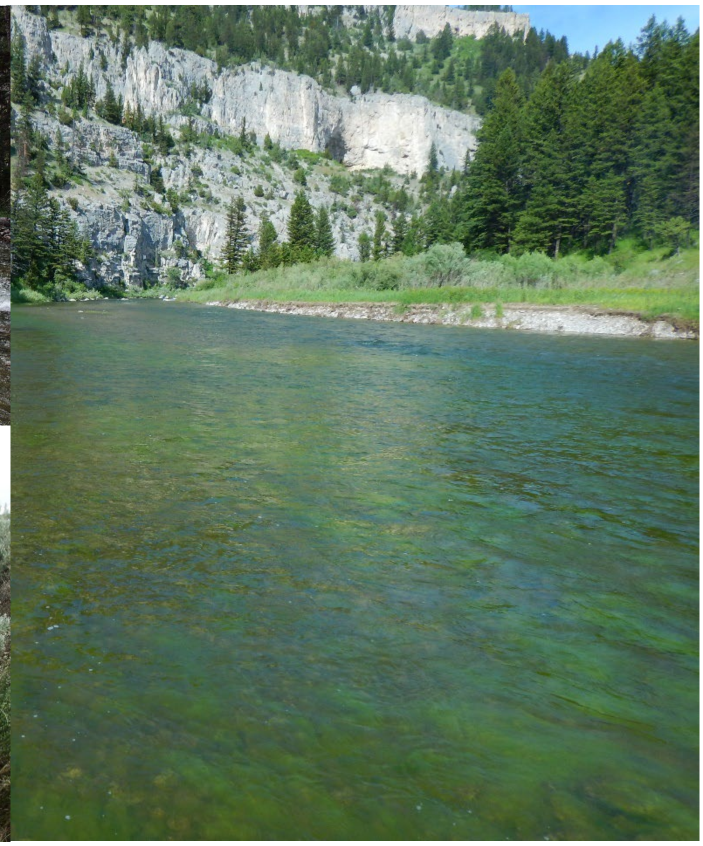
Eutrophication Assessment Method for Wadeable Streams and Medium Rivers



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Prepared by:
Montana Department of Environmental Quality
Water Quality Division
Water Quality Planning Bureau
Monitoring and Assessment Section

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ACRONYMS

AFDW	Ash Free Dry Weight
ARM	Administrative Rules of Montana
AU	Assessment Unit
CFR	Code of Federal Regulations
chl-*a*	chlorophyll-*a*
CWA	Clean Water Act
CWAIC	Clean Water Act Information Center
DEQ	Montana Department of Environmental Quality
DO	dissolved oxygen
DQA	data quality assessment
EPA	U.S. Environmental Protection Agency
ft	feet
ft/sec	feet per second
ft^2/sec	square feet per second
ft^3/sec	cubic feet per second
g/m^2	gram per meter squared
MCA	Montana Code Annotated
mg/L	milligram per liter
mg/m^2	milligram per meter squared
N	nitrogen
O/E	observed/expected
P	phosphorus
QA	quality assurance
QAPP	quality assurance project plan
QC	quality control
SAP	sampling and analysis plan
SOP	standard operating procedure
s.u.	standard units
TMDL	total maximum daily load
TN	total nitrogen
TP	total phosphorus
v3	version 3
WARD	Water Quality Assessment and Reporting Documentation
WQPB	Water Quality Planning Bureau

1.0 Introduction

This document details the Montana Department of Environmental Quality's (DEQ) eutrophication assessment method for wadeable streams and medium-sized (medium) rivers. This method will be used to assess eutrophication impacts on aquatic life and recreation beneficial uses for all water-use classifications applicable to wadeable streams and medium rivers. Beneficial uses are goals and expectations specified in water quality standards for how surface waters should be able to be used. DEQ determines whether waterbodies are supporting designated beneficial uses by conducting assessments of relevant water quality causal and response variables (Makarowski 2020). The DEQ document "Beneficial Use Assessment for Montana's Surface Waters" (Makarowski 2020) describes the overall process for conducting a beneficial use assessment for a waterbody.

1.1 Applicability

This assessment method only applies to wadeable streams and medium rivers; it does not apply to lakes and reservoirs, nor does it apply to large rivers. Definitions for wadeable stream, medium river, and large river are provided below. Large river eutrophication assessments are addressed in DEQ's Large River Eutrophication Assessment Method (2026). This assessment method is not a state rule or regulation. Assessors may use best professional judgement to deviate from this assessment method if necessary. Certain sections of this assessment method are better suited to the use of best professional judgement than others, therefore for such sections this document re-emphasizes the importance of best professional judgement.

Wadeable Stream means a perennial or intermittent stream in which most of the wetted channel is safely wadeable by a person during baseflow conditions.

Medium River means a perennial waterbody in which much of the wetted channel is unwadeable by a person during baseflow conditions.

Large River means a perennial waterbody which has, during summer and fall baseflow (August 1 to October 31 each year), a wadeability index (product of river depth [in feet] and mean velocity [in ft/sec]) of 7.24 ft^2 /sec or greater, a depth of 3.15 ft or greater, or a baseflow annual discharge of 1,500 ft^3 /sec or greater (Flynn and Suplee 2010).

State waters are classified in accordance with their present and future beneficial uses per the Montana Water Quality Act (75-5-301(1), Montana Code Annotated [MCA]). Waterbodies classified as A, B, C, and I are to be maintained suitable for growth and propagation of fishes and associated aquatic life, waterfowl, and furbearers; and be maintained suitable for bathing, swimming, and recreation (Administrative Rules of Montana [ARM] 17.30.621 through 629). This assessment method provides guidance on completing use support decisions for aquatic life and recreation beneficial uses pertaining to eutrophication.

1.2 Background Information

Eutrophication, which is ultimately the result of excess nutrients (nitrogen and phosphorus), causes a variety of water quality problems in flowing waters such as excess algal growth, low dissolved oxygen (DO), altered aquatic communities, and other undesirable water quality changes that impair beneficial uses (**Figure 1**). Filamentous algae, such as *Cladophora spp.,* benefit from excess nutrients and other environmental factors, resulting in growth to undesirable levels that impact recreation and aquatic life (Dodds 2006; Suplee et al. 2009; Suplee et al. 2016; Suplee 2023; Schulte and Craine 2023). Environmental factors influencing sensitivity to algae growth in rivers and streams include light availability, temperature, stream substrate type, and other factors. Altered diel DO and pH fluctuations are a common effect of increased algae growth (Walling and Webb 1992; Suplee 2023). Increased fluctuations in DO are associated with impacts on macroinvertebrate communities (Heiskary et al. 2013; Suplee 2023). These effects are disruptive to both humans and aquatic inhabitants.

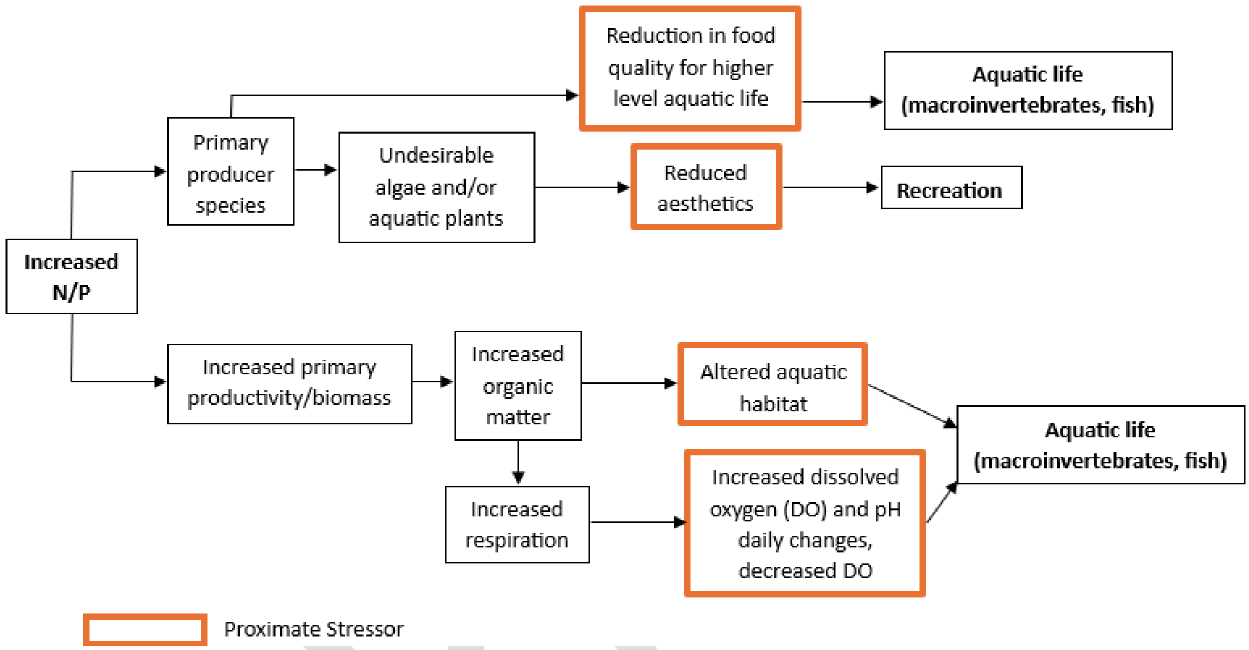


Figure 1. Relationships between Increased Nutrient Concentrations and River and Stream Beneficial Uses. This conceptual model was modified from the one developed in the Stream Nutrient Enrichment Workshop (EPA 2014) and that of Heiskary and Bouchard (2015).

This assessment method will review the following eutrophication-related stressors which can be attributed to human influences:

- undesirable algae growth,
- increases in daily DO changes (i.e., oscillations),
- negative impacts to macroinvertebrates communities and,
- changes in pH concentrations.

This assessment method evaluates the following uses which are the most sensitive to eutrophication:

- Recreation
- Aquatic life

1.3 Common Sources, Pathways, and Factors Influencing Eutrophication

Nutrients are a natural component of stream biological processes but can become over-enriched to levels at which undesirable algae conditions occur. Common sources of excess nitrogen and phosphorus in streams and medium rivers include municipal wastewater treatment plants, septic system discharges, runoff from fertilized fields used for crop production, soil erosion associated with crop production, blasting materials used in mining, and livestock production including both rangeland and animal feeding operations. For information on natural background concentrations of total nitrogen (TN) and total phosphorus (TP) in streams and medium rivers around Montana, see Suplee and Watson (2013).

2.0 Water Quality Standards

This assessment method implements Montana's narrative water quality standard at ARM 17.30.637(1)(e), which prohibits substances attributable to municipal, industrial, agricultural, or other discharges that create conditions producing undesirable aquatic life. Eutrophication, driven by excess nitrogen and phosphorus, is the primary mechanism by which such conditions arise in wadeable streams and medium rivers. As outlined in **Section 1.2**, eutrophication impacts aquatic life and recreation uses. This assessment method provides a consistent way of interpreting the narrative standard ARM 17.30.637(1)(e) by addressing applicable harm thresholds for response variables and then providing information about influencing factors, such as nutrients, to other DEQ programs.

As noted in **Section 1.2**, eutrophication affects pH, which is regulated under different water quality standards which are specific for each water use class (e.g., ARM 17.30.623(2)(c) for B-1 waters). The pH standard is analyzed separately under the pH Assessment Method for Streams and Rivers (DEQ 2025). The outcome from those decisions can be used to support eutrophication decisions made herein.

3.0 Monitoring and Data Quality Considerations for Assessment

Waterbody condition must be evaluated based on all existing and readily available data and information (75-5-702, MCA; 40 Code of Federal Regulations [CFR] § 130.7(b)(5)). This section describes considerations for developing monitoring designs and assessing data quality when performing assessments.

3.1 Monitoring Methods and Protocols

Montana DEQ’s Water Quality Planning Bureau (WQPB) has developed standard operating procedures (SOPs) and guidance manuals for collecting water chemistry, algae, macroinvertebrate, pH, and dissolved oxygen data. Guidance for water chemistry and benthic (bottom-attached) algae sample collection and handling are provided in the Chemistry SOP (Makarowski 2019) and Sample Collection and Laboratory Analysis of Chlorophyll-a SOP (DEQ 2026). Guidance for macroinvertebrate collection, handling, taxonomic identification, and analysis are provided in the Benthic Macroinvertebrate Communities SOP (DEQ 2012).

Guidance for collecting dissolved oxygen measurements is provided in the Instantaneous Water Quality Field Meter SOP (McWilliams and Bushnell 2025), Multiparameter Water Quality Sonde SOP (Milke 2025), and Small Water Quality Dataloggers SOP (McWilliams and Nixon 2020). Discrete dissolved oxygen measurements collected with instantaneous field meters must occur at dawn (4:00 a.m. to sunrise) and late afternoon to early evening to represent the lowest and highest daily dissolved oxygen conditions. Continuous dissolved oxygen measurements collected with deployed field meters are recommended to log measurements at least every 15 minutes.

3.2 Parameters and Sampling Timeframes Considered for Assessment

Water quality parameters and recommended sampling timeframes provided in **Table 1** will be considered when making standards attainment decisions. Geographic regions (ecoregions) across the state have variations in aquatic life communities (example: cold vs. warm water fishes) and annual runoff timeframes (Suplee et al. 2007). Montana streams and rivers are generally most vulnerable to excess nutrient impacts during the summer and early fall baseflow months (typically referred to as the growing season). Therefore, causal variable (nutrients) data collection should normally correspond in time with the response variable sampling timeframes in **Table 1**. Project sampling and analysis plans may call for nutrient sample collection outside of the growing season to characterize concentrations and loads reaching a waterbody when nutrient uptake by algae and aquatic plants is very low to non-existent. Consideration may be given to year-to-year variation in spring or early summer runoff timing and the onset of cold fall weather; approximately a two-week adjustment to the **Table 1** timeframe endpoints is acceptable.

Table 1. Response and causal variables and sampling timeframes considered during eutrophication assessment.

<table>
<tr><th>Parameter</th><th>Unit</th><th colspan="2">Sampling Timeframe</th></tr>
<tr><td colspan="4">Mountain and Transitional Ecoregions Response Variables</td></tr>
<tr><td colspan="4">Primary Response Variables</td></tr>
<tr><td>Benthic Chlorophyll-a</td><td>mg/m^2</td><td colspan="2" rowspan="4">July 1st - September 30th</td></tr>
<tr><td>Benthic Ash-Free Dry Weight</td><td>g/m^2</td></tr>
<tr><td>Filamentous Algae Percent Cover</td><td>%</td></tr>
<tr><td>Macroinvertebrate Community</td><td>Count</td></tr>
<tr><td colspan="4">Secondary Response Variables</td></tr>
<tr><td>pH</td><td>s.u.</td><td colspan="2" rowspan="2">July 1st - September 30th</td></tr>
<tr><td>Daily Dissolved Oxygen Delta</td><td>mg/L</td></tr>
<tr><td colspan="4">Plains Ecoregions Response Variables</td></tr>
<tr><td colspan="4">Primary Response Variables</td></tr>
<tr><td>Daily Dissolved Oxygen Delta</td><td>mg/L</td><td colspan="2">June 21st – September 30th</td></tr>
<tr><td>pH</td><td>s.u.</td><td colspan="2">June 21st – September 30th</td></tr>
<tr><td colspan="4">Optional Primary Response Variables</td></tr>
<tr><td>Benthic Chlorophyll-a[1]</td><td>mg/m^2</td><td colspan="2" rowspan="2">June 21st – September 30th</td></tr>
<tr><td>Benthic Ash-Free Dry Weight[1]</td><td>g/m^2</td></tr>
<tr><td colspan="4">Causal Variables for all Regions</td></tr>
<tr><td rowspan="2">Total Nitrogen and Total Phosphorus</td><td rowspan="2">mg/L</td><td>Mountains and Transitional Regions</td><td>July 1st - September 30th</td></tr>
<tr><td>Plains Regions</td><td>June 16th - September 30th</td></tr>
<tr><td>Nitrite - Nitrate as Nitrogen</td><td>mg/L</td><td>All Regions</td><td>Year Round</td></tr>
</table>

[1] In plains ecoregions, benthic chlorophyll-*a* and ash free dry weight may be used if a waterbody's dominant substrate is gravel or cobble and water clarity conditions allow for benthic algal growth.

3.3 Minimum Data Requirements and Temporal Independence

Table 2 below provides ambient monitoring minimum data requirements per response and causal variable, as well as temporal sampling independence recommendations. Discussion and analyses related to temporal independence are provided in **Appendix A**. To complete a standards attainment decision, DEQ strongly recommends meeting individual minimum data requirements for at least two primary response variables (**Table 1 and 2)**. However, in some circumstances datasets with only one primary response variable may be used to complete standards attainment decisions.

Note that pH data is reviewed separately based on minimal data requirements within the pH assessment method (DEQ 2025). Temporal independence does not apply to continuous dissolved oxygen datasets.

Table 2. Minimum data requirements for response and causal variables with associated temporal sampling independence recommendations.

Parameter	Minimum Data Requirements for Standard Attainment Decisions	Minimum Data Requirements for Delisting Decisions	Temporal Independence Recommendations
Response Variables			
Benthic Chlorophyll-*a*	Three individual samples[1]	Six individual samples[2]	14 days apart
Benthic Ash-Free Dry Weight	Three individual samples[1]	Six individual samples[2]	14 days apart
Filamentous Algae Percent Cover	Three individual samples[1]	Six individual samples[2]	14 days apart
Macroinvertebrate Community	Two individual samples[1]	Four individual samples[2]	About one year apart during summer baseflow conditions
Daily Dissolved Oxygen Delta	At least 10 continuous daily DO delta measurements during one growing season	Minimum of two growing seasons, each with at least 10 continuous daily DO delta measurements[3]	Not applicable
Causal Variables[4]			
Total Nitrogen	Six individual samples[1]	Six individual samples[2]	14 days apart
Total Phosphorus	Six individual samples[1]	Six individual samples[2]	14 days apart
Nitrate - Nitrite	Six individual samples[1]	Six individual samples[2]	14 days apart

[1] Samples must be collected during at least two separate calendar years following sampling timeframes outlined in **Table 1**.
[2] Samples must be collected during at least three separate calendar years following sampling timeframes outlined in **Table 1**.
[3] DO delta measurements should be collected during the hottest time of the growing season.
[4] It is recommended to meet minimum data requirements for causal variables, but it is not required to complete assessment decisions.

3.4 Monitoring Locations and Spatial Independence

Guidance for selecting sampling locations is provided in this section. The topic of spatial independence of data is also covered. See also the discussion and analyses related to spatial independence in **Appendix A**.

3.4.1 Assessment Unit Selection

Beneficial use assessment and impairment decisions are made for assessment units (AU) (i.e., waterbodies or waterbody segments). For eutrophication assessment, DEQ may prioritize monitoring of waters that have been previously identified as impaired due to eutrophication-related causes, waters at higher risk of eutrophication due to human activities, waters in areas where nutrient loads have been reduced, high use recreational areas, or other factors. DEQ may also receive requests to assess waterbodies.

3.4.2 Assessment Units and Reaches

During assessment, data may be aggregated and analyzed for the entire assessment unit or for an assessment reach. An assessment reach is a sub-segmented portion of an assessment unit. If an assessment unit exhibits one or more significant longitudinal shifts in type and intensity of potential nutrient sources, land use, land cover, surface water slope, or other characteristics such that clear breaks could be made to designate homogenous reaches, then breaking an assessment unit into homogenous assessment reaches may be justified. For example, if a relatively unimpacted upstream reach can be isolated and its condition is likely to be substantially different from other downstream parts of the assessment unit, the assessment unit may be split into two reaches for assessment purposes. The assessment steps should be performed separately for each assessment reach. The following general principles and guidance apply:

- If two reaches in an assessment unit are each assessed but only one indicates impairment, the entire assessment unit receives the impairment determination.
- Each reach has the same general data requirements (e.g., dataset minimums) as the parent assessment unit would have had if it hadn't been divided.
- It is better to lump than split reaches to avoid excessive consequential administrative and sampling requirements that result.
- An assessor should, to the best of their ability, decide whether to split an assessment unit into two or more assessment reaches before data collection; this will help ensure that reach breaks are based on considerations of land use and sources.
    - However, if collected data indicate that splitting into reaches is appropriate, proceed with additional data collection if necessary to adequately populate all data needs for both reaches.

3.4.3 Total Number of Sites

Impairment determination is made using data pooled for the entire assessment unit or assessment reach per **Section 3.4.2**, not individual sites. Accordingly, minimum data requirements are set for assessment units, not for individual sites. Best professional judgement is used to determine how many sites are needed to adequately represent the range of potential human sources influencing the assessment unit. It is best to incorporate data collected at multiple sites to better capture variability throughout the assessment unit.

Also, as resources allow, it is preferable to collect multiple samples from each monitoring site selected as long as spatial requirements are met (see **Section 3.4.5**). This enables a multifaceted approach to data analysis; for example, in addition to pooling data from the entire assessment unit to make impairment determinations, an assessor may also strive for enough data to analyze individual sites to perform a thorough source assessment to assist in determining probable sources. Increased monitoring efforts will also provide better information for total maximum daily load (TMDL) development. Assessment decisions can be based on data collected from a single sampling location if that single sampling location can reasonably be considered representative of the assessment unit/reach or if a site represents an area where sources are most likely to impact uses. The assessor will determine if spatial representation of data is sufficient to justify moving forward on a eutrophication assessment.

3.4.4 Site Selection

Data used to make beneficial use assessment decisions for an assessment unit must be collected within the assessment unit. Generally, sampling should target sites that are most likely influenced by sources of concern rather than random sampling designs intended to represent all potential impairment and non-impairment conditions throughout the assessment unit or reach.

Sampling locations that may be prioritized for monitoring include:

- Up- and downstream of point sources
- Up- and downstream of known or likely nonpoint sources
- Up- and downstream of incoming tributaries that may, themselves, have human-caused nutrient sources

Other site locations that may be useful for source assessment purposes include sites that represent natural background conditions (e.g., upstream from human sources), sites that bracket tributary confluences or source areas, and sites on waters that are hydrologically connected to the assessment unit (e.g., ditches, point source discharges, wetlands, reservoirs).

3.4.5 Spatial Independence

The following guidance for generally achieving spatial independence aligns with similar guidance found in other DEQ assessment methods, and is consistent with discussion and analyses presented in **Appendix A**:

- Select sites that are at least one stream mile apart unless there is a flowing tributary that confluences with the segment or a discrete source is located between the two sites.

- Consider land use and landform changes to help identify potential sources of excess nutrients as well as sites representative of natural background conditions.

3.5 Data Currency

Data collected within the past ten years can be considered current and may be used in making assessment decisions (Makarowski 2020). If during this period, significant changes in sources have been documented, the assessor may use best professional judgment when determining which data are appropriate to include in the assessment. The assessor should document in Water Quality Assessment and Reporting Documentation (WARD) the specific changes, identify data currency alternatives, and determine which years of data are appropriate to include in the assessment process.

4.0 Data Quality

Established policies and procedures of DEQ's Water Quality Division for quality assurance and quality control, beneficial use assessment, and data management apply to this assessment method. Data quality requirements apply to all data incorporated while making assessment decisions, whether collected internally by DEQ or externally. All readily available data that passes data quality assessment must be considered when performing beneficial use assessments.

Data quality assessment information is provided in Section 6.0 of the Beneficial Use Assessment Method for Montana's Surface Waters (Makarowski 2020).

5.0 Data Analysis to Support Water Quality Standards Attainment Decisions

5.1 Overview of the Assessment Decision Framework

Assessors apply the same process and decision framework whether or not a waterbody was previously listed on Montana's impaired waters list as impaired for total nitrogen (TN), total phosphorus (TP), nitrate-nitrite, excess algal growth, chlorophyll-*a*, or benthic macroinvertebrates, or other related cause names. For each assessment unit or assessment reach an assessor will compile data, evaluate data sufficiency and credibility, and organize the data for analysis (**Section 5.2**). Once data are prepared, an assessor will first compare response variable data to applicable thresholds. If excess algal growth or macroinvertebrate thresholds are exceeded, an assessor will compare causal variable results to applicable ecoregional ranges and thresholds. The process to determine overwhelming evidence of eutrophication is outlined in **Section 5.4**. Beneficial use determinations for an assessment unit will follow the process outlined in Section 7.0 in the Beneficial Use Assessment Method for Montana's Surface Waters (Makarowski 2020).

5.2 Preparing the Data for Assessment

Assessors will follow these steps when preparing data for assessment decisions:

1. Complete a general review of the watershed to determine if human influenced sources of nutrients exist in the watershed (**Figure 2**). Assessors may use aerial imagery, geographic information system layers, or other relevant resources. If human influenced sources of nutrients do not exist in the watershed, consult with the MAS supervisor on next steps.
2. Compile all response and causal parameters available for an assessment unit or reach (**Sections 3.2 and 3.4**).
3. Organize data for each parameter by site and evaluate spatial independence of sites according to **Section 3.4**. Compile each data point from spatially-dependent sites prior to the next step.
4. Organize data for each parameter by year, month, and day for each site. The data within the required sampling timeframe (**Section 3.2**) will be used for the following steps and attainment decision.
5. Perform data quality assessment for each parameter to identify the usable dataset. Follow guidance provided in Section 6.0 of the Beneficial Use Assessment Method for Montana's Surface

Waters (Makarowski 2020). Address non-detects according to **Section 5.3**. Document any data that cannot be used.

6. Determine temporal independence of data for each site and parameter (e.g., samples collected 14 days apart) (**Section 3.3**). If multiple sample results were collected within a 14-day period, calculate the average for each 14-day period.
7. Determine if minimum data requirements are met for each parameter using data from all sites of an assessment unit or reach (**Section 3.3**).
8. If at least one primary response variable meets minimum data requirements, move forward with assessment analysis based on geographic location (**Section 5.5 or 5.6**).
9. Total nitrogen, total phosphorus, and/or nitrate-nitrite data must meet minimum data requirements (**Section 3.3, Table 2**) to complete an analysis of the likely causal nutrient(s). If minimum data requirements are not met, an assessor will not complete an analysis of the likely causal nutrient(s).

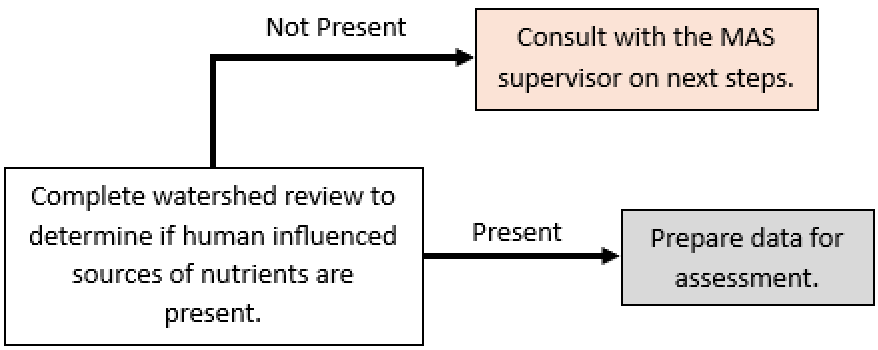


Figure 2. Flow chart outlining preparing data for assessment.

5.3 Handling Non-Detects

Concentrations of causal variables reported below detectable limits are presumed to be present in the waterbody, albeit below the lower reporting limit. Required reporting limits for nutrients should be documented in the Quality Assurance Project Plan (QAPP), Sampling and Analysis Plan (SAP), or similar quality assurance document. For total nitrogen, total phosphorus, and nitrate-nitrite data points reported below the required reporting limit, the non-detect values should be replaced with numeric values which are half the required reporting limit if the non-detect values represent approximately 15% (or less) of the dataset. If more than approximately 15%, consult your supervisor on potential alternative methods for handling the non-detects.

5.4 Harm-to-Beneficial Use Thresholds and Exceedance Rates

Response variable thresholds and exceedance rates for wadeable streams and medium rivers are listed in **Tables 3** and **4**. If DEQ approves use of a model that derives assessment unit- or waterbody-specific thresholds for response or causal variables, assessors may use the approved model-derived thresholds instead of the regionally-based response or causal variable thresholds in **Tables 3** and **4**. Assessors may use alternative allowable exceedance rates for large data sets.

If documentation is not available that demonstrates multiple excessive and extensive algal blooms over a 3-year period, and a large data set (n ≥20) is available, an assessor may use best professional judgement when evaluating the dataset's exceedance rate (≥20) for benthic algae parameters (chl-*a*, AFDW, and filamentous percent cover). To interpret the one-in-three-year exceedance rate for a large benthic algae dataset, the recommendation is to use a binomial distribution test with a 90% confidence level, an allowable exceedance rate is ≤10%, and an effect size of 15%.

5.4.1 Rationale for Aquatic Life Based Response Variable Thresholds and Allowable Exceedance Rates

5.4.1.1 Macroinvertebrate Metrics

Schulte and Craine (2023) and Suplee (2023) provide the basis for macroinvertebrate metric thresholds for mountains, low valleys, and transitional ecoregions in accordance with their water surface slope (**Table 4**). Thresholds were selected at the mid-point of change in the nutrient-biometric logistic relation; this approach is well supported in technical and scientific literature (Schulte and Craine 2024). Beck's Biotic Index (v3), a metric based on macroinvertebrate population structure, had a consistent correlation with total nitrogen and total phosphorus concentrations (Schulte and Craine 2024). Hilsenhoff Biotic Index and Beck's Biotic Index (v3) respond to increases in dissolved oxygen delta. Indices indicate a loss of sensitive species with increases in dissolved oxygen delta (Suplee 2023). Additional macroinvertebrate metrics and thresholds selected from Schulte and Craine (2023) are provided in **Table 5.** When available, an assessor can use observed/expected (O/E) metric results. Assessors will interpret O/E results following guidance in Section 5.6 in the macroinvertebrate SOP (DEQ 2012).

An assessor will select macroinvertebrate metrics based on stream slope. However, an assessor may use best professional judgement when necessary. In addition to water surface ≤1%, other naturally occurring characteristics that qualify a waterbody as being viewed as a low gradient site are:

- The stream substrate is more dominated by small gravels and sand and silt rather than gravels and cobbles;
- Presence of sparse macrophyte (aquatic vascular plant) populations; and
- Glides with minimal water surface disturbance interspersed between more turbulent riffles and runs in the longitudinal geomorphic pattern.

The opposite situation may occur as well. A reach of stream or river could have water surface slope <1% but have substrate dominated by gravels and cobbles, no macrophytes, and have turbulent riffles and few or no glides. It would be more suitable to apply the >1% thresholds in this case. Allowable exceedance rates for macroinvertebrate metrics are established as one allowable year in three years, consistent with EPA recommendations (Stephan et al. 1985) where it is presumed that most aquatic systems can recover from exceedances in about three years.

5.4.1.2 Dissolved Oxygen Delta

Mountain and Transitional Wadeable Streams and Medium Rivers

Suplee (2023) provides the basis for dissolved oxygen delta thresholds for mountain/transitional and plains ecoregions. The dissolved oxygen delta threshold for mountain and transitional waterbodies is based on the relationship between macroinvertebrate metric harm thresholds and corresponding dissolved oxygen deltas.

Allowable exceedance rates reviewed for dissolved oxygen delta were recommended by EPA or used by other states. According to CA (2004), allowable exceedance rates commonly used by states for conventional pollutants and bacteria range from 10% - 25%. To select an allowable exceedance rate for the mountain and transitional ecoregion a waterbody was selected for a case study. A eutrophication rating assessment was completed for each site following guidance outlined in Section 2.6 of Suplee

(2023). The site with a eutrophication rating of two (low eutrophication impacts) had an exceedance rate of 25%. Therefore, an allowable exceedance rate of 25% was selected and considered protective.

Plains Wadeable Streams and Medium Rivers
The plains dissolved oxygen delta threshold is based on the relationship between weekly dissolved oxygen delta and dissolved oxygen minimum standards (specifically, during non-drought conditions) (GLEC 2021). The dissolved oxygen delta threshold applies to wadeable streams and medium rivers with a slope less than one percent. The allowable exceedance rate selected (15%) was informed by the recommendation provided in Suplee 2023 to ensure dissolved oxygen delta is not exceeded too often and supports aquatic life.

5.4.1.3 Algae Thresholds

Suplee and Flynn (2014) provide the basis for a benthic chlorophyll-*a* threshold (125 mg Chl-*a*/m^2) for mountain and transitional ecoregions. In Suplee and Flynn (2014), results from a nutrient dosing study on a stream in the plains (Suplee et al. 2019) were modeled and simulated for similar size and sloped streams for a mountainous waterbody. The results indicated vulnerability to low dissolved oxygen problems caused by algae density exceeding the threshold, which would impact aquatic life. Accordingly, allowable exceedance rates for algae thresholds are established as one allowable year in three years, consistent with EPA recommendations (Stephan et al. 1985) where it is presumed that most aquatic systems can probably recover from exceedances in about three years.

5.4.2 Rationale for Recreation Based Response Variable Thresholds

Algae Thresholds

Multiple studies provide the basis for recreation-based algae thresholds for mountain and transitional ecoregions of Montana. The benthic chlorophyll-*a* thresholds are based on acceptable levels from public opinion surveys in both Montana and Utah (Suplee et al. 2009; Jakus et al. 2017). The ash free dry weight threshold is based on considerations presented in **Appendix B**. Percent filamentous algae cover is based on public opinion work in Utah and West Virginia (WV DEP 2012; Ostermiller et al., 2019) and is consistent with cover percentages associated with the chlorophyll-*a* harm-to-use level documented in Montana's public opinion survey (Suplee et al. 2009). In wadeable streams there is often close alignment between aquatic life and recreational impacts (see Suplee et al. 2019) and, accordingly, allowable exceedance rates for algae thresholds are established as one allowable year in three years, consistent with EPA recommendations (Stephan et al. 1985).

5.4.3 Causal Variables

Total nitrogen and total phosphorus ecoregional ranges and a nitrate-nitrite threshold are provided in **Table 5**. The total nitrogen and total phosphorus ranges represent the range of nutrient concentrations based on dose-response studies and associated with likely harm-to-beneficial uses (aquatic life, recreation) that have been documented in the scientific literature for the ecoregion(s) shown (see Suplee and Watson, 2013; Schulte and Craine 2023). In virtually all cases, natural background ecoregion concentrations are substantially lower than these concentrations (Suplee and Watson 2013). Therefore, a logical way to compare a wadeable stream or medium river's dataset to the ranges is via a measure of the waterbody's central tendency, and DEQ selected the mean for this assessment method. An assessor will calculate a summer growing season mean for total nitrogen and total phosphorus and compare the mean to the applicable ecoregional range (**Section 5.5 and 5.6**).

The nitrate-nitrite concentration threshold is a concentration which, if measured during routine assessment, indicates the stream/river is largely saturated for nitrate and, as a result, detrimental eutrophication impacts may ensue (Suplee 2013). The nitrate-nitrate allowable exceedance rate selected is one exceedance in three years. This lower exceedance rate for nitrate-nitrite is attributed to evaluating a saturated condition, instead of a condition that would likely limit algal growth. Nitrate concentrations that would control algae growth are usually lower than this value.

Table 3. Algae and dissolved oxygen delta thresholds and exceedance rates for wadeable rivers and streams.

Beneficial Use	Ecoregions	Parameter	Threshold	Duration*	Allowable Exceedance Rate
Recreation & Aquatic Life	Mountain and Transitional	Benthic Chlorophyll-*a*	≤ 125 mg/m^2	Single Sample	1 in 3 Years
Recreation & Aquatic Life	Mountain and Transitional	Benthic Ash-Free Dry Weight	≤ 35 g/m^2	Single Sample	1 in 3 Years
Aquatic Life	Plains[1]	Benthic Chlorophyll-*a*	≤ 150 mg/m^2	Single Sample	1 in 3 Years
Aquatic Life	Plains[1]	Benthic Ash-Free Dry Weight	≤ 35 g/m^2	Single Sample	1 in 3 Years
Recreation	Mountain and Transitional	Filamentous Algae Percent Cover	≤ 30%	Single Observation	1 in 3 Years
Aquatic Life	Mountain and Transitional (with a slope less than or equal 1%)	Dissolved Oxygen Delta	≤ 3 mg/L	7-Day Average	25%
Aquatic Life	Plains	Dissolved Oxygen Delta	≤ 6 mg/L	7-Day Average	15%

[1] Threshold and exceedance rate may be applied to a plains wadeable river or stream if the waterbody's dominant substrate is gravel or cobble and water clarity conditions allow for benthic algal growth.

* Duration Definition: A period of time over which a parameter is averaged or evaluated.

Table 4. Macroinvertebrate metric thresholds and exceedance rates for wadeable rivers and streams.

Beneficial Use	Slope Designation	Metric	Threshold*	Duration**	Allowable Exceedance Rate
Aquatic Life	> 1% Slope	Beck's Biotic Index v3	≥ 35.1	Seasonal Average	1 in 3 Years
Aquatic Life	> 1% Slope	Hilsenhoff Biotic Index	≤ 4	Seasonal Average	1 in 3 Years
Aquatic Life	> 1% Slope	Percent of Intolerant Taxa	≥ 42.2	Seasonal Average	1 in 3 Years
Aquatic Life	> 1% Slope	Number of Plecoptera Taxa	≥ 4.8	Seasonal Average	1 in 3 Years
Aquatic Life	> 1% Slope	Number of EPT Taxa	≥ 18.1	Seasonal Average	1 in 3 Years
Aquatic Life	≤ 1% Slope	Beck's Biotic Index v3	≥ 18.7	Seasonal Average	1 in 3 Years
Aquatic Life	≤ 1% Slope	Hilsenhoff Biotic Index	≤ 5	Seasonal Average	1 in 3 Years
Aquatic Life	≤ 1% Slope	Percent of Insect Taxa	≥ 84.2	Seasonal Average	1 in 3 Years
Aquatic Life	≤ 1% Slope	Number of Intolerant EPT Taxa	≥ 10.6	Seasonal Average	1 in 3 Years

*Macroinvertebrate metric scores will either increase or decrease with increasing pollution. Metrics preceded by ≥ indicate a metric where higher values are better (healthier) scores; those preceded by ≤ indicates that lower values are better (healthier) scores.

** Duration Definition: A period of time over which a parameter is averaged or evaluated.

Table 5. Total nitrogen (TN) and total phosphorus (TP) ranges within which harm-to-beneficial uses may ensue.
A nitrate-nitrite threshold for wadeable rivers and streams is also provided in Section 5.4.3.

Region	**Ecoregion** (Level III)	**Ecoregion** (Level IV)	**TP** (mg/L)	**TN** (mg/L)	**Nitrate-Nitrite** (mg/L)
Mountains	Northern Rockies (15)	all	0.02 – 0.04	0.139[a] – 0.750	0.1
Mountains	Canadian Rockies (41)	all	0.02 – 0.06	0.139[a] – 0.750	0.1
Mountains	Idaho Batholith (16)	all	0.02 – 0.06	0.139[a] – 0.750	0.1
Mountains	Middle Rockies (17)	all except 17i	0.02 – 0.06	0.139[a] – 0.980	0.1
Mountains	Middle Rockies (17)	**17i.** Absaroka-Gallatin Volcanic Mountains	0.043 – 0.106[b]	Use values from the lower end of the Middle Rockies ecoregion range	0.1
Transitional	Northwestern Glaciated Plains (42)	**42i.** Sweetgrass Upland **42n.** Milk River Pothole Upland **42q.** Rocky Mountain Front Foothill Potholes **42r.** Foothill Grassland	0.020 – 0.206[c]	0.199 – 0.775[d]	0.1
Transitional	Northwestern Great Plains (43)	**43s.** Non-calcareous Foothill Grassland **43t**. Shields Smith Valleys **43u.** Limy Foothill Grassland **43v.** Pryor-Bighorn Foothills **43o.** Unglaciated Montana High Plains[g]	0.020 – 0.041[e]	0.199 -1.125[f]	0.1
Plains	Northwestern Glaciated Plains (42)	All except those listed above for 42	0.070 – 0.150	0.540 – 1.830	0.1
Plains	Northwestern Great Plains (43) and Wyoming Basin (18)	All except for those listed 42 and 43c	0.070 – 0.150	0.540 – 1.830	0.1
Plains	Northwestern Great Plains (43)	River Breaks (43c)	None Recommended	None Recommended	0.1

[a]Lower end based on macroinvertebrate response (page 18 of Schulte and Craine 2023).
[b]Based on the 10th and 90th percentiles of the natural background concentrations for this level IV ecoregion (Suplee and Watson 2013).
[c]Lower end based on streams' origins in the Canadian Rockies; upper end on 90th percentile of natural background for these ecoregions (Suplee and Watson 2013).

[d]Lower end based on macroinvertebrate response (page 20 of Schulte and Craine 2023); upper end on region-specific Chl-a computation (see page 3-24 of Suplee and Watson 2013).
[e]Lower end based on similarity to Middle Rockies, upper end based on Elk Creek reference site (Suplee and Watson 2013).
[f]Lower end based on macroinvertebrate response (page 20 of Schulte and Craine 2023); upper end on region-specific Chl-a computation (see page 3-37 of Suplee and Watson 2013).
[g]For the Unglaciated High Plains ecoregion (43o), range applies to the polygon located just south of Great Falls, MT.

5.5 Mountain and Transitional Wadeable Stream and Medium River Assessment Process

The eutrophication assessment decision framework outlined in this section applies when determining impairment decisions for eutrophication in mountain or transitional streams and medium rivers (**Figures 3, 4,** and **5**). To complete a standards attainment decision, DEQ strongly recommends meeting individual minimum data requirements for at least two primary response variables. However, in some circumstances datasets with only one primary response variable may be used to complete standards attainment decisions. When completing delisting decisions, minimum data requirements must be met for all primary response variables and the causal variables of total nitrogen, total phosphorus, and nitrate-nitrite.

The following steps should be followed when determining attainment of the narrative standard as it pertains to eutrophication.

Primary Response Variables (Section 3.2)

1. Compare sufficient and credible primary response variable sample results to thresholds provided in **Tables 3** and **4**.
    a. Compare chlorophyll-*a* sample results to the mountain and transitional ecoregional threshold. Document the number of exceedances and the years exceedances occurred.
    b. Compare ash free dry weight sample results to the mountain and transitional ecoregional threshold. Document the number of exceedances and the years exceedances occurred.
    c. Compare filamentous algae percent cover observation to the mountain and transitional threshold. Document the number of exceedances and the years exceedances occurred.
    d. For each site, calculate a site seasonal average for each macroinvertebrate metric. Compare macroinvertebrate seasonal average metrics to slope designation-based thresholds. Document the number of exceedances for each metric type and the years exceedances occurred.
2. Assess each primary response variable independently. If more than one sample within a three-year period exceeds the threshold, the response variable fails. It is recommended that a primary response variable exceeds the threshold during two separate growing seasons to list the waterbody for eutrophication (**Figure 3**).
    a. Assessors may use best professional judgement when assessing macroinvertebrate metrics. For example, if an individual metric does not fail the more than one in three years exceedance rate, then an assessor may look at the exceedance rates for multiple metrics combined.
3. Move to analyze causal variables if chlorophyll-*a* and/or ash free dry weight fail, or if two primary response variables fail (**Figure 5**).
4. Move to assess secondary response variables if filamentous algae percent cover ***or*** macroinvertebrates was the only response variable able to be assessed (**Figure 4**). Move to assess secondary response variables if chlorophyll-*a* and/or ash free dry weight were the only response variables able to be assessed and the analysis resulted in a pass. Assessing secondary response variables assists with standards attainment decisions.

Secondary Response Variables

1. Review and document pH assessment outcomes (if completed) for an assessment unit or reach.

   a. If an assessment unit is listed as impaired due to pH exceedances above *or* above and below the pH standard, the response variable fails.
2. Calculate 7-day rolling averages for continuous dissolved oxygen delta datasets. The analysis steps provided are completed per site.
   a. Format continuous data files following guidance in Appendix A of the Small Water Quality Datalogger SOP (McWilliams and Nixon 2020).
   b. Use the DO_delta_v1_alpha3 Excel macro file tool to calculate dissolved oxygen deltas for each day.
   c. Calculate rolling dissolved oxygen delta 7-day averages.
      i. Seven consecutive dissolved oxygen deltas are required to calculate a 7-day average.
      ii. An assessor may use best professional judgement if there is a one- or two-day gap in a continuous dataset.
   d. Per site, count the number of exceedances greater than the threshold provided in **Table 3**.
   e. Calculate the percentage of exceedances for each site.
   f. If a single site has a greater exceedance percentage than the allowable exceedance rate (**Table 3**), the response variable fails.
3. Calculate 7-day averages for discrete dissolved oxygen delta datasets. The analysis steps provided are completed per site.
   a. For each day, subtract the dissolved oxygen daily maximum from the dissolved oxygen daily minimum to determine the dissolved oxygen delta.
   b. Calculate rolling dissolved oxygen delta 7-day averages.
      i. Seven consecutive dissolved oxygen deltas are required to calculate a 7-day average.
      ii. An assessor may use best professional judgement if there is a one- or two-day gap in a continuous dataset.
   c. Per site, count the number of exceedances greater than the threshold provided in **Table 3**.
   d. Calculate the percentage of exceedances for each site.
   e. If a single site has a greater exceedance percentage than the allowable exceedance rate, then the response variable fails.
4. Follow guidance in **Figure 4** to determine next steps. If the primary response variable findings conflict with the secondary response variable findings, more sampling may be required in order to make an assessment decision.

Causal Variables

The following steps are completed only if the assessment of response variables results in a eutrophication impairment listing (**Figure 5**).

1. Using the complete dataset for an assessment unit or reach, calculate the total nitrogen and total phosphorus growing season means.
   a. Compare the total nitrogen growing season mean to the applicable ecoregional range provided in **Table 5**.
      i. If the growing season mean is <u>within</u> the ecoregional range, nitrogen may be influencing eutrophication, and assessors should evaluate nitrogen on a case-by-case basis in relation to the phosphorus concentration.
      ii. If the growing season mean is <u>above</u> the ecoregional range, nitrogen is likely influencing eutrophication.

   iii. If the growing season mean is below the ecoregional range and excess algal growth is present, nitrogen may be a limiting nutrient.

   b. Compare the total phosphorus growing season mean to the applicable ecoregional range provided in **Table 5**.
      i. If the growing season mean is within the ecoregional range, phosphorus may be influencing eutrophication, and assessors should evaluate phosphorus on a case-by-case basis in relation to the nitrogen concentration.
      ii. If the growing season mean is above the ecoregional range, phosphorus is influencing eutrophication. Consult with MAS supervisor.
      iii. If the growing season mean is below the ecoregional range and excess algal growth is present, phosphorus may be a limiting nutrient.

2. Using the complete dataset for an assessment unit or reach, count the number of exceedances greater than the nitrate-nitrite threshold provided in **Table 5**. If more than one sample within a three-year period exceeds the threshold, nitrate-nitrite may be influencing eutrophication.

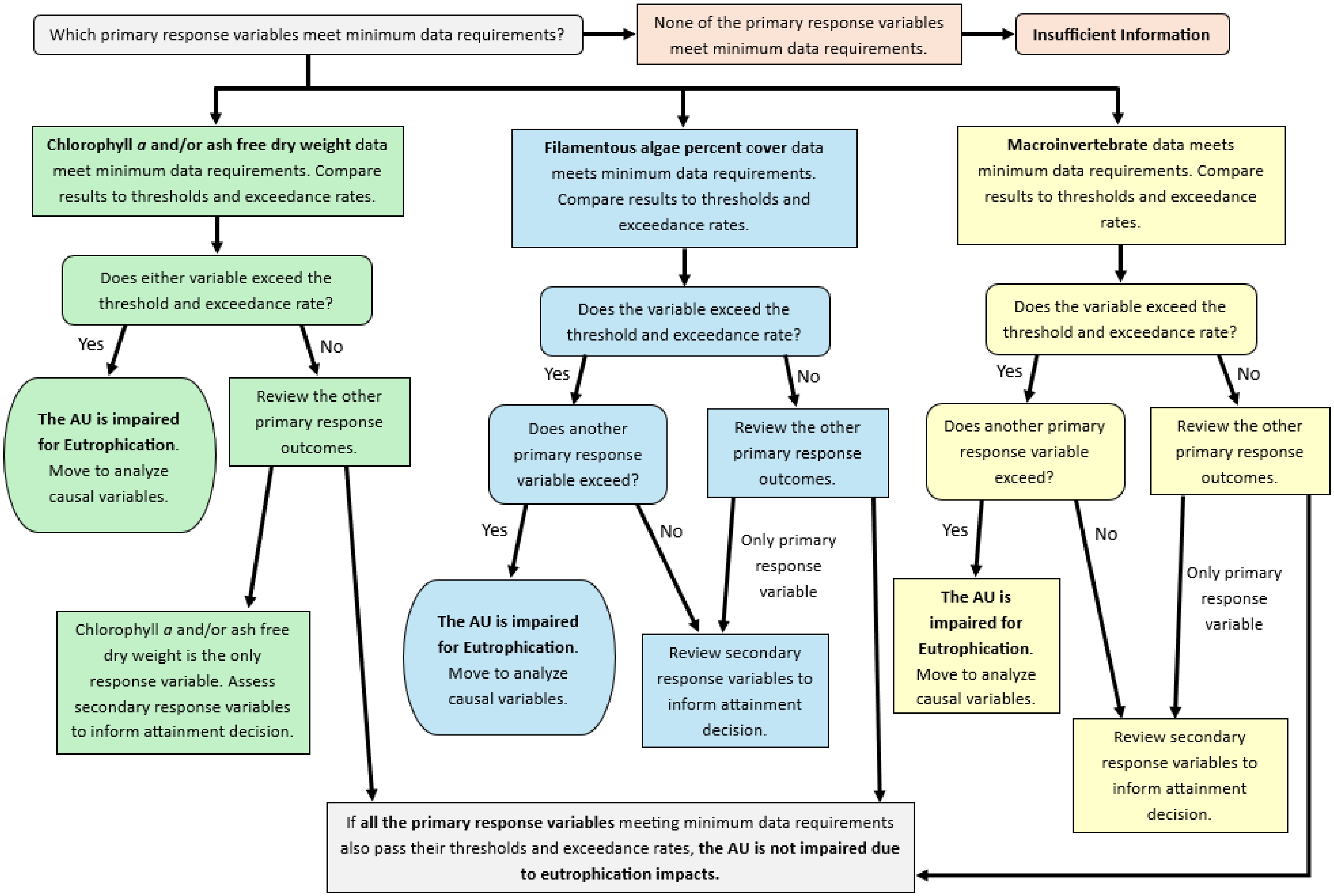


Figure 3. Mountain and transitional primary response variable decision framework for wadeable rivers and streams.

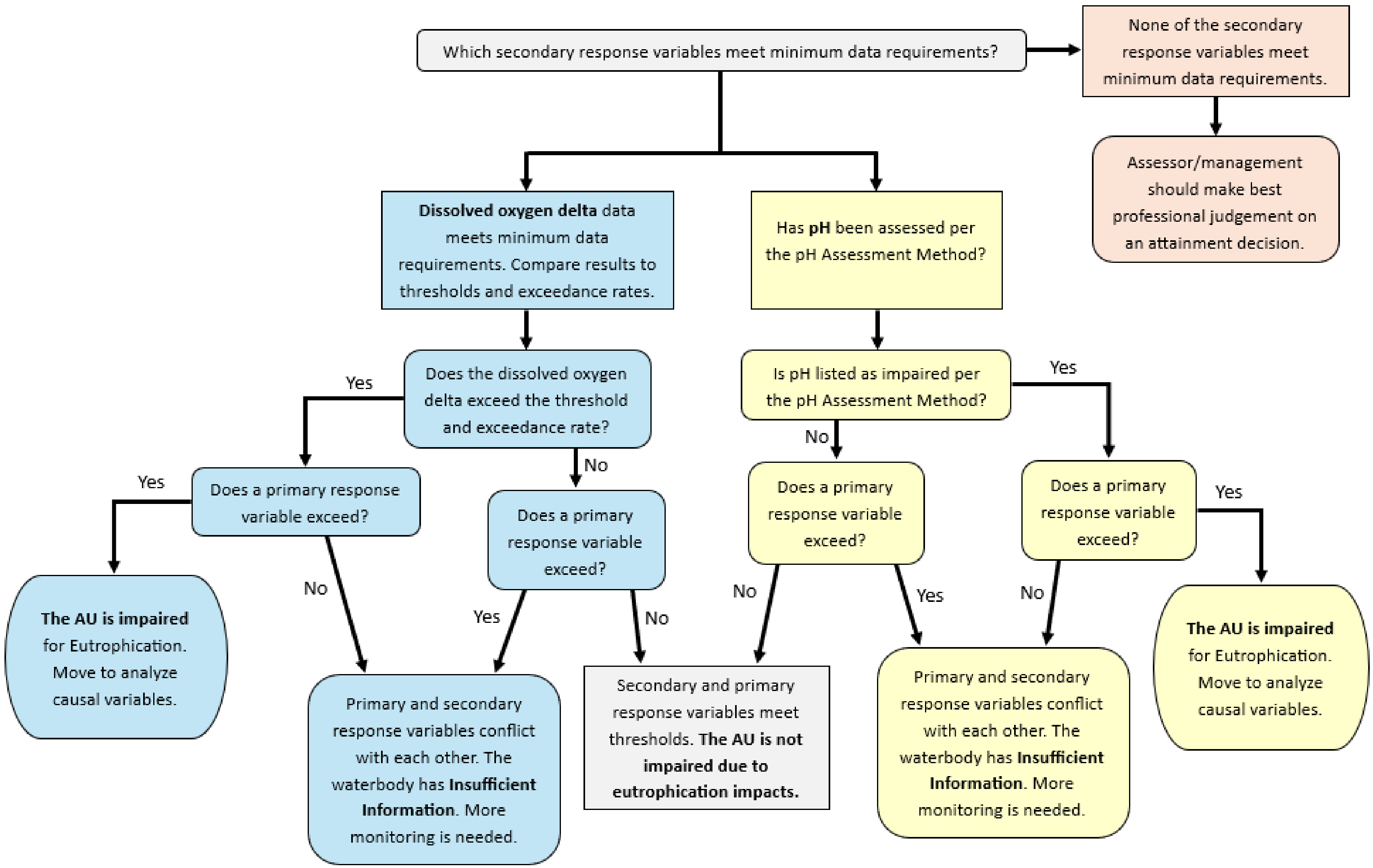
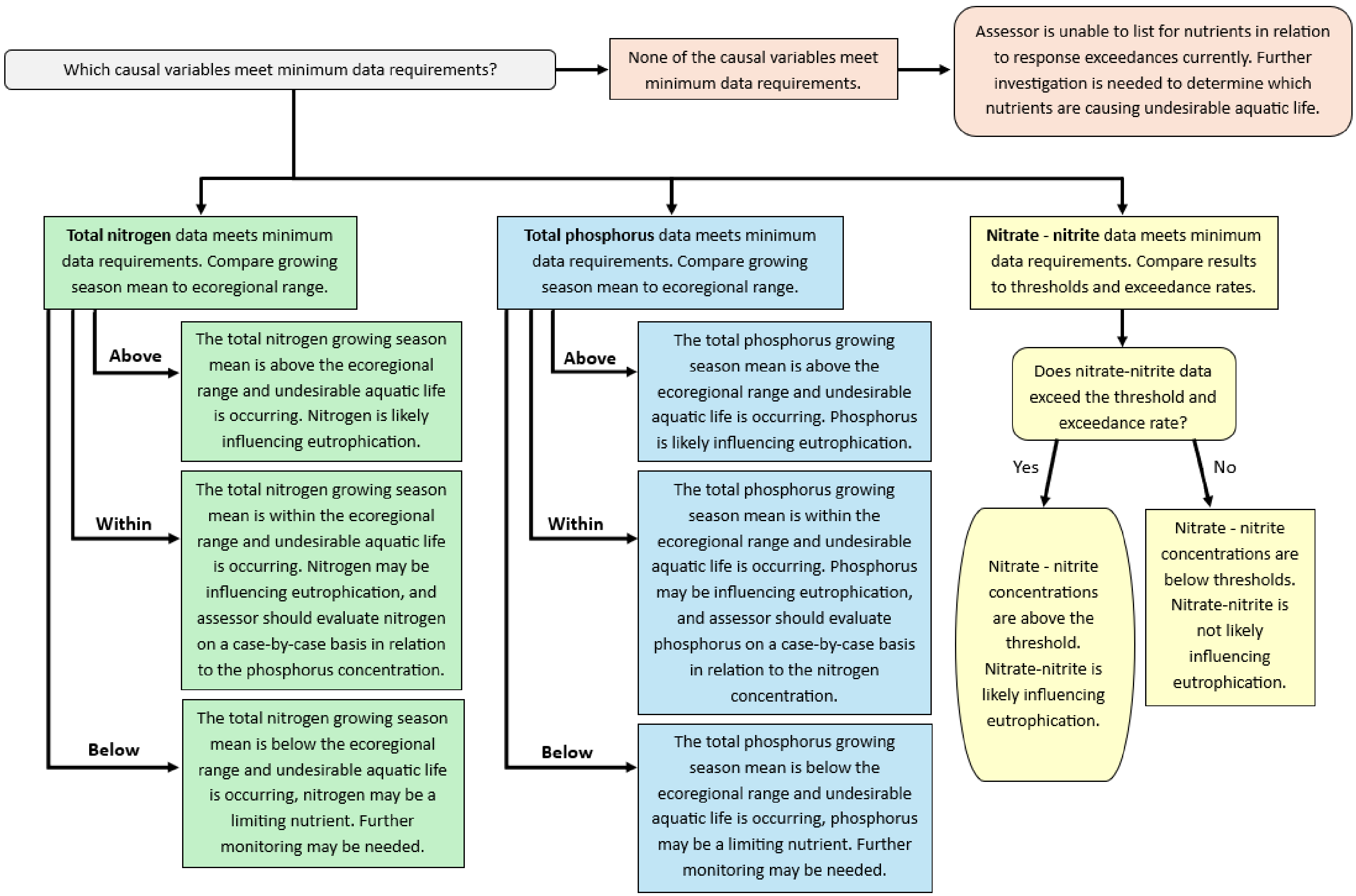


Figure 4. Mountain and transitional secondary response variable decision framework for wadeable rivers and streams.



NOTE: If DEQ approves use of a model that derives assessment unit- or site-specific thresholds for causal variables, assessors may use the approved model-derived thresholds.

Figure 5. Mountain/transitional and plains causal variable decision framework for wadeable rivers and streams.

5.6 Plains Wadeable Stream and Medium River Assessment Process

The eutrophication assessment decision framework outlined in this section applies when determining impairment decisions for identifying eutrophication as a cause of impairment to aquatic life and/or recreation in plains wadeable streams and medium rivers (**Figures 4** and **5**). When completing delisting decisions, minimum data requirements must be met for all primary response variables and causal variables (total nitrogen, total phosphorus, and nitrate-nitrite).

The following steps should be followed when determining attainment of the narrative standard relating to eutrophication.

Primary Response Variables (Section 3.2)

1. Calculate 7-day averages for continuous dissolved oxygen delta datasets. The analysis steps provided are completed per site.
    a. Format continuous data files following guidance in Appendix A of the Small Water Quality Datalogger SOP (McWilliams and Nixon 2020).
    b. Use the DO_delta_v1_alpha3 Excel macro file tool to calculate dissolved oxygen deltas for each day.
    c. Calculate rolling dissolved oxygen delta 7-day averages.
        i. Seven consecutive dissolved oxygen deltas are required to calculate a 7-day average.
        ii. An assessor may use best professional judgement if there is a one- or two-day gap in a continuous dataset.
    d. If a dataset was collected during drought conditions, run the drought correction tool according to Section 3.1 and Appendix C from the Addendum: Daily Patterns of Dissolved Oxygen Change in Montana Waters (Suplee 2026) prior to comparing results to the threshold and exceedance rate.
    e. Per site, count the number of exceedances greater than the threshold provided in **Table 3**.
    f. Calculate the percentage of exceedances for each site.
    g. If a single site has a greater exceedance percentage than the allowable exceedance rate, the response variable fails.
2. Calculate 7-day averages for discrete dissolved oxygen delta datasets. The analysis steps provided are completed per site.
    a. For each day, subtract the dissolved oxygen daily maximum from the dissolved oxygen daily minimum to determine dissolved oxygen delta.
    b. Calculate rolling dissolved oxygen delta 7-day averages.
        i. Seven consecutive dissolved oxygen deltas are required to calculate a 7-day average.
        ii. An assessor may use best professional judgement if there is a one- or two-day gap in a continuous dataset.
    c. If a dataset was collected during drought conditions, run the drought correction tool according to Section 3.1 and Appendix C from the Addendum: Daily Patterns of Dissolved Oxygen Change in Montana Waters (Suplee 2026) prior to comparing results to the threshold and exceedance rate.
    d. Per site, count the number of exceedances greater than the threshold provided in **Table 3**.
    e. Calculate the percentage of exceedances for each site.

   f. If a single site has a greater exceedance percentage than the allowable exceedance rate (**Table 3**), the response variable fails.
3. If dissolved oxygen delta fails, move to assess causal variables **(Figure 4)**.
4. If dissolved oxygen delta passes, follow guidance provided in **Figure 5.**

Optional Primary Response Variables

1. Compare sufficient and credible primary response variable sample results to thresholds provided in **Table 3**.
   a. Compare chlorophyll-*a* sample results to the plains ecoregional threshold. Document the number of exceedances and the years exceedances occurred.
   b. Compare ash free dry weight sample results to the plains ecoregional threshold. Document the number of exceedances and the years exceedances occurred.
2. Assess each response variable independently. If more than one sample within a three-year period exceeds the threshold, the response variable fails. For listing decisions, it is recommended for a primary response variable to have exceedances during two separate growing seasons.
3. Move to assess causal variables if chlorophyll-*a* or ash free dry weight fail **(Figure 4)**.

Causal Variable

The following steps are completed only if the assessment of response variables results in a eutrophication impairment listing.

1. Using the complete dataset for an assessment unit or reach, calculate the total nitrogen and total phosphorus growing season means.
   a. Compare the total nitrogen growing season mean to the applicable ecoregional range provided in **Table 5**.
      i. If the growing season mean is within the ecoregional range, nitrogen may be influencing eutrophication, and assessors should evaluate nitrogen on a case-by-case basis in relation to the phosphorus concentration.
      ii. If the growing season mean is above the ecoregional range, nitrogen is influencing eutrophication.
      iii. If the growing season mean is below the ecoregional range and excess algal growth is present, nitrogen may be a limiting nutrient.
   b. Compare the total phosphorus growing season mean to the applicable ecoregional range provided in **Table 5**.
      i. If the growing season mean is within the ecoregional range, phosphorus may be influencing eutrophication, and assessors should evaluate phosphorus on a case-by-case basis in relation to the nitrogen concentration.
      ii. If the growing season mean is above the ecoregional range, phosphorus is influencing eutrophication. Consult with MAS supervisor.
      iii. If the growing season mean is below the ecoregional range and excess algal growth is present, phosphorus may be a limiting nutrient.
2. Using the complete dataset for an assessment unit or reach, count the number of exceedances greater than the nitrate-nitrite threshold provided in **Table 5**. If more than one sample within a three-year period exceeds the threshold, nitrate-nitrite may be influencing eutrophication.

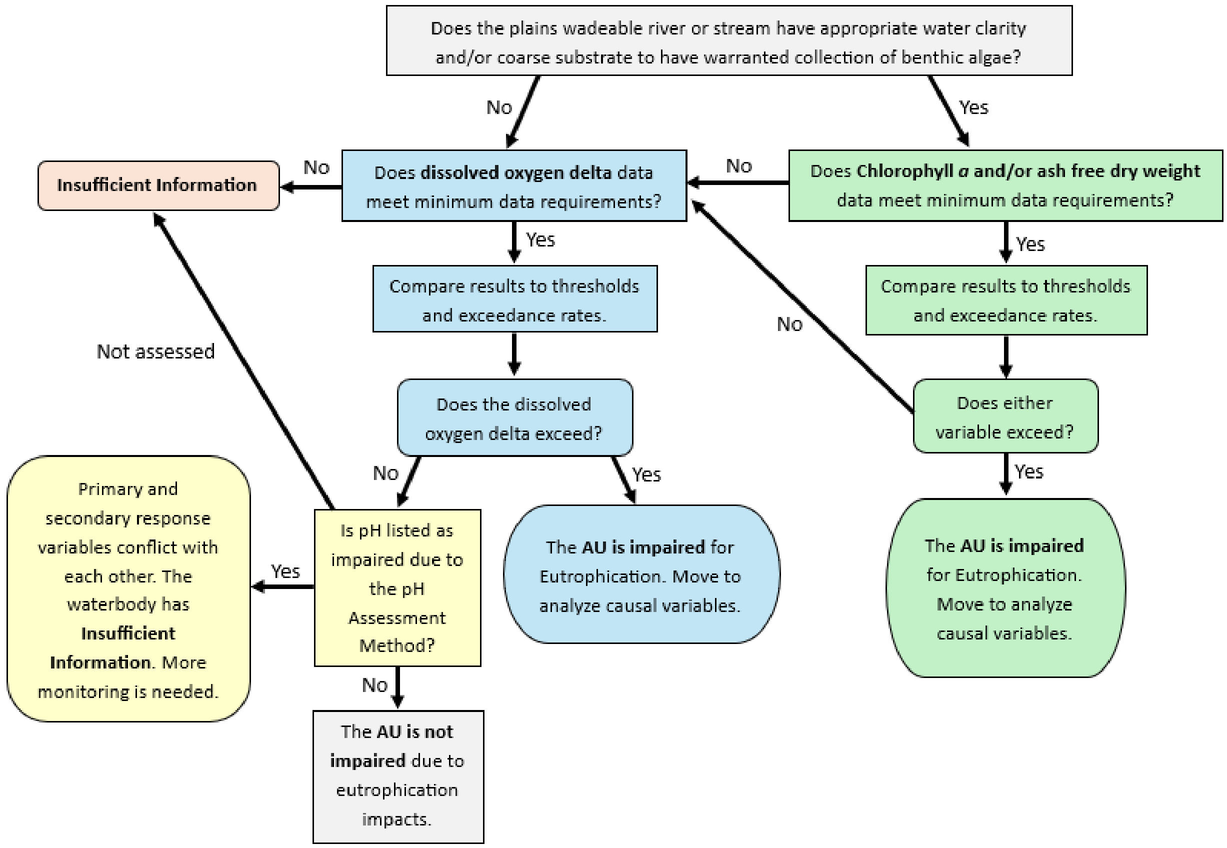


Figure 2. Plains Stream response variable decision framework for wadeable rivers and streams.

5.7 Overwhelming Evidence of Eutrophication Impairment -All Regions

Some circumstances related to eutrophication are severe enough that a rigorous data collection effort for multiple data parameters is not required; the narrative standard at ARM 17.30.637(1)(e) as it pertains to eutrophication can be assessed with limited information. Photo documentation with associated latitude and longitude and description of location and date will suffice. Below are conditions that can be considered overwhelming evidence; these apply equally to all wadeable streams and medium rivers across the state. These conditions are likely to be intertwined with organic pollution problems which impact dissolved oxygen concentrations.

- Fish kills involving massive growths of senescing algae mats. These mats may be attached to the bottom or floating. Dissolved oxygen levels at dawn (if measured) may at times be <1 mg/L.

- Filamentous algal growth covering the entire bottom from bank to bank or nearly so and extending continuously for a substantial longitudinal distance (>150m) at multiple sites along an AU. Use the photographs below (**Figures 6** and **7**) as guides. Don't confuse these conditions with sporadic, longitudinally-patchy growths of heavy filamentous growth, in between which there is lighter algal growth. The latter scenario is not extreme enough to warrant overwhelming evidence and should be sampled/assessed per methods in this assessment method.

Figure 7. Photographs of heavy, bank-to-bank and longitudinally continuous Cladophora growth.

Figure 8. Massive Cladophora growth in the Clark Fork River, MT, 1984.
This excess algae is aptly named "blanket weed". *Photo courtesy of Dr. Vicki Watson, University of Montana.*

5.8 Spring Creeks

Cold water spring creeks have special conditions (e.g., more macrophytes, higher nitrate concentrations, larger diel dissolved oxygen changes) that set them apart from spring runoff influenced streams. The narrative standard at ARM 17.30.637(1)(e) applies to cold water spring creeks but will require the assessor's best professional judgement to identify the most appropriate response and causal thresholds to apply when assessing eutrophication. Note also that Big Spring Creek (Casino Creek to mouth (Judith

River), MT41S004_020) is not included among the spring creeks described in this section because Big Spring Creek is influenced by 23 non-spring tributaries. **Instead, use the plains assessment process in Section 5.6 for this specific assessment unit of Big Spring Creek.**

Most Montana spring creeks are inventoried (Decker-Hess 1989), making it clear which waterbodies should be excluded from the current assessment method. Assessors should use Decker-Hess (1989) to determine if an assessment unit or reach is a spring creek. Verified spring creeks that do not appear in Decker-Hess 1989 should be evaluated and assessed on a case-by-case basis. **If the assessor suspects they are working in an unidentified spring creek, next steps should be discussed with their supervisor.**

5.8.1 Overwhelming Evidence of Eutrophication Impairment in Spring Creeks

Healthy spring creeks will naturally have high densities of macrophytes (aquatic vascular plants), but usually only limited densities of benthic filamentous algae or filamentous algae attached or entangled with the macrophytes. If benthic algae conditions in a spring creek resemble those shown in the figures in **Section 5.7** above, the spring creek may be listed as impaired for eutrophication due to not attaining the narrative standard at ARM 17.30.637(1)(e) based on overwhelming evidence.

5.9 Consideration of Site-specific Conditions

Although ecoregion boundaries are clearly mapped, until a site visit has occurred there may remain a certain degree of uncertainty as to whether a waterbody is better viewed as a higher or lower gradient stream within the mountainous regions. Primary response variable thresholds in **Table 3** and **Table 4** are influenced by environmental factors besides total nitrogen and total phosphorus concentrations; for example, water surface slope is a key variable influencing macroinvertebrate metrics (Suplee 2026). DEQ has analyzed confounding environmental factors that influence the response variables (see Schulte and Craine 2023; Suplee 2023; Suplee 2026) and DEQ's goal is that the most reasonable response variables and thresholds be applied when assessing a waterbody.

In deciding how a mountainous or transitional waterbody is best assessed, consider whether the site has a water surface slope ≤1% (i.e., it is low gradient). In addition to water surface ≤1%, other naturally occurring characteristics that qualify a waterbody as being viewed as a low gradient site are:

- The stream substrate is more dominated by small gravel, sand, and silt rather than gravels and cobbles;
- Presence of sparse macrophyte (aquatic vascular plant) populations; and
- Glides with minimal water surface disturbance interspersed between more turbulent riffles and runs along the longitudinal geomorphic pattern.

The opposite situation may occur as well. A reach of stream or river could have water surface slope <1% but have substrate dominated by coarse gravels and cobbles, no macrophytes, and have turbulent riffles and few or no glides. It would be more suitable to apply the mountainous or transitional thresholds in this case.

There may have been site-specific modeling or similarly detailed project work carried out on the waterbody in question. Results from such work, for example site-specific identification of the key causal variable, site specific causal-response relationships, or site-specific adjustments to response variables may be given consideration.

An assessor using this assessment method may, through data collection, encounter situations that may warrant site specific thresholds. Such situations should be brought to the attention of their supervisor.

5.9.1 Influence of Dams

In Montana, conditions resulting from the reasonable operation of dams in existence as of July 1, 1971, are natural (§ 75-5-306(2), MCA; ARM 17.30.602(17)). Wadeable streams and medium rivers below dams have special conditions (e.g., more macrophytes and larger diel dissolved oxygen changes) that set them apart from rivers and streams that are not influenced by dams. Assessing conditions related to eutrophication in waterbodies influenced by dams must employ best professional judgement when selecting appropriate response and causal variable thresholds.

5.9.2 Influence of Salinity on Beck's Biotic Index (version 3) in Low Gradient Sites (Western and Transitional Ecoregions Only)

If an assessment reach has a low gradient (≤ 1% water surface slope) and elevated specific conductance, and Beck's Biotic Index (but not dissolved oxygen delta) has not attained its threshold, an assessor should complete watershed source assessment to determine if there are likely anthropogenic salinity sources. If **no** anthropogenic sources of elevated salinity are present, an assessor will review the specific conductivity adjustments (**Appendix C**) to the Beck's Biotic Index scores to adjust the result. The adjusted Beck's Biotic Index score result is then compared to the thresholds provided in **Table 5**.

5.10 Document Assessment Decisions and Review with Management

The assessor must document all data and decisions made pertaining to eutrophication standards attainment decisions and beneficial use support determinations for assessment units. Waterbodies identified as impaired due to eutrophication are classified as a category five pollutant, which means the impairment needs to be addressed by a TMDL. Causal variable(s) determined to likely be influencing eutrophication will be documented within the Water Quality Assessment and Reporting Documentation (WARD) database. Assessment decisions are reviewed by the Monitoring and Assessment Section Supervisor and may be reviewed by the Quality Assurance Officer and managers or staff from other DEQ programs. Impairment listings in Montana's Integrated Report will be identified by "Eutrophication" listings.

6.0 Potential Sources

Probable sources of impairment are the activities, facilities, or conditions that generate the pollutants that prevent surface waters from meeting water quality standards. The following sources are most commonly associated with eutrophication impairment listings in Montana; additional selections are available in the WARD database if needed:

- Livestock (Grazing or Feed Operations)
- Agriculture
- Animal Feeding Operations (NPS)
- Confined Animal Feeding Operations – CAFOS (Point Source)
- Crop Production (Crop Land or Dry Land)
- Crop Production (Irrigated)
- Crop Production (Non-Irrigated)
- Dam or Impoundment

- Discharge from Municipal Separate Strom Sewer System (MS4)
- Golf Courses
- Grazing in Riparian or Shoreline Zones
- Internal Nutrient Recycling
- Manure Runoff
- Municipal (Urbanized High Density Area)
- Municipal Point Source Discharges
- Natural Sources
- On-site Treatment Systems (Septic Systems and Similar Decentralized Systems)
- Urban Runoff/Storm Sewers
- Watershed Runoff following Forest Fire
- Waterfowl
- Mining

7.0 Assessment Documentation

Data collected by DEQ is stored in DEQ's MT-eWQX Enterprise (EQuIS) database and is uploaded weekly to the Water Quality Portal (EPA, USGS and NWQMC 2018). Assessment outcomes for individual assessment units, including data summaries, impairment determinations, and beneficial use support determinations, are documented in WARD system and publicly available via Montana DEQ's Clean Water Act Information Center (CWAIC) and within Montana's integrated reports (available at https://deq.mt.gov/water/resources).

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APPENDIX A. DETERMINING SAMPLE INDEPENDENCE

According to definitions in Hurlbert (1984), much sampling carried out by DEQ on individual streams tends to violate spatial and temporal independence assumptions and results in pseudoreplication. For example, samples collected over time at a site can be serially correlated, which precludes temporal independence (Hurlbert 1984). However, the statistical views advocated by Hurlbert are not universally supported; contrary opinions on the matter can be found in the literature (Stewart-Oaten and Murdock 1986; Stewart-Oaten et al. 1992; Osenberg et al. 1994) and have led to what one journal referred to as a "healthy debate" (*Ecological Applications*, volume 4, No. 1, 1994). In general, more needs to be known about detection of non-independence and the frequency with which temporally independent samples can be collected (Underwood 1994).

A.1 TEMPORAL INDEPENDENCE

Time-series collected samples from a site may be used in inferential statistical testing, if used cautiously; this requires that one assumes that actual trends in time are identical in magnitude and direction for all the sites across the study (Norris and Georges 1993). Osenberg et al. (1994) examine time-series serial correlation of physical and biological measurements in a BACI (Before-After-Control-Impact) study and conclude that, in the marine environment they study, sampling can occur at a site every 60 days without yielding substantial serial correlation.

DEQ recognizes the issue of temporal pseudoreplication, but also needs to be practical about the reality of sampling streams which, by their very nature, make collection of independent samples difficult. In DEQ's reference project (Suplee et al. 2005), 30 days has generally been used as a minimum time span between sampling events at a site to infer temporal independence of water samples. This time span was based on the experiential observation that, during the brief Montana summer, substantial changes in flow, temperature, and vegetation (both riparian and instream) occur from month to month, changes that would likely affect water quality. But Stewart-Oaten et al. (1986) recommend that the assumption of temporal independence be tested, rather than assumed. The Durbin-Watson test statistic is widely used to check for time-series serial correlation. Stream sites with monthly nutrient sampling during the summer were available in Montana, and some of these sites were tested using the Durbin-Watson statistic Results are shown in **Table A-1** below.

Table A-1. Durbin-Watson values for time-series collected nutrient samples at selected sites. All Samples were Collected Approximately 30 Days apart. Nutrients Showing Probable Time-series Serial Correlation (95% Confidence Level) are Highlighted.

				Nutrient	**Nutrient**	**Nutrient**
Stream Site	**Months Sampled**	**Time Range**	**n**	**Total N**	**Total P**	**NO_{2+3}**
Rock Creek Site 2	June, July, Aug, Sept	2001-2004	12	1.18	1.43	2.3
Clark Fork R. at Deer Lodge (site 9)	July, Aug, Sept	1998-2006	25	1.81	1.78	1.68
Clark Fork R. above Little Blackfoot R. (site 10)	July, Aug, Sept	1998-2006	26	2.01	1.57	1.46
Clark Fork R. above Flathead R. (site 25)	July, Aug, Sept	1998-2006	26	1.76	1.21	2.08

In general, Durbin-Watson values around 2 mean there is no serial correlation, whereas values greater than approximately 2.5 or less than about 1.5 lead one to suspect negative or positive serial correlation, respectively (Neter, et al. 1989; Ott 1993). What can be concluded from this limited analysis? Most nutrients did not show serial correlation, and one of the three that did is borderline cases (statistic =1.43, but power of test very low). Overall, it appears that serial correlation is present in nutrient samples collected a month apart, but the effect is very weak. It is evident that 30-day separated water samples can provide a fairly high degree of independence for nutrients.

2016 Update:

Methods. In 2016 we made an in-depth analysis of temporal sample independence patterns as manifested by Montana stream nutrient datasets. Historic nutrient data (1968-2012) from legacy STORET and MT-eWQX were queried to find contiguous daily, weekly, or biweekly datasets (or close approximations thereof). TP, TKN, and NO_{2+3} data were queried (little TN data exists in older datasets). Time-series datasets were located for 87 different wadeable streams, from eastern and western MT, representing both heavily polluted and relatively un-impacted sites (**Table A-2**). Datasets with >16.6% non-detects (EPA 2006) or with other flagged data were eliminated. Durbin-Watson significance tables from Savin and White (1977) were used. The critical bound for decisions (dU, the upper bound[1]) on each side of the ideal score of 2 was used to determine if a given dataset demonstrated serial correlation or not (95% confidence level). We incorporated the previously-completed monthly dataset results (**Table A-1** above), with updated dU decision thresholds as necessary, per Savin and White (1977). Time-series datasets were then categorized as representing 3-day (i.e., sampling occurred about every three days), weekly, or biweekly sampling intervals. If a dataset did not exactly match one of these categorical intervals, it was placed in the closest category.

Table A-2. Sites with nutrient datasets regularly sampled (every 3-days, weekly, biweekly).

Station ID	Site Name	Nutrient	Interval
3732PR02	PRICKLY PEAR CK JUST ABOVE KAISER CEMENT	TKN	3-day
5614AS03	ASHLEY CR JUST ABV KALISPELL WWTP OUTFALL	TP, TKN	3-day, weekly
CBM-SW-1	0.25 MI UPSTREAM OF CONFLUENCE WITH DAISY CREEK	NO3	weekly
5514AS01	ASHLEY CR @ BRIDGE ABV CONFL WITH FLATHD	TP, NO3	weekly
5613AS02	ASHLEY CR @ BRIDGE ABV FOREST PROD CO.	TP, TKN, NO3	weekly
5613AS03	ASHLEY CR @ BRIDGE BEL FOREST PROD CO.	TP, TKN, NO3	weekly
5614AS08	ASHLEY CR @ BRIDGE NR DEMERSVILLE SCHOOL	TP, NO3	weekly
5614AS01	ASHLEY CR @ GAUGE STAT ABV STORM SEWER	TP, TKN	weekly
5513AS01	ASHLEY CR AT BRIDGE ABOVE SMITH LAKE	TP, TKN	weekly
5613AS01	ASHLEY CR AT BRIDGE BEL SMITH LAKE	TP, TKN	weekly
5512AS01	ASHLEY CR AT BRIDGE ON ROGERS LAKE ROAD	TP	weekly

[1] The procedure tests the null hypothesis of zero autocorrelation in the residuals *vs.* the alternative that residuals are positively autocorrelated. If a test result is greater than dU, the null is not rejected (i.e., no serial correlation exists). Between dU and dL is the test's gray zone (result is inconclusive). If the result is lower than dL, the null is always rejected (there is serial correlation). Either boundary (dU or dL) can be used as the critical threshold. The dU boundary, which we used, is more conservative as more cases will be found with serial correlation.

Station ID	Site Name	Nutrient	Interval
3127BL01	BLACKTAIL CREEK ABOVE SILVER BOW	TP, TKN, NO3	weekly
5317BO01	BOND CREEK-UPPER	NO3	weekly
SEELEY1	CLEARWATER RIV AB RAINY LK	NO3	weekly
SEELEY4	CLEARWATER RIV BL LK ALVA	NO3	weekly
SEELEY8	CLEARWATER RIV BL SEELEY LK	NO3	weekly
SEELEY9	DEER CREEK NR SEELEY LAKE	NO3	weekly
3125GE05	GERMAN GULCH CREEK ABV CONF W SILVERBOW C	TP, TKN, NO3	weekly
5317HA01	HALL CREEK-UPPER	NO3	weekly
3326MI02	MILL-WILLOW BYPASS NR WARMSPRINGS POND	NO3	weekly
SEELEY10	SEELEY CRK AT SEELEY LAKE TOWN	NO3	weekly
3225SI03	SILVER BOW CR AT STUART ST BRIDGE OPPORT	TP,TKN, NO3	weekly
3127SI07	SILVER BOW CR BEL COLO TAILS & SLTR HOUSE	TKN, NO3	weekly
3126SI01	SILVER BOW CREEK 1 MILE BELOW RAMSAY	TP, TKN, NO3	weekly
3125SI02	SILVER BOW CREEK AT ROAD TO FAIRMONT	TP, TKN, NO3	weekly
3326SI01	SILVER BOW CR-LOWER PH SHACK NR WARM SPRG	TP, NO3	weekly
3127SI01	SILVERBOW CR ABV CONFL OF BLACKTAIL CREEK	TP, TKN, NO3	weekly
3324WA01	WARM SPRINGS CR 1 MILE BELOW MYERS DAM	TP, NO3	weekly
FL1003	BOHANNON CR 11 MI SSE BIG FORK, MT.	NO3	weekly, biweekly
3326WA01	WARM SPRINGS CR AT MOUTH NR SILVER BOW CR	TKN, NO3	weekly, biweekly
FL8008	ALDER CR 20 MI. WNW WHITEFISH MT	NO3	biweekly
2529BI01	BIGHOLE RIVER NEAR TWIN BRIDGES	TP	biweekly
2354BU01	BUTCHER CR - NR COONEY DAM RD	TP, TKN,NO3	biweekly
2453BU01	BUTCHER CR-NR MOUTH	TP, TKN,NO3	biweekly
2253BU01	BUTCHER CR-NR SH78	TP, TKN,NO3	biweekly
3555CA01	CARELESS CREEK AT MOUTH NR RYEGATE	TP	biweekly
3526CL01	CLARK FORK R -BRIDGE JUST ABV DEER LODGE	TP, NO3	biweekly
SEELEY7	CLEARWATER RIV AB SEELEY LK	NO3	biweekly
4814CR02	CROW CR BLW LOWER CROW RES NR RONAN	TP,TKN	biweekly
4815CR02	CROW CREEK ABOVE LOWER CROW RES NR RONAN	TP,TKN	biweekly
4714DU01	DUBLIN GULCH AT MOUTH NR MOIESE	TP,TKN	biweekly
4415FI01	FINLEY CK AT MOUTH NR ARLEE	TP, TKN	biweekly
2738GO04	GODFREY CK EAST FORK MOUTH	TP, NO3	biweekly
2738GO02	GODFREY CR - EAST FORK MOUTH	TP	biweekly
2738GO05	GODFREY CR - NEAR CHURCHILL	TP, NO3	biweekly

Station ID	Site Name	Nutrient	Interval
FL8014	GRIFFIN C, LOWER 20 MI WSW WHITEFISH MT	NO3	biweekly
FL8015	HAND C LOWER 21 MI WSW WHITEFISH, MT	NO3	biweekly
FL8016	HAND C, UPPER 23 MI WSW WHITEFISH,MT	NO3	biweekly
4515JO01	JOCKO R ABOVE VALLEY CK NR ARLEE	TP	biweekly
4416JO01	JOCKO RIVER ABOVE FINLEY CK NR ARLEE	TP,TKN	biweekly
4614JO01	JOCKO RIVER NEAR MOUTH AT HW 212 BRIDGE	TP,TKN	biweekly
4814LI01	LITTLE BITTERROOT R NEAR MOUTH AT SLOAN	TP,TKN	biweekly
FL8011	LOGAN C, LOWER 14 MI W WHITEFISH, MT	NO3	biweekly
FL8012	LOGAN C, MIDDLE 15 MI. SW WHITEFISH MT	NO3	biweekly
FL8013	LOGAN C, UPPER 16 MI SW WHITEFISH, M T.	NO3	biweekly
4615MI02	MISSION CK ABOVE SABINE CK NR ST IGNATIUS	TP,TKN	biweekly
4714MI01	MISSION CK AT HWY 212 NR MOIESE	TP,TKN	biweekly
4616MI03	MISSION CK BLW MISSION RES ABV DRY CR	TP,TKN	biweekly
4916MU01	MUD CK (UPPER) NR PABLO	TP	biweekly
4815MU01	MUD CREEK AT HWY 211 NR RONAN	TP,TKN	biweekly
5034MU01	MUDDY CREEK ABOVE DRAIN E	TP, NO3	biweekly
5034MU03	MUDDY CREEK ABOVE DRAIN M	TP, NO3	biweekly
5134MU02	MUDDY CREEK ABOVE EAST AND WEST FORKS	TP, NO3	biweekly
5034MU02	MUDDY CREEK ABOVE SPRING COULEE	TP, NO3	biweekly
5134MU03	MUDDY CREEK AT BRIDGE IN CORDOVA-WEST OF	TP, NO3	biweekly
3866MU01	MUSSELSHELL R AT HWY BRIDGE E OF MELSTONE	TP, NO3	biweekly
3455MU01	MUSSELSHELL RIVER AT BRIDGE IN RYEGATE	TP	biweekly
3650MU01	MUSSELSHELL RIVER AT HARLOWTON UPSTREAM	TP	biweekly
3457MU01	MUSSELSHELL RIVER NEAR LAVINA	TP	biweekly
3553MU01	MUSSELSHELL RIVER SOUTH OF SHAWMUT	TP	biweekly
3660MU01	MUSSLSHELL RIVER AT USGS STATION ROUNDUP	TP	biweekly
4815NI01	NINEPIPE RES OUTLET NR CHARLO	TP,TKN	biweekly
FL1001	NO FK LOST CR 3 MI. SE SWAN LAKE, MONTANA	NO3	biweekly
4916NO01	NORTH CROW CK ABOVE RONAN	TP	biweekly
4816NO02	NORTH CROW CK AT MOUTH NR RONAN	TP,TKN	biweekly
3646NO01	NORTH FORK MUSSELSHELL @ HWY 12	TP	biweekly
FL1002	PORCUPINE CR 5 MI. SW SWAN LAKE, MT	NO3	biweekly
4716PO01	POST CK ABOVE KICKING HORSE RES NR RONAN	TP,TKN	biweekly
4715PO01	POST CK AT HIGHWAY 93 BRIDGE	TP,TKN	biweekly
4715PO04	POST CK NR MOUTH AT CO.RD BRIDGE NR ST IG	TP,TKN	biweekly
4615SA01	SABINE CK AT MOUTH NR ST IGNATIUS	TP,TKN	biweekly
HUN103	SF FLATHEAD RIV 500FT BL H-H DAM	NO3	biweekly
4916SP01	SPRING CK NR RONAN	TP	biweekly
4815SP01	SPRING CREEK NEAR MOUTH NEAR RONAN	TP,TKN	biweekly
5231TE03	TETON R BELOW PRIEST BUTTE LAKE DISCH	NO3	biweekly
FL8021	TRIB TO SQUAW MEADOWS C 23 MI SW WHITEFISH MT	TP	biweekly
4815WE01	WEST FORK MUD CK AT HWY 211 NR RONAN	TP,TKN	biweekly

Results. The most common dataset time interval was biweekly (n=78), followed by weekly (n=55) and 3-day (n=3). The most common dataset size was n=7 samples (55 cases), the largest was n=20 (a 3-day interval dataset). Results are plotted in **Figure A-1.** This is the best fit relationship (natural log) and is curvilinear, with an R^2 of 0.84. **Figure A-2** is the same data but includes one assumed data point; it was assumed that if streams were sampled every minute (i.e., high frequency), all case studies would have serial correlation (think of TSS being sampled minutely on the rising limb of a hydrograph). The assumed data point provides a reasonable anchor point on the Y-intercept when deriving a best-fit curvilinear relationship using an algorithm (Gauss-Newton in this case). The relationship in **Figure A-2** is the best fit (i.e., lowest error sum of squares), but a 2nd order polynomial also reasonably fits these data (R^2 = 0.81; y = $0.0016x^2 - 0.0764x + 1$). The weekly and biweekly results do not plot exactly where one would expect, but the overall pattern of a curvilinear relationship with increasing serial correlation with fewer days between sampling events is clearly evident in **Figures A-1** and **A-2**.

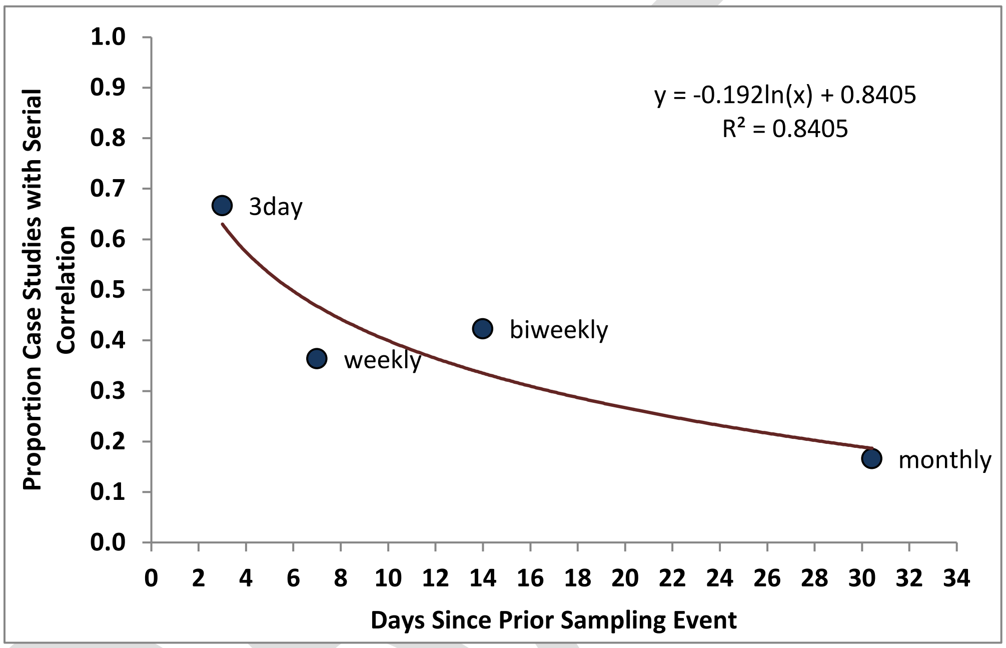

Figure A-1. Best-fit curvilinear relationship (natural log) between days since prior sampling event (X) vs. the proportion of nutrient sampling case studies with serial correlation (Y).

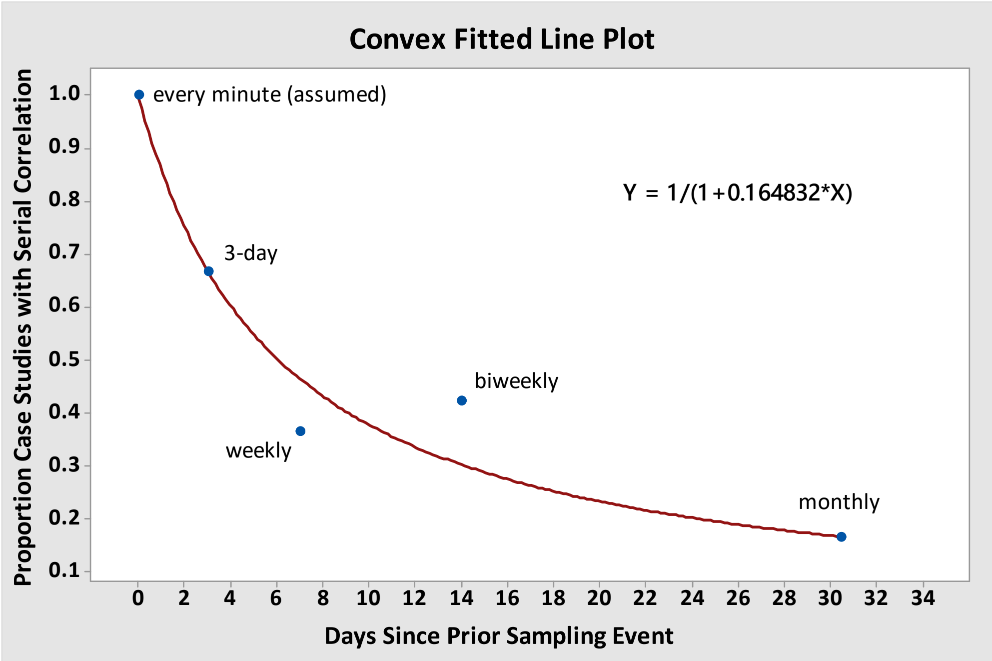

Figure A-2. Best-fit curvilinear relationship between days since prior sampling event (X) *vs.* the proportion of nutrient sampling case studies with serial correlation (Y).
The line was fit using the Gauss-Newton algorithm in MiniTab 17 and includes an assumed data point at the Y-intercept.

Discussion and Conclusion. Based on the earlier work (**Table A-1**), DEQ has accepted—per this assessment method—that 17% of nutrient-sampling events would have serial correlation at the monthly interval sampling frequency. This was considered a tolerable level. This updated, more in-depth analysis was reviewed by **DEQ Standards Modeling staff and management in March 2016 and we concluded that the sampling interval could be reduced to two weeks.** The relationship between days since the prior sampling event and percentage serial correlation in dataset case studies is clearly curvilinear (**Figures A-1, A-2**). In **Figure A-2** the curve's apex is about 7 days, and at two weeks the curve is into the flattening part of the curved relationship. In both figures, at two-week sampling intervals, the percent of sampling events with serial correlation would be about 30%—below a critical concern level of 50%. Based on experience using this document's assessment method over the past 5 years, allowing sampling to occur at two-week intervals provides big advantages for completing field work and assembling adequately sized nutrient datasets during the short Montana field season. Given our better understanding of the sampling interval/serial correlation relationship, we believed the pros of allowing two-weeks between events greatly outweigh the cons of somewhat more cases with serial correlation.

A.2 Spatial Independence

DEQ is aware that spatial independence is also a concern. Water flows from upstream to downstream, consequently influencing the spatial independence of downstream sampling sites. No generally applicable spatial minimums were found as of this writing. U.S. EPA guidance (EPA 2002) generally glosses over the topic of spatial independence in streams.

To address spatial independence, we tested a Montana dataset. We used the pre-dosing baseline data collected as part of the Box Elder Creek nutrient dosing study (Suplee et al. 2016). We found that total nutrient samples collected within hours of one another at two sites located 0.73 stream miles apart were not spatially correlated. We compared nutrient samples collected from the Low Dose site to those collected on the same day at the High Dose site which is 0.73 miles downstream. Box Elder Creek is perennial and was flowing during all sampling events. No tributary intervenes between the sites. Samples were collected within 1-2 hours of one another, during the summer index period. We only considered samples collected *prior to* nutrient dosing, as these are comparable to what one would encounter during routine stream sampling/assessment. Using the Rank von Neumann test , we found that there was no serial correlation for total N or total P (i.e., we could not reject the null hypothesis “no serial correlation”), at an alpha of 0.05. There was serial correlation for Soluble Reactive Phosphate (SRP). We were unable to assess soluble N as there were too many non-detects in the datasets, which led to too many rank-ties; too many rank-ties precludes proper statistical evaluation (Gilbert 1987).

Spatial independence can therefore be established (albeit as rules of thumb) for total nutrients as a minimum of about 1 mile between two sites. Other factors leading to spatial independence include a tributary confluencing on a stream between two sampling sites, or if major landform or land use changes occur along the reach.

Giving consideration to our findings, below are guidelines for establishing independence of samples collected within an assessment reach:

- Sites (or short reaches equivalent to sites) should be located a minimum of 1 stream mile apart.
- Sites may be placed < 1 mile apart along the assessment reach if there is a flowing tributary confluencing with the segment between the two sites.
- Try to collect water samples starting at the downstream end of the assessment reach moving upstream, to avoid re-sampling the same water.
- Land use changes and land form changes should be considered and can be used to help define (1) breaks between assessment reaches and/or (2) additional sampling sites within an assessment reach.
- And, per **Section A.1**, nutrient samples collected at the same site (or short reach) should be collected at least 14 days apart.

Total nutrient samples that meet the above conditions may generally be considered both spatially and temporally independent for the purposes of determining compliance with the nutrient criteria. As such, they may be used in inferential statistical analyses and to make conclusions about the assessment reach in question.

Precautionary Considerations: The last bullet above (temporal independence resulting from approximate 14-day time spans) is <u>not</u> applicable for some bioassemblage samples (e.g., macroinvertebrates, fish). These organism populations operate on different (longer) time scales from water samples and diatoms and may show considerable year-to-year stability. Please see **Section 9.0** of Suplee (2004) and Bramblett *et al.* (2005) for more details on temporal patterns of these biological assemblages. Diatom populations tend to shift quickly, within 1-5 weeks, in response to environmental changes (LaVoie et al. 2008). Thus, this rate of change is sufficient to be able to consider diatom sampling events spaced 30 days apart as being largely independent of one another.

APPENDIX B. ASH FREE DRY WEIGHT THRESHOLD IDENTIFICATION

Ash Free Dry Weight (AFDW) collected from natural stream-sediment surfaces is a useful measurement for estimating algal biomass. The laboratory method basically oxidizes and reports back the mass of all organic material in the sample (American Public Health Association 1998). It provides an additional means of assessing accumulated algal biomass independent of Chl-*a.* Chl-*a* levels tend to be highest during peak growth, and then decline later as the Chl-*a* molecules degrade as the algae senesce (Stevenson et al. 1996). For varying reasons, including late season algal senescence, an assessor may find low Chl-*a* values in spite of the presence of a large biovolume of algal material (e.g., see DEQ 2024).

AFDW should be determined for all samples when Chl-*a* is collected. AFDW can be determined from the same algae sample in a subsequent analysis that follows the Chl-*a* analysis. Site average AFDW can be determined from individual replicates, or as a weighted average.

Note: AFDW results from core samples should never be included in determining a site's average AFDW. The method measures organic material from the entire core sample, not just the surface where the algae are growing, and will therefore over-report AFDW. Core samples are only for Chl*a*.

DEQ has good estimates of what comprises too much algal AFDW. In Suplee et al. (2009), the threshold Chl-*a* level of 150 mg/m^2 corresponds to 36 g AFDW/m^2. In New Zealand, extensive analysis of algal AFDW resulted in a recommendation of 35 g AFDW/m^2 as the maximum level for gravel/cobble streams, to protect recreation use (Biggs 2000). In DEQ's whole-stream nutrient dosing study (Suplee et al. 2019), the AFDW corresponding to 127 mg Chl *a*/m^2 (the Chl-*a* level linked to late-season DO problems) is 33 g/m^2. Long-term monitoring in the Clark Fork River (1998-2009) shows that the average summer AFDW at sites that do not develop nuisance algae (i.e., they consistently have <150 mg Chl-*a*/m^2) ranged from 17 to 48 g AFDW/m2 (mean: 27 g AFDW/m^2). These considerations indicate that site average AFDW (i.e., the weighted average of replicates collected at a site, excluding cores) should be no greater than **35 g AFDW/m^2.** This value should generally be protective of fish and aquatic life uses and the recreation use.

APPENDIX C: SPECIFIC CONDUCTIVITY VS. BECK'S BI (V3) LOOK-UP TABLES

logConductance	Specific Conductivity (µS/cm)	Residual (add this value to Beck's Biotic Index (v3) Threshold)
1.90855	81.0	14.52
1.91855	82.9	14.44
1.92855	84.8	14.34
1.93855	86.8	14.22
1.94855	88.8	14.08
1.95855	90.9	13.91
1.96855	93.0	13.72
1.97855	95.2	13.51
1.98855	97.4	13.28
1.99855	99.7	13.02
2.00855	102.0	12.74
2.01855	104.4	12.44
2.02855	106.8	12.12
2.03855	109.3	11.78
2.04855	111.8	11.42
2.05855	114.4	11.04
2.06855	117.1	10.64
2.07855	119.8	10.23
2.08855	122.6	9.80
2.09855	125.5	9.37
2.10855	128.4	8.92
2.11855	131.4	8.46
2.12855	134.4	7.99
2.13855	137.6	7.52
2.14855	140.8	7.04
2.15855	144.1	6.57
2.16855	147.4	6.09
2.17855	150.9	5.62
2.18855	154.4	5.15
2.19855	158.0	4.68
2.20855	161.6	4.23
2.21855	165.4	3.78
2.22855	169.3	3.35
2.23855	173.2	2.93
2.24855	177.2	2.52
2.25855	181.4	2.14
2.26855	185.6	1.77
2.27855	189.9	1.42
2.28855	194.3	1.08
2.29855	198.9	0.77
2.30855	203.5	0.48
2.31855	208.2	0.21
2.32855	213.1	-0.04
2.33855	218.0	-0.28
2.34855	223.1	-0.49
2.35855	228.3	-0.68
2.36855	233.6	-0.85
2.37855	239.1	-1.00
2.38855	244.7	-1.14
2.39855	250.4	-1.26

logConductance	Specific Conductivity (µS/cm)	Residual (add this value to Beck's Biotic Index (v3) Threshold)
2.40855	256.2	-1.36
2.41855	262.2	-1.45
2.42855	268.3	-1.52
2.43855	274.5	-1.59
2.44855	280.9	-1.64
2.45855	287.4	-1.69
2.46855	294.1	-1.74
2.47855	301.0	-1.78
2.48855	308.0	-1.82
2.49855	315.2	-1.85
2.50855	322.5	-1.90
2.51855	330.0	-1.94
2.52855	337.7	-1.99
2.53855	345.6	-2.05
2.54855	353.6	-2.12
2.55855	361.9	-2.20
2.56855	370.3	-2.28
2.57855	378.9	-2.38
2.58855	387.8	-2.49
2.59855	396.8	-2.60
2.60855	406.0	-2.73
2.61855	415.5	-2.87
2.62855	425.2	-3.01
2.63855	435.1	-3.17
2.64855	445.2	-3.33
2.65855	455.6	-3.50
2.66855	466.2	-3.67
2.67855	477.0	-3.85
2.68855	488.1	-4.04
2.69855	499.5	-4.22
2.70855	511.2	-4.42
2.71855	523.1	-4.61
2.72855	535.2	-4.81
2.73855	547.7	-5.01
2.74855	560.5	-5.21
2.75855	573.5	-5.40
2.76855	586.9	-5.60
2.77855	600.6	-5.80
2.78855	614.5	-5.99
2.79855	628.9	-6.19
2.80855	643.5	-6.38
2.81855	658.5	-6.56
2.82855	673.8	-6.75
2.83855	689.5	-6.93
2.84855	705.6	-7.11
2.85855	722.0	-7.28
2.86855	738.8	-7.45
2.87855	756.1	-7.62
2.88855	773.7	-7.78
2.89855	791.7	-7.94
2.90855	810.1	-8.10