed
- Pollutant ID
- Permitted discharge amount
- Location of discharge

Meteorological data
- Average rainfall
- Average high and low air temps
- Average snowfall
- Annual hydrograph of stream or of comparable nearby stream
- Baseflow cfs
- Average high and low water temps

Aerial photo of affected watershed showing:
- Land use
- Dischargers
- Surface water withdrawals
- Hydrologic features
- Historical aerial photos of same area
- Direction of water flow to stream from the landscape

Assessment of the physical integrity of the stream and riparian zone
- Red flag areas
- Specify method used to assess physical integrity(see Table 5)
- Include field sheets
- Include photos of sites
- Suggested monitoring sites:
- At site of impairments
  - Upstream
  - Downstream
  - Tributaries upstream from mouth
  - At your discretion

List of Impairments (from first page). You may have multiple impairments (e.g., fish with lesions and invert community structure). Be sure to complete this section for each impairment.
1)
2)
3)
4)
Impairment Summary:
Impairment:
Notes:

Candidate Causes (check all that apply)
_____Sediment _____Temperature _____Dissolved Oxygen _____Metals _____Nutrients _____Flow Alteration _____Toxic substances _____Ionic Strength _____Other (define):
Impairment Summary:
Impairment:
Notes:
Candidate Causes (check all that apply)
_____Sediment _____Temperature _____Dissolved Oxygen _____Metals _____Nutrients _____Flow Alteration _____Toxic substances _____Ionic Strength _____Other (define):
Impairment Summary:
Impairment:
Notes:
Candidate Causes (check all that apply)
_____Sediment _____Temperature _____Dissolved Oxygen _____Metals _____Nutrients _____Flow Alteration _____Toxic Substances _____Ionic Strength _____Other (define):
Impairment Summary:
Impairment:
Notes:

Candidate Causes (check all that apply)
_____Sediment _____Temperature _____Dissolved Oxygen _____Metals _____Nutrients _____Flow Alteration _____Toxic Substances _____Ionic Strength _____Other (define):

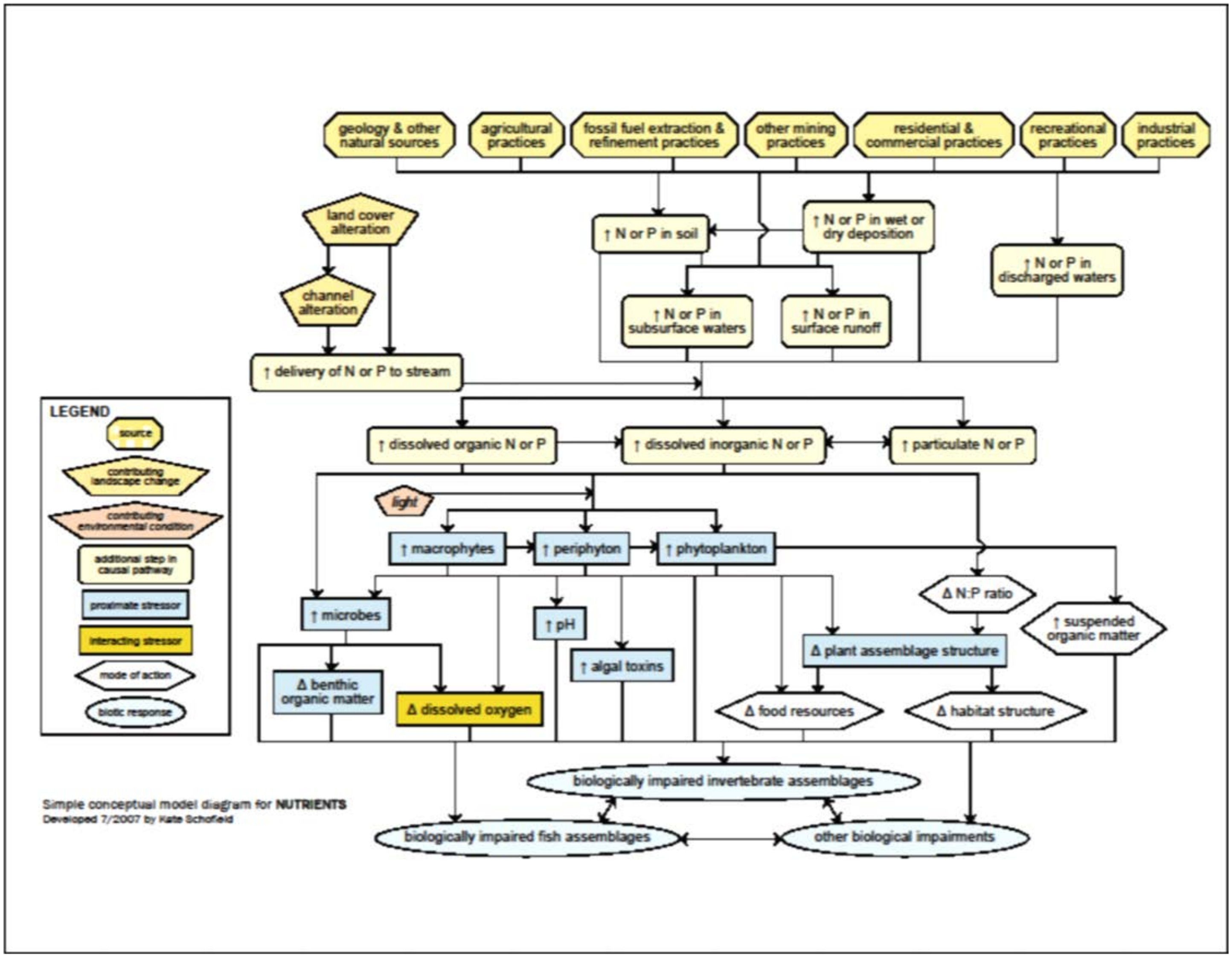

Figure A-1. Conceptual model for each candidate cause for each impairment: Nutrients.

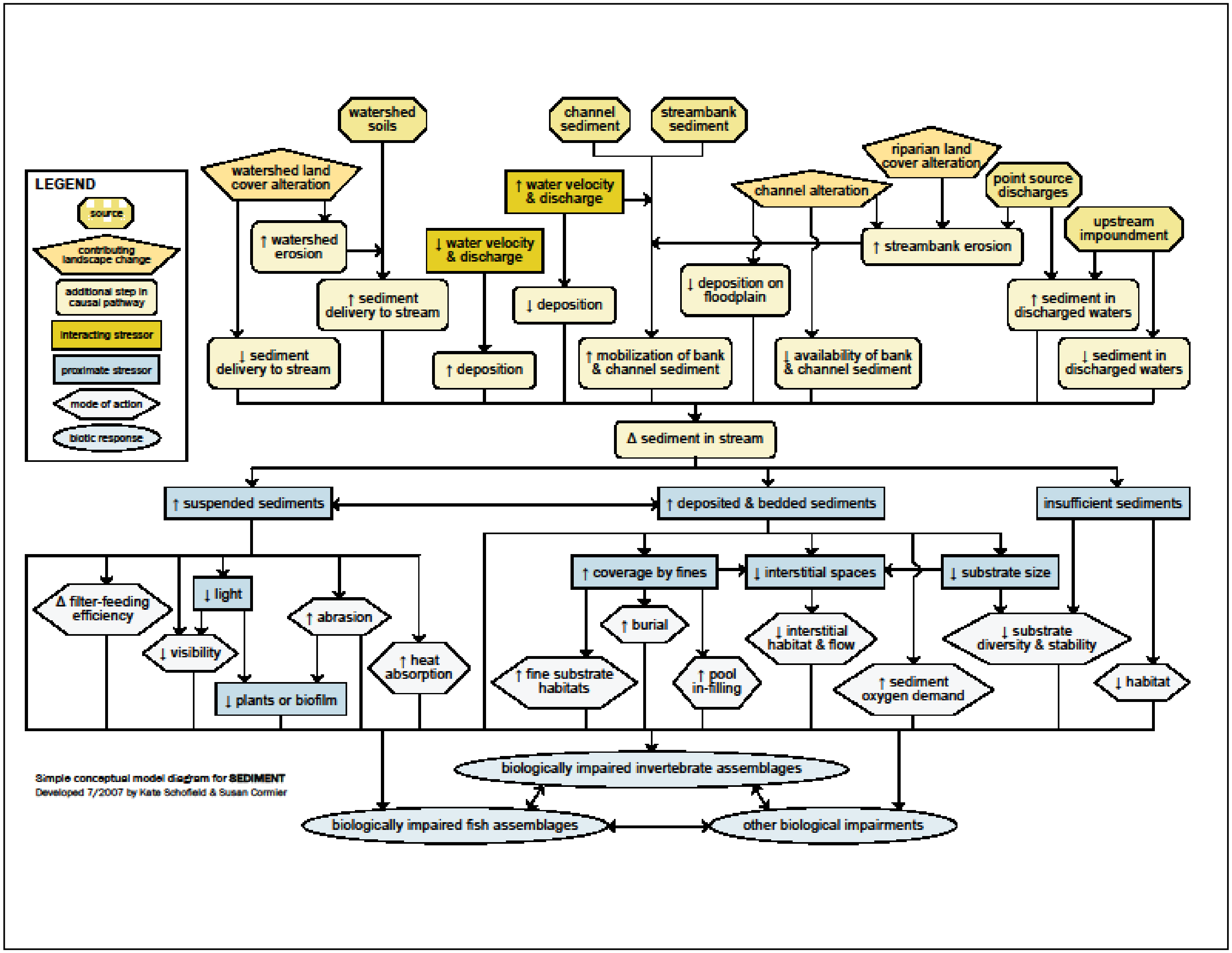

Figure A-2. Conceptual model for each candidate cause for each impairment: Sediment.

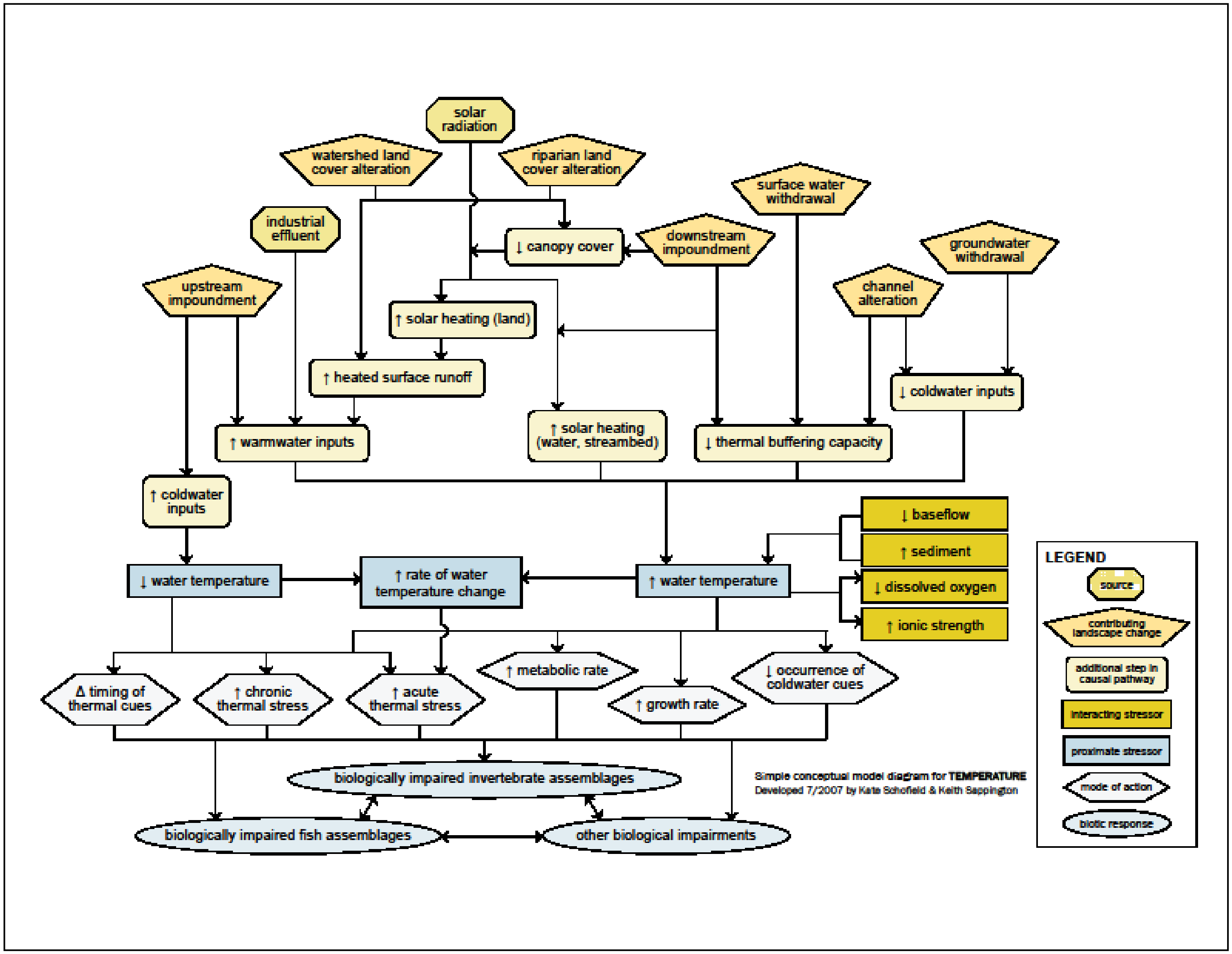

Figure A-3. Conceptual model for each candidate cause for each impairment: Temperature.

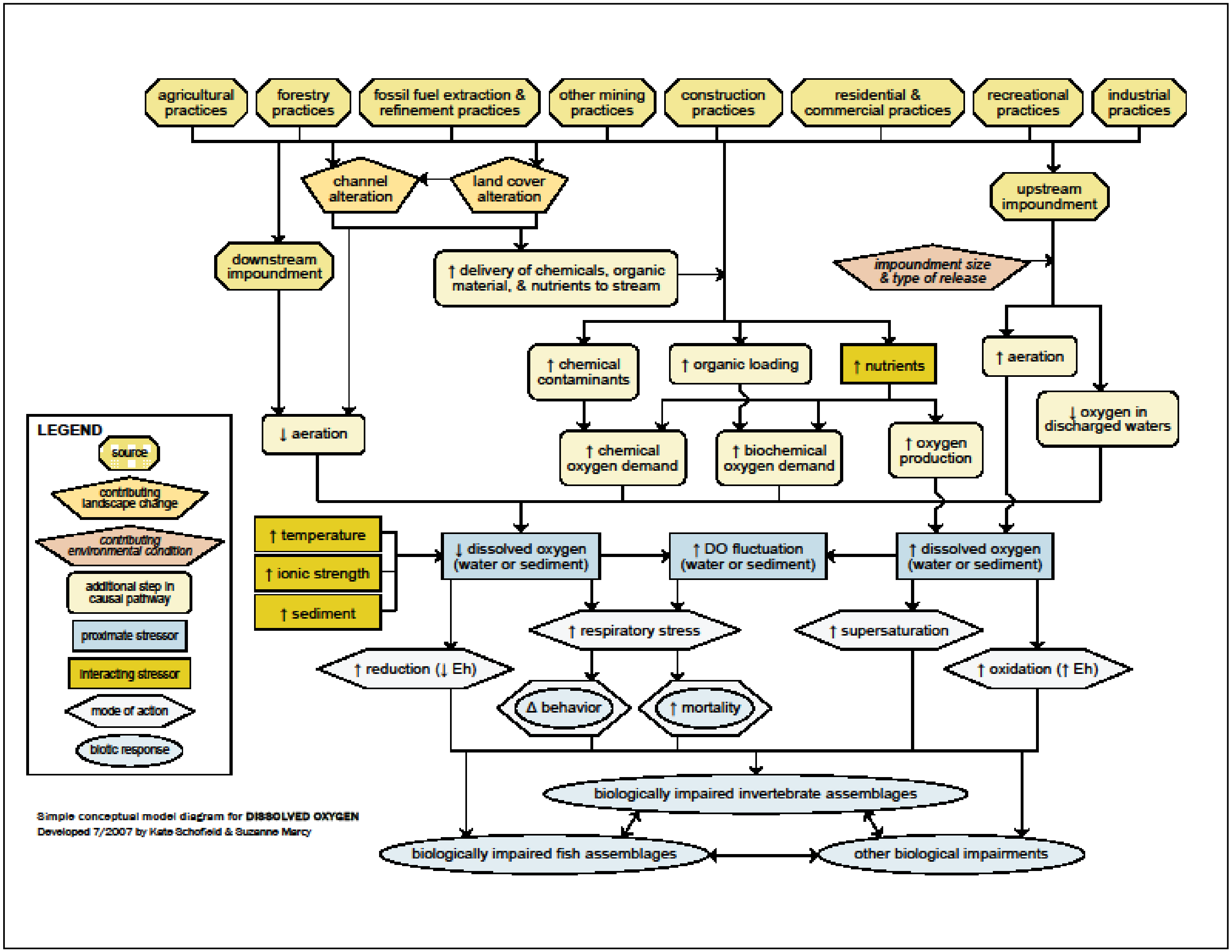

Figure A-4. Conceptual model for each candidate cause for each impairment: Dissolved Oxygen.

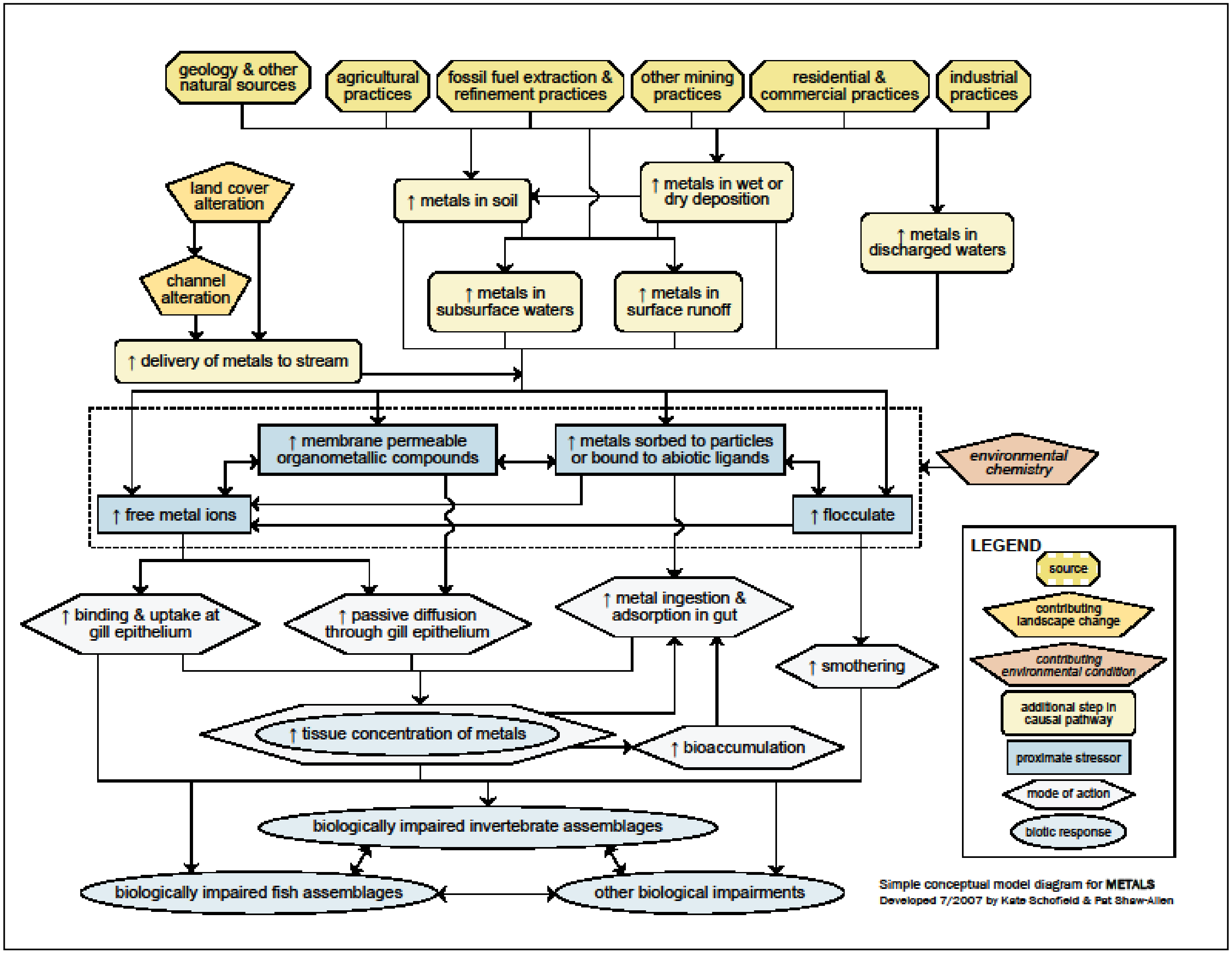

Figure A-5. Conceptual model for each candidate cause for each impairment: Metals.

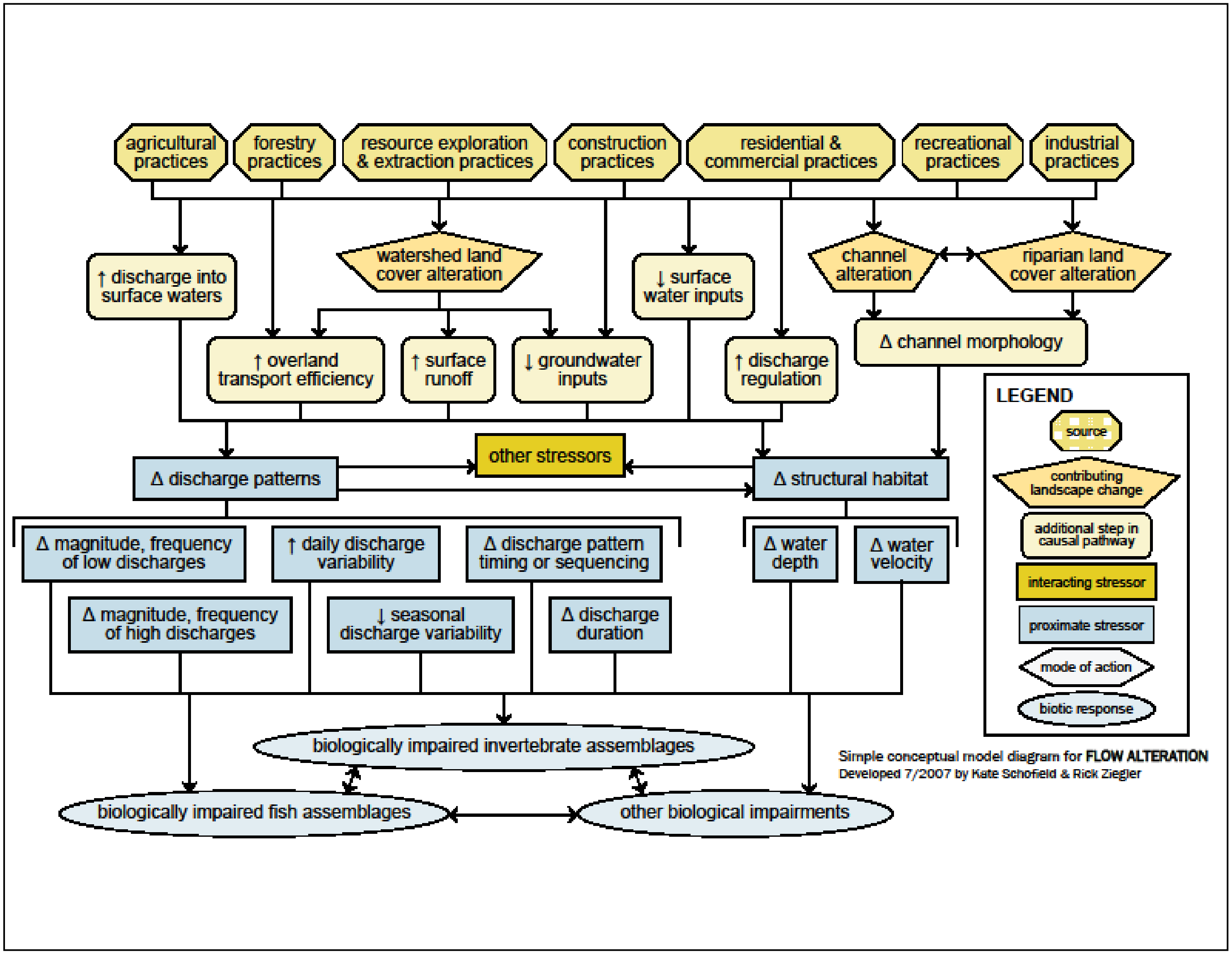

Figure A-6. Conceptual model for each candidate cause for each impairment: Flow Alteration.

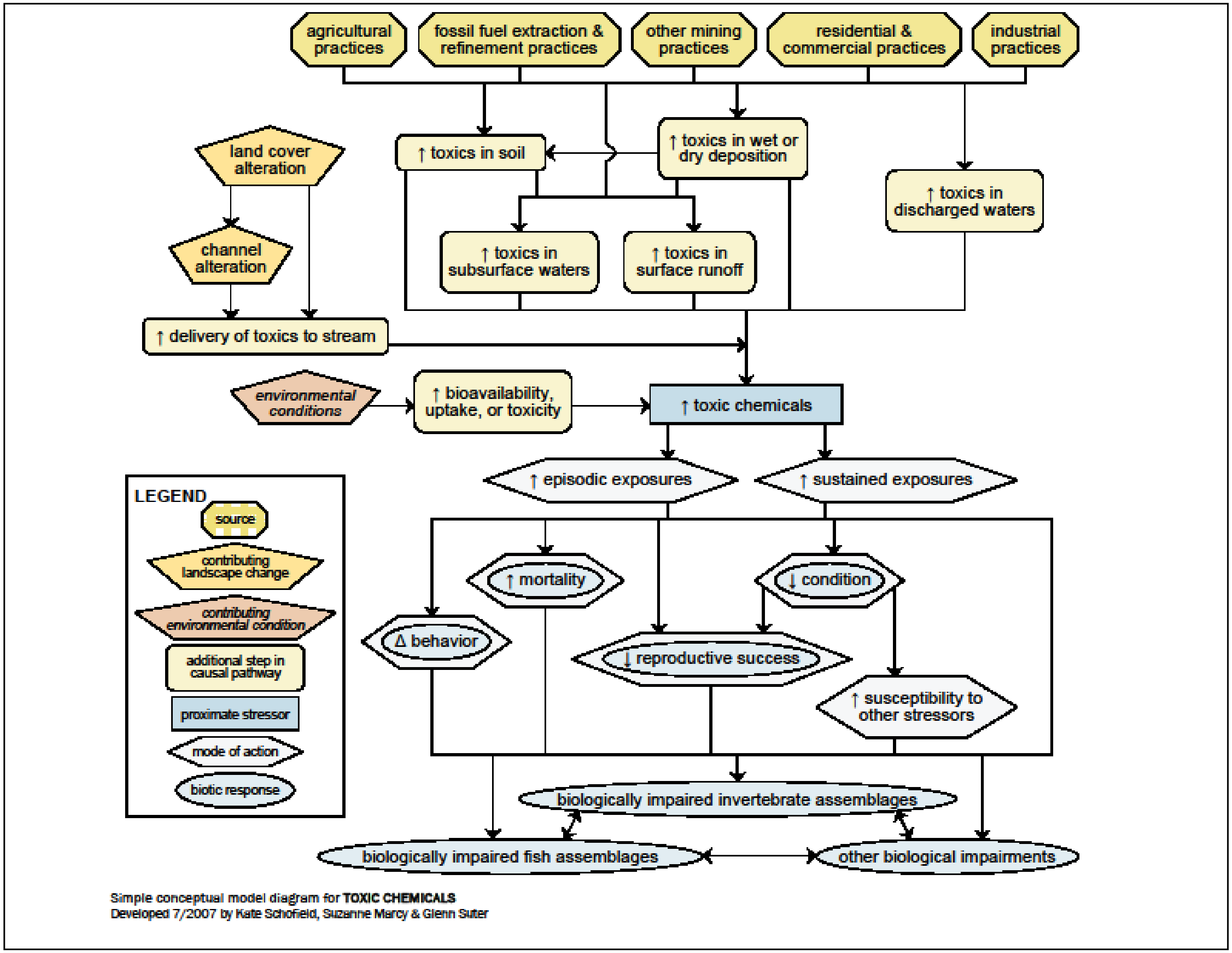

Figure A-7. Conceptual model for each candidate cause for each impairment: Toxic Substances.

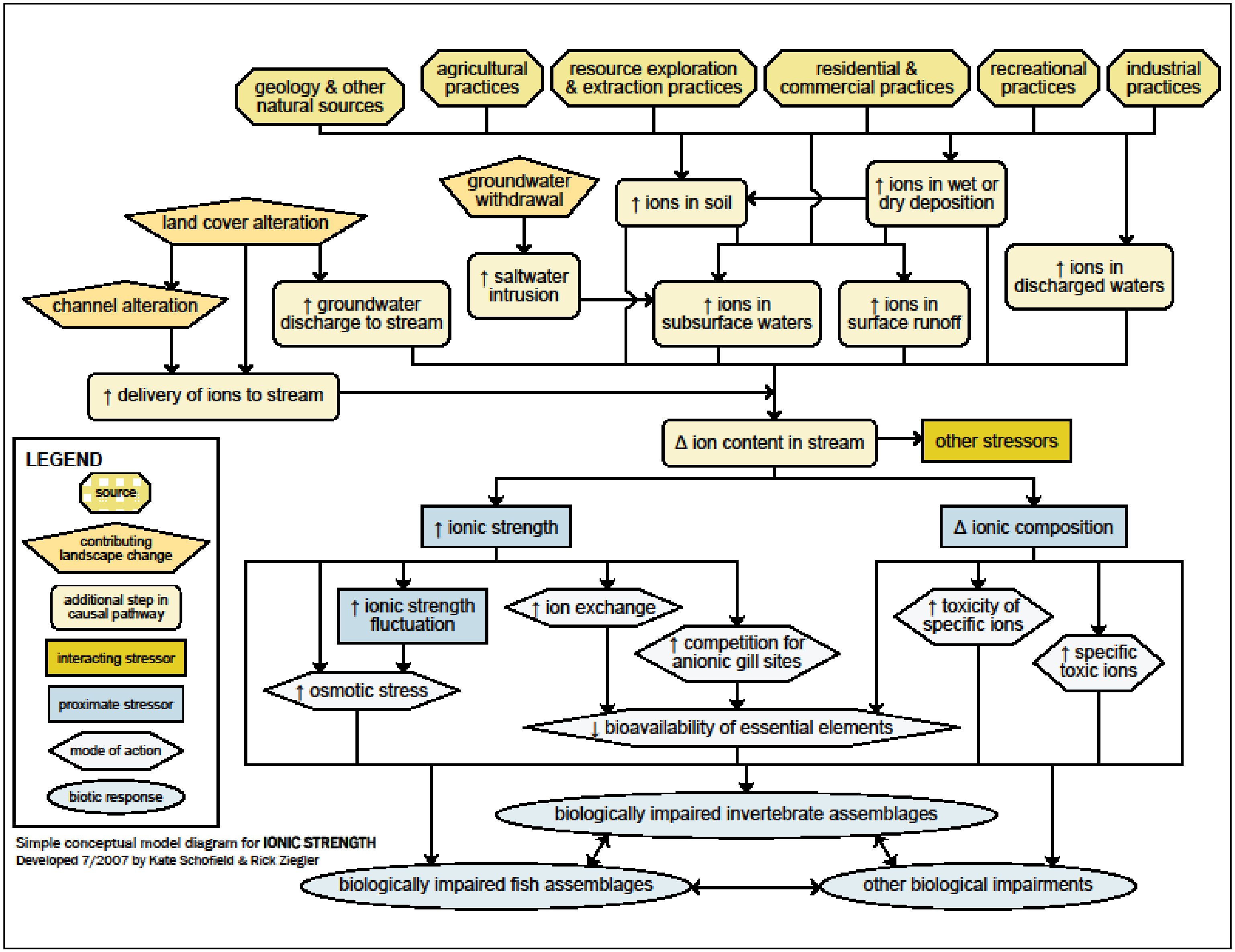

Figure A-8. Conceptual model for each candidate cause for each impairment: Ionic Strength.

(Study author create model here)

Figure A-9. Conceptual model for each candidate cause for each impairment: Other – Define as appropriate to your case.

Table A-3. Considerations to investigate for candidate cause:

From these websites, you should be able to develop a list of data to collect that will support or refute these primary candidate causes. Use this information as a guide to help develop the same list for "other" causes.	
Candidate Cause	**Resource**
Sediment	http://cfpub.epa.gov/caddis/candidate.cfm?section=134&step=24&parent_section=132
Temperature	http://cfpub.epa.gov/caddis/candidate.cfm?section=137&step=24&parent_section=132
Dissolved Oxygen	http://cfpub.epa.gov/caddis/candidate.cfm?section=136&step=24&parent_section=132
Metals	http://cfpub.epa.gov/caddis/candidate.cfm?section=133&step=24&parent_section=132
Nutrients	http://cfpub.epa.gov/caddis/candidate.cfm?section=135&step=24&parent_section=132
Flow Alteration	http://cfpub.epa.gov/caddis/candidate.cfm?section=139&step=24&parent_section=132
Toxic substances	http://cfpub.epa.gov/caddis/candidate.cfm?section=140&step=24&parent_section=132
Ionic Strength	http://cfpub.epa.gov/caddis/candidate.cfm?section=138&step=24&parent_section=132
Map of sources for each candidate cause.	

On the next page, insert an aerial or other map and locate the possible sources for each candidate cause.

Lines of Evidence: Spatial Co-Occurrence					
Site:					
List Candidate Causes Supported by Spatial Co-occurrence at this Site:					
1)					
2)					
3)					
4)					
Candidate Cause	Measurement or Data Collected	Site 1	Upstream	Increase?	Score

Score	Interpretation
D	Diagnoses
+++	Strongly Supports
+	Strengthens Case
0	No effect
-	Weakens case
- - -	Strongly Weakens
R	Refutes
NA	Does not apply to a line of evidence
NE	Can't be analyzed to address a particular type of evidence

Lines of Evidence: **Sequential or Temporal Co-Occurrence**					
Site:					
List Candidate Causes Supported by Spatial Co-occurrence at this Site:					
1)					
2)					
3)					
4)					
Candidate Cause	Measurement	Before	After	Increase?	Score

Score	Interpretation
D	Diagnoses
+++	Strongly supports
+	Strengthens case
0	No effect
-	Weakens case
- - -	Strongly weakens
R	Refutes
NA	Does not apply to a line of evidence
NE	Can't be analyzed to address a particular type of evidence

Lines of Evidence: Exposure
Site:
List Candidate Causes Supported by Spatial Co-occurrence at this Site:
1)
2)
3)
4)

Candidate Cause	Measurement	Site 1	Upstream	Increase?	Score

Score	Interpretation
D	Diagnoses
+++	Strongly supports
+	Strengthens case
0	No effect
-	Weakens case
- - -	Strongly weakens
R	Refutes
NA	Does not apply to a line of evidence
NE	Can't be analyzed to address a particular type of evidence

Lines of Evidence: Causal Pathway					
Site:					
List Candidate Causes Supported by Spatial Co-occurrence at this Site:					
1)					
2)					
3)					
4)					
Candidate Cause	Measurement	Site 1	Upstream	Increase?	Score

Score	Interpretation
D	Diagnoses
+++	Strongly supports
+	Strengthens case
0	No effect
-	Weakens case
- - -	Strongly weakens
R	Refutes
NA	Does not apply to a line of evidence
NE	Can't be analyzed to address a particular type of evidence

Lines of Evidence: Field Response Relationship					
Site:					
List Candidate Causes Supported by Spatial Co-occurrence at this Site:					
1)					
2)					
3)					
4)					
Candidate Cause	Measurement	Site 1	Upstream	Increase?	Score
Score	Interpretation				
D	Diagnoses				
+++	Strongly supports				
+	Strengthens case				
0	No effect				
-	Weakens case				
- - -	Strongly weakens				
R	Refutes				
NA	Does not apply to a line of evidence				
NE	Can't be analyzed to address a particular type of evidence				

Lines of Evidence: Manipulation
Site:
List Candidate Causes Supported by Spatial Co-occurrence at this Site:
1)
2)
3)
4)

Candidate Cause	Measurement	ambient	manipulated level	Increase?	Score

Score	Interpretation
D	Diagnoses
+++	Strongly supports
+	Strengthens case
0	No effect
-	Weakens case
- - -	Strongly weakens
R	Refutes
NA	Does not apply to a line of evidence
NE	Can't be analyzed to address a particular type of evidence

Lines of Evidence: Lab Tests Of Field Collected Media					
Site:					
List Candidate Causes Supported by Spatial Co-occurrence at this Site:					
1)					
2)					
3)					
4)					
Candidate Cause	Measurement	Field measurement	Lab measurement	Result	Score

Score	Interpretation
D	Diagnoses
+++	Strongly supports
+	Strengthens case
0	No effect
-	Weakens case
- - -	Strongly weakens
R	Refutes
NA	Does not apply to a line of evidence
NE	Can't be analyzed to address a particular type of evidence

Lines of Evidence: Verified Predictions					
Site:					
List Candidate Causes Supported by Spatial Co-occurrence at this Site:					
1)					
2)					
3)					
4)					
Candidate Cause	Prediction	Verified	Unverified	Increase?	Score
Score	Interpretation				
D	Diagnoses				
+++	Strongly supports				
+	Strengthens case				
0	No effect				
-	Weakens case				
- - -	Strongly weakens				
R	Refutes				
NA	Does not apply to a line of evidence				
NE	Can't be analyzed to address a particular type of evidence				

Lines of Evidence: Symptoms
Site:
List Candidate Causes Supported by Spatial Co-occurrence at this Site:
1)
2)
3)
4)

Candidate Cause	Symptom	Present	Absent	Increase?	Score

Score	Interpretation
D	Diagnoses
+++	Strongly supports
+	Strengthens case
0	No effect
-	Weakens case
- - -	Strongly weakens
R	Refutes
NA	Does not apply to a line of evidence
NE	Can't be analyzed to address a particular type of evidence

Outside Evidence: List of Supporting Evidence from Studies.				
	Citation	Evidence supported	Summary	Score
1				
2				
3				
4				
5				
6				
7				

Final Scoring: Weigh the number of lines of evidence that are supportive and the strength of their support. Do not just add up the plusses and minuses				
Type of Evidence	**Scores**			
	Candidate #	Candidate #	Candidate #	Candidate #
Spatial Co-Occurrence				
Exposure				
Causal Pathway				
Stressor-Response in Field				
Manipulation of Exposure				
Lab Test of Site Media				
Temporal Sequence				
Verified Predictions				
Symptoms				
Mechanistically Plausible Cause				
Stressor-Response from Other Studies				
Stressor-Response from Lab Studies				
Stressor-Response from Models				
Manipulation of Exposure Elsewhere				
Analogous Stressors				
Consistency of Evidence				
Explanation of Evidence				
TOTAL SCORES				

Score	Interpretation
D	Diagnoses
+++	Strongly Supports
+	Strengthens Case
0	No effect
-	Weakens case
- - -	Strongly Weakens
R	Refutes
NA	Does not apply to a line of evidence
NE	Can't be analyzed to address a particular type of evidence

Appendix B. Scoring Evidence for Stressor Identification in Washington State

Type of Evidence	Finding	Interpretation	Score
Types of Evidence that Use Data from the Case			
Spatial/Temporal Co-occurrence	The effect occurs where or when the candidate cause occurs, OR the effect does not occur where or when the candidate cause does not occur.	This finding *somewhat supports* the case for the candidate cause, but is not strongly supportive because the association could be coincidental.	+
	It is uncertain whether the candidate cause and the effect co-occur.	This finding *neither supports nor weakens* the case for the candidate cause, because the evidence is ambiguous.	0
	The effect does not occur where or when the candidate cause occurs, OR the effect occurs where or when the candidate cause does not occur.	This finding *convincingly weakens* the case for the candidate cause, because causes must co-occur with their effects.	- - -
	The effect does not occur where and when the candidate cause occurs, OR the effect occurs where or when the candidate cause does not occur, and the evidence is indisputable.	This finding *refutes* the case for the candidate cause, because causes must co-occur with their effects.	R
Temporal Sequence	The candidate cause occurred prior to the effect.	This finding *somewhat supports* the case for the candidate cause, but is not strongly supportive because the association could be coincidental.	+
	The temporal relationship between the candidate cause and the effect is uncertain.	This finding *neither supports nor weakens* the case for the candidate cause, because the evidence is ambiguous.	0
	The candidate cause occurs after the effect.	This finding *convincingly weakens* the case for the candidate cause, because causes cannot precede effects. (Note that this should be evaluated with caution when multiple sufficient causes are present.)	- - -
	The candidate cause occurs after the effect, and the evidence is indisputable.	This finding *refutes* the case for the candidate cause, because effects cannot precede causes.	R
Stressor-Response Relationship from the Field	A strong effect gradient is observed relative to exposure to the candidate cause, at spatially linked sites, and the gradient is in the expected direction.	This finding *strongly supports* the case for the candidate cause, but is not convincing due to potential confounding.	+ +
	A weak effect gradient is observed relative to exposure to the candidate cause, at spatially linked sites, OR a	This finding *somewhat supports* the case for the candidate cause, but is not strongly supportive due to potential confounding or random error.	+

Type of Evidence	Finding	Interpretation	Score
	strong effect gradient is observed relative to exposure to the candidate cause, at non-spatially linked sites, and the gradient is in the expected direction.		
	An uncertain effect gradient is observed relative to exposure to the candidate cause.	This finding *neither supports nor weakens* the case for the candidate cause, because the evidence is ambiguous.	0
	An inconsistent effect gradient is observed relative to exposure to the candidate cause, at spatially linked sites, OR a strong effect gradient is observed relative to exposure to the candidate cause, at non-spatially linked sites, but the gradient is not in the expected direction.	This finding *somewhat weakens* the case for the candidate cause, but is not strongly weakening due to potential confounding or random error.	-
	A strong effect gradient is observed relative to exposure to the candidate cause, at spatially linked sites, but the relationship is not in the expected direction.	This finding *strongly weakens* the case for the candidate cause, but is not convincing due to potential confounding.	- -
Causal Pathway	Data show that all steps in at least one causal pathway are present.	This finding *strongly supports* the case for the candidate cause, because it is improbable that all steps occurred by chance. It is not convincing because these steps may not be sufficient to generate sufficient levels of the cause.	+ +
	Data show that some steps in at least one causal pathway are present.	This finding *somewhat supports* the case for the candidate cause.	+
	Data show that the presence of all steps in the causal pathway is uncertain.	This finding *neither supports nor weakens* the case for the candidate cause.	0
	Data show that there is at least one missing step in each causal pathway.	This finding *somewhat weakens* the case for the candidate cause, but is not strongly weakening because it may be due to temporal variability, problems in sampling or analysis, or unidentified alternative pathways.	-
	Data show, with a high degree of certainty, that there is at least one missing step in each causal pathway.	This finding *convincingly weakens* the case for the candidate cause, assuming critical steps in each pathway are known, and are not found at the impaired site after a well-designed, well-performed, and sensitive study.	- - -

Type of Evidence	Finding	Interpretation	Score
Evidence of Exposure or Biological Mechanism	Data show that exposure or the biological mechanism is clear and consistently present.	This finding *strongly supports* the case for the candidate cause, but is not convincing, because it does not establish that the level of exposure or mechanistic action was sufficient to cause the effect.	+ +
	Data show that exposure or the biological mechanism is weak or inconsistently present.	This finding *somewhat supports* the case for the candidate cause.	+
	Data show that exposure or the biological mechanism is uncertain.	This finding *neither supports nor weakens* the case for the candidate cause.	0
	Data show that exposure or the biological mechanism is absent.	This finding *strongly weakens* the case for the candidate cause, but is not convincing, because the exposure or the mechanism may have been missed.	- -
	Data show that exposure or the biological mechanism is absent, and the evidence is indisputable.	This finding *refutes* the case for the candidate cause.	R
Manipulation of Exposure	The effect is eliminated or reduced when exposure to the candidate cause is eliminated or reduced, OR the effect starts or increases when exposure to the candidate cause starts or increases.	This finding *strongly supports* the case for the candidate cause, but is not convincing, because it may result from other factors (e.g., removal of more than one agent or other unintended effects of the manipulation).	+ + +
	Changes in the effect after manipulation of the candidate cause are ambiguous.	This finding *neither supports nor weakens* the case for the candidate cause.	0
	The effect is not eliminated or reduced when exposure to the candidate cause is eliminated or reduced, OR the effect does not start or increase when exposure to the candidate cause starts or increases.	This finding *convincingly weakens* the case for the candidate cause, because such manipulations can avoid confounding. However, effects may continue if there are impediments to recolonization or if another sufficient cause is present.	- - -
	The effect is not eliminated or reduced when exposure to the candidate cause is eliminated or reduced, OR the effect does not start or increase when exposure to the candidate cause starts or increases, and the evidence is indisputable.	This finding *refutes* the case for the candidate cause, given that data are based on a well-designed and well-performed study.	R
Laboratory Tests of Site Media	Laboratory tests with site media show clear biological effects that are closely related to the observed impairment.	This finding *convincingly supports* the case for the candidate cause.	+ + +
	Laboratory tests with site media show ambiguous effects, OR clear effects that	This finding *somewhat supports* the case for the candidate cause.	+

Type of Evidence	Finding	Interpretation	Score
	are not closely related to the observed impairment.		
	Laboratory tests with site media show uncertain effects.	This finding *neither supports nor weakens* the case for the candidate cause.	0
	Laboratory tests with site media show no toxic effects that can be related to the observed impairment.	This finding *somewhat weakens* the case for the candidate cause, but is not strongly weakening, because test species, responses, or conditions may be inappropriate relative to field conditions.	-
Verified Predictions	Specific or multiple predictions of other effects of the candidate cause are confirmed.	This finding *convincingly supports* the case for the candidate cause, because predictions confirm a mechanistic understanding of the causal relationship, and verification of a predicted association is stronger evidence than associations explained after the fact.	+ + +
	A general prediction of other effects of the candidate cause is confirmed.	This finding *somewhat supports* the case for the candidate cause, but is not strongly supportive because another cause may be responsible.	+
	It is unclear whether predictions of other effects of the candidate cause are confirmed.	This finding *neither supports nor weakens* the case for the candidate cause.	0
	A prediction of other effects of the candidate cause fails to be confirmed.	This finding *somewhat weakens* the case for the candidate cause, but is not strongly weakening, because other factors may mask or interfere with the predicted effect.	-
	Multiple predictions of other effects of the candidate cause fail to be confirmed.	This finding *convincingly weakens* the case for the candidate cause.	- - -
	Specific predictions of other effects of the candidate cause fail to be confirmed, and the evidence is indisputable.	This finding *refutes* the case for the candidate cause.	R
Symptoms	Symptoms or species occurrences observed at the site are diagnostic of the candidate cause.	This finding is sufficient to *diagnose* the candidate cause as the cause of the impairment, even without the support of other types of evidence.	D
	Symptoms or species occurrences observed at the site include some but not all of a diagnostic set, OR symptoms or species occurrences observed at the site characterize the candidate cause and a few others.	This finding *somewhat supports* the case for the candidate cause, but is not strongly supportive because symptoms or species are indicative of multiple possible causes.	+
	Symptoms or species occurrences observed at the site are ambiguous or occur with many causes.	This finding *neither supports nor weakens* the case for the candidate cause.	0

Type of Evidence	Finding	Interpretation	Score
	Symptoms or species occurrences observed at the site are contrary to the candidate cause.	This finding *convincingly weakens* the case for the candidate cause.	- - -
	Symptoms or species occurrences observed at the site are indisputably contrary to the candidate cause.	This finding *refutes* the case for the candidate cause.	R
	Symptoms or species occurrences observed at the site are indisputably contrary to the candidate cause.	This finding *refutes* the case for the candidate cause.	R
Types of Evidence that Use Data from Elsewhere			
Mechanistically Plausible Cause	A plausible mechanism exists.	This finding *somewhat supports* the case for the candidate cause, but is not strongly supportive, because levels of the agent may not be sufficient to cause the observed effect.	+
	No mechanism is known.	This finding *neither supports nor weakens* the case for the candidate cause.	0
	The candidate cause is mechanistically implausible.	This finding strongly weakens the case for the candidate cause, but is not convincing because the mechanism could be unknown.	- -
Stressor-Response Relationships from Laboratory Studies	The observed relationship between exposure and effects in the case agrees quantitatively with stressor-response relationships in controlled laboratory experiments.	This finding *strongly supports* the case for the candidate cause, but is not convincing, because the correspondence could be coincidental due to confounding or differences in organisms or conditions between the case and the laboratory.	+ +
	The observed relationship between exposure and effects in the case agrees qualitatively with stressor-response relationships in controlled laboratory experiments.	This finding *somewhat supports* the case for the candidate cause, but is not strongly supportive, because the correspondence is only qualitative, and the degree of correspondence could be coincidental due to confounding or differences in organisms or conditions between the case and the laboratory.	+
	The agreement between the observed relationship between exposure and effects in the case and stressor-response relationships in controlled laboratory experiments is ambiguous.	This finding *neither supports nor weakens* the case for the candidate cause.	0
	The observed relationship between exposure and effects in the case does not agree with stressor-response relationships in controlled laboratory experiments.	This finding *somewhat weakens* the case for the candidate cause, but is not strongly weakening, because there may be differences in organisms or conditions between the case and the laboratory.	-

Type of Evidence	Finding	Interpretation	Score
	The observed relationship between exposure and effects in the case does not even qualitatively agree with stressor-response relationships in controlled laboratory experiments, or the quantitative differences are very large.	This finding *strongly weakens* the case for the candidate cause, but is not convincing, because there may be substantial and consistent differences in organisms or conditions between the case and the laboratory.	- -
Stressor-Response Relationships from Other Field Studies	The stressor-response relationship in the case agrees quantitatively with stressor-response relationships from other field studies.	This finding *strongly supports* the case for the candidate cause, but is not convincing, because the correspondence could be coincidental due to confounding or differences in organisms or conditions between the case and elsewhere.	+ +
	The stressor-response relationship in the case agrees qualitatively with stressor - response relationships from other field studies.	This finding *somewhat supports* the case for the candidate cause, but is not strongly supportive, because the correspondence is only qualitative, and the degree of correspondence could be coincidental due to confounding or differences in organisms or conditions between the case and elsewhere.	+
	The agreement between the stressor-response relationship in the case and stressor-response relationships from other field studies is ambiguous.	This finding *neither supports nor weakens* the case for the candidate cause.	0
	The stressor-response relationship in the case does not agree with stressor-response relationships from other field studies.	This finding *somewhat weakens* the case for the candidate cause, but is not strongly weakening, because there may be differences in organisms or conditions between the case and elsewhere.	-
	There are large quantitative differences or clear qualitative differences between the stressor-response relationship in the case and the stressor-response relationships from other field studies.	This finding *strongly weakens* the case for the candidate cause, but is not convincing, because there may be substantial and consistent differences in organisms or conditions between the case and elsewhere.	- -
Stressor-Response Relationships from Ecological Simulation Models	The observed relationship between exposure and effects in the case agrees with the results of a simulation model.	This finding *somewhat supports* the case for the candidate cause, but is not strongly supportive, because models may be adjusted to simulate the effects.	+
	The results of simulation modeling are ambiguous.	This finding *neither supports nor weakens* the case for the candidate cause.	0
	The observed relationship between exposure and effects in the case does not agree with the results of simulation modeling.	This finding *somewhat weakens* the case for the candidate cause, but is not strongly weakening, because it may be due to lack of correspondence between the model and site conditions.	-

Type of Evidence	Finding	Interpretation	Score
Manipulation of Exposure at Other Sites	At other sites, the effect is consistently eliminated or reduced when exposure to the candidate cause is eliminated or reduced, OR the effect consistently starts or increases when exposure to the candidate cause starts or increases.	This finding *convincingly supports* the case for the candidate cause, because consistent results of manipulations at many sites are unlikely to be due to chance or irrelevant to the site being investigated.	+ + +
	At other sites, the effect is eliminated or reduced at most sites when exposure to the candidate cause is eliminated or reduced, OR the effect starts or increases at most sites when exposure to the cause starts or increases.	This finding *somewhat supports* the case for the candidate cause, but is not strongly supportive, because consistent results of manipulation at one or a few sites may be coincidental or irrelevant to the site being investigated.	+
	Changes in the effect after manipulation of the candidate cause are ambiguous.	This finding *neither supports nor weakens* the case for the candidate cause.	0
	At other sites, the effect is not consistently eliminated or reduced when exposure to the cause is eliminated or reduced, OR the effect does not consistently start or increase when exposure to the cause starts or increases.	This finding *strongly weakens* the case for the candidate cause, but is not convincing, because failure to eliminate or induce effects at one or a few sites may be due to poorly conducted studies, or results may be irrelevant due to differences among sites.	- -
Analogous Stressors	Many similar agents at other sites consistently cause effects similar to the impairment.	This finding *strongly supports* the case for the candidate cause, but is not convincing, because of potential differences among the agents or in conditions among the sites.	+ +
	One or a few similar agents at other sites cause effects similar to the impairment.	This finding *somewhat supports* the case for the candidate cause, but is not strongly supportive, because of potential differences among the agents or in conditions among the sites.	+
	One or a few similar agents at other sites do not cause effects similar to the impairment.	This finding *somewhat weakens* the case for the candidate cause, but is not strongly weakening, because of potential differences among the agents or in conditions among the sites.	-
	Many similar agents at other sites do not cause effects similar to the impairment.	This finding *strongly weakens* the case for the candidate cause, but is not convincing, because of potential differences among the agents or in conditions among the sites.	- -
	Many similar agents at other sites do not cause effects similar to the impairment.	This finding *strongly weakens* the case for the candidate cause, but is not convincing, because of potential differences among the agents or in conditions among the sites.	- -
Evaluating Multiple Lines of Evidence			

Type of Evidence	Finding	Interpretation	Score
Consistency of Evidence	All available types of evidence support the case for the candidate cause.	This finding *convincingly supports* the case for the candidate cause.	+ + +
	All available types of evidence weaken the case for the candidate cause.	This finding *convincingly weakens* the candidate cause.	- - -
	All available types of evidence support the case for the candidate cause, but few types are available.	This finding *somewhat supports* the case for the candidate cause, but is not strongly supportive, because coincidence and errors may be responsible.	+
	All available types of evidence weaken the case for the candidate cause, but few types are available.	This finding *somewhat weakens* the case for the candidate cause, but is not strongly weakening, because coincidence and errors may be responsible.	-
	The evidence is ambiguous or inadequate.	This finding *neither supports nor weakens* the case for the candidate cause.	0
	Some available types of evidence support and some weaken the case for the candidate cause.	This finding *somewhat weakens* the case for the candidate cause, but is not convincing, because a few inconsistencies may be explained.	-
Explanation of the Evidence	There is a credible explanation for any negative inconsistencies or ambiguities in an otherwise positive body of evidence that could make the body of evidence consistently supporting.	This finding can save the case for a candidate cause that is weakened by inconsistent evidence. However, without evidence to support the explanation, the cause is barely strengthened.	+ +
	There is no explanation for the inconsistencies or ambiguities in the evidence.	This finding neither strengthens nor weakens the case for a candidate cause.	0
	There is a credible explanation for any positive inconsistencies or ambiguities in an otherwise negative body of evidence that could make the body of evidence consistently weakening.	This finding further weakens an inconsistent case. However, without evidence to support the explanation, the cause is barely weakened.	-

Appendix C. Additional Tools to Facilitate Stressor Identification in Washington State

This information is taken in part from the LaMotte Company's, The Monitor's Handbook, 1992.

pH	Effect on Aquatic Species
3.0-3.5	Unlikely that fish can survive for more than a few hours in this range although some plant and invertebrates can be found at pH levels this low.
3.5-4.0	Known to be lethal to all salmonids.
4.0-4.5	All fish, most frogs and insects are not present.
4.5-5.0	Mayfly and many other insect species are not found. Most fish eggs will not hatch.
5.0-5.5	Bottom-dwelling decomposing bacteria begin to die off. Leaf litter and dead plant and animal materials begin to accumulate. Plankton begin to disappear.
6.0-6.5	Freshwater shrimp are not present.
6.5-8.5	**Optimal for most organisms.**
8.5-9.0	Unlikely to be harmful to fish, but indirect effects from chemical changes in the water may occur.
9.0-10.5	Harmful to perch and salmonids if prolonged exposure.
10.5-11.0	Prolonged exposure is lethal to carp and perch.
11.0-11.5	Lethal to all species of fish.

Appendix D. Glossary

This Glossary is taken from the CADDIS website (http://cfpub.epa.gov/caddis/info_sources.cfm?Section=30&From=A&To=Z)

Term	Definition
Agent	A physical, chemical or biological entity that may affect a biotic system positively or negatively. This term is similar to but more general than *stressor*. For example, dissolved oxygen and woody debris are agents; low dissolved oxygen and reduced woody debris may be stressors.
Anthropogenic	Induced by humans.
Associations	Relationships between different types of observations; these relationships become *lines of evidence* supporting or weakening the case for a *candidate cause*.
Benthic invertebrates	Bottom-dwelling organisms without backbones (e.g., aquatic insects, crustaceans, worms).
Biota	Flora (plants) and fauna (animals).
Biotic	Produced or caused by living organisms.
Bioassessment (biological assessment)	Evaluation of ecosystem condition using biological surveys and other direct measurements of resident biota.
Biocriteria (biological criteria)	Numerical values or narrative expressions describing the *reference* biological condition of aquatic communities inhabiting waters of a given designated aquatic life use. Biocriteria are benchmarks for evaluation and management of water resources.
Biological gradient	A regular increase or decrease in a measured biological attribute with respect to space (e.g., below an outfall), time (e.g., since a flood), or an environmental property (e.g., temperature). Biological gradients are analyzed to generate *stressor-response* relationships based on field data.
Biological mechanism	The process by which a *cause* induces a biological *effect*. A biological mechanism is a *causal mechanism* emphasizing biological processes.
CADDIS	The Causal Analysis/Diagnosis Decision Information System, a web-based technical support system for implementing the *Stressor Identification* process.
CERCLA	Comprehensive Environmental Response, Compensation, and Liability Act
Candidate cause	A hypothesized cause of an environmental *impairment*, that is sufficiently credible to be analyzed.
Case	The situation that is the subject of a causal analysis. For example, the case may be an impaired stream reach and the upstream and downstream reaches, or an estuary and the lower reaches of its tributaries.
Case study	An example illustrating a complete *causal analysis* or a component of the process.
Causal analysis	A process by which data and other information are organized and evaluated, using quantitative and logical techniques, to determine the likely *cause* of an observed condition.
Causal mechanism	The process by which a *cause* induces an *effect*.
Causal pathway	The sequence of processes and states that causally connect a *source* to *exposure* to a *candidate cause*, potentially including release, transport, transformation, and direct effects (if the effect of concern is indirect).
Causal relationship	The relationship between a *cause* and its *effect*.
Cause	1. That which produces an *effect* (a general definition). 2. A *stressor* or set of stressors that occurs at an intensity, duration, and frequency of exposure sufficient to result in a change in an identified biological effect (a definition specific to *Stressor Identification*).
Co-occurrence	The spatial or temporal co-location of the *candidate cause* and the *effect*. Synonymous with *spatial/temporal co-occurrence*.
Coherence	See *reasonable explanation*.
Conceptual model	A graphic depiction of the *causal pathways* linking *sources* and *effects*, that ultimately is used to communicate why some pathways are unlikely and others are very likely.

Term	Definition
Consistency of evidence	The degree to which *types of evidence* in a *strength-of-evidence analysis* are in agreement in either supporting or weakening the case for a *candidate cause*.
Control	The treatment in a toxicity test or other experiment in which the test chemical or other experimental condition is absent. *Reference* implies comparison without control. Thus *reference*, rather than control is the appropriate term for observational studies.
Correlation	A statistical relationship between two or more variables such that systematic changes in the value of one variable are accompanied by systematic changes in the other.
Define the Case	A step in the *Stressor Identification* process in which the *impairment* and its spatial and temporal scope are defined. Also see *case*.
Designated use	Terminology used in the Clean Water Act to describe classes of expectations for waterbodies, including their ability to support aquatic life.
Detrital	Weathered mineral and organic particles – organisms may feed on detrital material or live in detrital habitats.
Diagnosis	A type of *inference* in the *Stressor Identification* process that uses *symptomatology* or a set of specific observations to identify a *probable cause*.
Direct causation	The induction of an *effect* through a single cause-effect relationship; for example, the direct effect of an herbicide may be reduced algal production. Compare this to *indirect causation*.
Ecoregion	A geographic area having relatively uniform ecological properties.
Effect	In general, an effect is something that inevitably follows an antecedent (cause or agent). A biological effect is the biological result of *exposure* to a *candidate cause*. This term is similar to *response*, but emphasizes the *agent* that acts (e.g., the effect of cadmium) rather than *receptor* that responds to it (e.g., the response of trout).
Elimination	Rejection of a *candidate cause* based on *evidence* that an expected *association* between that *cause* and the *effect* does not occur.
Eutrophication	Enrichment of a waterbody with nutrients, often resulting in high levels of primary production and leading to depletion of dissolved oxygen.
Evaluate Data	A step in the *Stressor Identification* process in which data are analyzed to generate *associations* constituting *types of evidence*, and these results are then scored.
Evidence	1. Knowledge that changes one's degree of belief in a proposition (a general definition). 2. Results of data analysis concerning *associations* between the *candidate cause* and the *effect*, or between *sources* or steps in the causal chain and the candidate cause (a definition specific to *Stressor Identification*).
Evidence from the case	*Evidence* based on data or observations from the impaired system or *reference* systems that are adjoining or closely spatially related (e.g., reaches in the same stream or watershed).
Evidence of exposure	*Evidence* indicating that organisms took up or contacted a *stressor*.
Experiment	Manipulation of a *candidate cause* through elimination of a *source* or alteration of *exposure*, to evaluate the candidate cause's relationship to an *effect*.
Exposure	The co-occurrence or contact of a *stressor* with the biological resource demonstrating *impairment*.
Exposure-response	1. The relationship between the intensity, frequency, or duration of exposure to a *stressor* and the intensity, frequency, or duration of the biological response. 2. A model of that relationship. This term is similar to *concentration-response* and *stressor-response*.
Field studies	Observational or experimental studies carried out in nature.
Goodness of Fit Tests	A quantification of how well a statistical model describes a set of observations. Usually, such tests quantify the differences between observed values and those expected under a model of interest and calculate the probability the observed differences would occur if there were actually no difference between the observed and expected values.
Hypothesis	A proposed theory concerning a *causal relationship*; for example, identification of a *candidate cause* for *impairment* constitutes a causal hypothesis. Note that these are not statistical hypotheses.
Impairment	A detrimental effect on the biological integrity of a waterbody that prevents attainment of the designated use.

Term	Definition
Indirect effect	Change in a resource due to *indirect causation*.
Inference	The act of reasoning from *evidence*.
Life history	Developmental processes and behaviors that sustain and reproduce a species. For example, case formation and net-spinning can be components of the life history of certain caddisflies.
Manipulation of exposure	A *type of evidence* in which human action induces, eliminates, or modifies *exposure* to a *stressor* (e.g., shutting down an effluent source, fencing cattle from a stream, or caging fish in a contaminated lake).
Mechanism	The process by which a system is changed.
Natural	A state that occurs in the absence of human actions; natural conditions can be approximated but never achieved in the real world.
Nonpoint	Pollutants discharged from large area or from several small inputs rather than from one distinct source.
Parameter	Water quality constituent being measured (analyte). A physical, chemical, or biological property whose values determine environmental characteristics or behavior.
Piece of evidence	A specific data analysis or observation that relates to a *type of evidence*. For example, the *type of evidence* 'stressor-response relationships from laboratory studies' may include a chronic value for fathead minnows and an acute *species sensitivity distribution* for freshwater fish as pieces of evidence.
Point Source	Source of pollution that discharge at a specific location from pipes, outfalls, and conveyance channels to a surface water. Examples of point source discharges include municipal wastewater treatment plants, municipal stormwater systems, industrial waste treatment facilities, and construction sites that clear more than 5 acres of land.
Plausibility	The degree to which a cause-effect relationship would be expected, given known facts.
Pollutant	Any substance introduced into the environment that adversely affects a resource.
Probable cause	The *cause* that is most likely to be the true cause of an *effect*.
Proximate cause	The *cause* that induces the *effect* through direct exposure. Compare to an *indirect cause*.
Proximate stressor	The *stressor* that directly induces the biological *effect* of concern. This is equivalent to *candidate cause*, but emphasizes the negative consequences of exposure.
Reasonable explanation	The final consideration in a *strength-of-evidence analysis*. If the results of a strength-of-evidence analysis are not consistent, a mechanistic, conceptual, or mathematical model reasonably may explain the apparent inconsistencies. This concept is called coherence in the *Stressor Identification* guidance document.
Reference	1. A *reference site* or set of reference sites. 2. An environmental attribute of a reference site or a set of reference sites; for example, dissolved oxygen concentrations at a reference site represent reference concentrations.
Reference site	A location or waterbody selected for comparison with the impaired location or waterbody being assessed. The type of sites selected and the type of comparative measures used will vary with the purpose of the comparisons. References that lack a *source*, *stressor*, or *impairment* are termed negative or clean references; references that have well-defined and elevated levels of a *stressor* or well-characterized *sources* or *impairments* are referred to as positive or dirty references.
Refutation	The logical process of demonstrating the impossibility of a *candidate cause*, thus allowing it to be eliminated from further consideration.
Regional reference	A set of sites within a region that represent the best conditions of some environmental characteristic (e.g., a biological index or a pollutant concentration).
Replicate	1. One of a set of independent systems that have been randomly assigned a single treatment. 2. The process of generating a set of such systems.
Response	The biological result of an *exposure*. This term is synonymous with *effect*, but emphasizes the *receptor* that responds (e.g., the response of trout) rather than the *agent* that acts upon it (e.g., the effect of cadmium).
Scoring	Categorization of the results of analyses based on *types of evidence* for a particular *candidate cause*, based on text descriptors and +, -, and 0 symbols.
Shredders	Invertebrates that feed by shredding large pieces of plant matter.

Term	Definition
Simulation model	Mathematical representation of the entities and processes in a system.
Site	A specific location or body of water (e.g., a *stream reach*, a pond, an embayment of a lake, or an area of an estuary).
Site media	Water, sediment, fish tissue, or other materials collected from an impaired *site* or *reference sites* for testing or analysis.
Source	An origination point, area, or entity that releases or emits an *agent* that may be an *indirect cause* or a *proximate cause*.
Spatial/temporal co-occurrence	A *type of evidence* that involves observation of two entities or conditions at the same place or time; it is sometimes shortened to *co-occurrence*.
Specificity	The degree to which an *effect* is known to result from one or very few possible *causes*, or a cause is known to have a distinct effect. Specificity is a *causal consideration* in the *Stressor Identification* guidance. In CADDIS, the concept is incorporated into the *Symptoms* type of evidence.
Stakeholders	People or organizations with an interest in the outcome of an assessment, including *causal analyses* for *bioassessments*.
Stream reach	A segment of a stream delimited in some way (e.g., by occurrence of tributaries or effluents).
Stressor	Any physical, chemical or biological entity that can induce an adverse *effect*. A change in the level of a stressor may be the *proximate cause* of the biological effect under investigation, or may be one event of several required to produce the effect, or may not contribute to causation. *Stressor Identification* focuses on stressors that can induce biological effects. Also see *agent*.
Stressor Identification (SI)	A methodology for determining the most *probable cause* of an observed biological *impairment*, using *elimination*, *diagnosis*, and *strength-of-evidence analysis*. The *CADDIS* website is based on the Stressor Identification process, which is described in a U.S. EPA guidance document.
Stressor-response	1. The relationship between the intensity, frequency, or duration of *exposure* to a *stressor* and the intensity or frequency of a biological *response*. 2. A model of that relationship. Equivalent to *exposure-response* and *concentration-response*.
Structural equation modeling	A family of multivariate statistical methods that use covariance analysis to estimate parameters associated with a series of structural equations that express the hypothetical relationships among several variables that can be either directly observed or manifest variables or unobserved hypothetical or latent variables. It is similar to multiple regression, but uses assumptions concerning the causal network to structure the relationships into one or more equations. It is, in effect, a means of quantifying the links in a conceptual model.
Symptom	A property of affected organisms, populations, communities, or ecosystems that is indicative of a specific *cause* or a very few causes.
Symptomatology	A set of signs indicating the action of a specific *candidate cause* on organisms.
Synergistic	The combined effect of two or more items that is greater than the sum of each one.
Taxa richness	Number of different taxa at a site or in a sample.
Temporal sequence	A *type of evidence* based on the relationship between the time of occurrence of a *candidate cause* and the *effect* of concern. This type of evidence is called temporality in the *Stressor Identification* guidance document.
Tolerance	Measure of degree to which a particular taxon can persist in anthropogenically-disturbed systems. We expect to find highly tolerant taxa at severely degraded sites.
Total Maximum Daily Load (TMDL)	The total allowable pollutant load to a receiving waterbody, such that any additional loading will produce a violation of water quality standards.
Toxicant	A chemical with known toxic properties.
Toxicity identification and evaluation (TIE)	A process that identifies the toxic components of an effluent or ambient medium by chemically manipulating the effluent or medium and testing the resulting material.
Type of evidence	A category of relationships that provides a logically distinct way to support, weaken, or refute the case for a *candidate cause*. A type of evidence may contain multiple *lines of*

Term	Definition
	evidence. It is synonymous with *causal consideration* in the *Stressor Identification* guidance document.
Ultimate cause	The action or policy that is responsible for creating or sustaining a *source*.
Uncertainty	Lack of knowledge concerning an event, state, model, or parameter. Uncertainty, unlike *variability*, may be reduced by research or observation.
Variability	Differences among entities or states of an entity attributable to heterogeneity. Variability is an inherent property of nature and may not be reduced by measurement.
Verified predictions	A *type of evidence* in which predictions about conditions in the receiving system, based on knowledge of the mode of action of the *candidate cause*, are confirmed by observation or measurement; in the *Stressor Identification* guidance document, this is referred to as *predictive performance*.
Watershed	An area of land from which any released or deposited water flows into the same waterbody. Equivalent to catchment.
303 (d) List	Federal Clean Water Act 303(d) list of impaired waters for Washington State.

Acronyms and Abbreviations

BACI	Before-after-control-impact
BIBI	Benthic Index of Biotic Integrity
BOD	Biochemical oxygen demand
CADDIS	Casual Analysis/Diagnostic Decision Information System
CWA	Clean Water Act
DELT	Deformities, erosion, lesions, and tumors
DO	Dissolved oxygen
Ecology	Washington State Department of Ecology
ECOTOX	ECOTOXicology database
EPA	U.S. Environmental Protection Agency
EPT	Ephemeroptera, Plecoptera, and Trichoptera
GIS	Geographic Information System
GPS	Global Positioning System
mg/l	Milligrams per liter
MMI	Multi-metric indices
NA	Not applicable
NHD	National Hydrography Dataset
NPDES	National Pollutant Discharge Elimination System
NTU	Nephelometric turbidity units
O/E	Observed/Expected
RIVPACS	River Invertebrate Prediction and Classification System
SI	Stressor Identification
TMDL	(See Glossary above)

USGS	U.S. Geological Survey
WQIP	Water Quality Implementation Plan
WRIA	Water Resource Inventory Area