Cramer Fish Sciences

Interpretation of Periphyton Samples from Montana Streams

December 2010

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Introduction

This paper provides guidance for interpretation of diatom sample results from wadeable streams in Montana using Increaser Taxa identified by Teply (2010). Increaser Taxa are taxa that, as a group, exist in detectable amounts in all stream classes and demonstrate a meaningful, measurable, and significant response to environmental stressors. They are used to predict, with stated reliability, the probability of impairment under 303(d) guidelines. Of specific interest to the State is discriminating impairment to aquatic life use support in instances where diatom community response was most likely; that is, impairment due to sediment, nutrients, and/or metals. Increaser Taxa are regionally specific, reflecting meaningful differences in geology and hydrology and resulting natural variability taxa assemblages. Five sets of Increaser Taxa were identified by Teply (2010) that met these water quality assessment objectives:

1) Warm Water Fisheries – Sediment Increaser Taxa
2) Warm Water Fisheries – Nutrient Increaser Taxa
3) Cold Water Fisheries, Middle Rockies Ecoregion – Sediment Increaser Taxa
4) Cold Water Fisheries, Northern/Canadian Rockies – Sediment Increaser Taxa
5) Cold Water Fisheries, Northern/Canadian Rockies – Nutrients Increaser Taxa

This document is organized into two sections. The first provides necessary background regarding stream groups, impairment causes, sampling and laboratory methods, and discriminant models underlying this guidance. The second provides step-by-step procedures for using Increaser Taxa to interpret periphyton sample results. It also includes suggested language. A detailed accounting of the Increaser Taxa development is presented in Teply (2010), a Technical Report that was developed concurrently with this guidance document. This companion document is suggested reading as it provides the empirical basis for the Increaser Taxa presented herein.

Background

Stream Groups

Increaser Taxa lists are differentiated by stream groups within which there is a high degree of similarity among taxa assemblages. Stream groups are based on the analysis of reference taxa assemblages across Montana (Teply and Bahls 2007). Two alternatives were presented; this guidance uses rules based on Montanan Department of Environmental Quality (MT DEQ) fisheries classification (cold or warm) and Level III ecoregions (see **Figure 1**). These rules make effective use of a streams fisheries classification and Level III ecoregion (US EPA 2000). To classify streams into cold or warm water fisheries, we used rules implemented by the State based on Level III and IV ecoregions (see **Appendix A**). Overall classification accuracy using these rules was found to be about 71%.

Figure 1. Dichotomous key to stream groupings using MDEQ Fisheries Classification and Predominant Level III Ecoregion (Alternative I).

1. Site in a warm water fishery?
    - a. Yes..... Warm-Water Stream Group (I.1a)
    - b. No ..... Go To 2
2. Site within the Middle Rockies Ecoregion?
    - a. Yes..... Middle Rockies Stream Group (I.2a)
    - b. No ..... Northern/Canadian Rockies Stream Group (I.2b)

Impairment Causes

Increaser Taxa lists are differentiated according to impairment causes. Teply (2010) developed distinct lists for each impairment cause of interest to the State – sediment, nutrients, or metals. Although metals impairment was found to be naturally occurring along plains streams, it was not considered in our analysis. **Impaired** streams were classified by the subject cause of impairment where aquatic life use support was listed

as none. **Non-impaired** streams were classified as those where the cause of impairment was other than that being considered. Non-impaired streams also included those listed as impaired but for causes other than sediment, nutrients, and metals, such as habitat degradation, flow alteration, or thermal loading; i.e., these stressors are not considered to have an affect on the diatom community.

Sampling/Laboratory Methods

Increaser Taxa lists were developed through analysis of hundreds of samples collected over the last decade from streams throughout Montana and analyzed by several labs and diatomists. Samples collected by the State followed the PERI-1 or PER-1mod protocol (MT DEQ 2005; MT DEQ 2006) and were analyzed by two laboratories (*Hannaea* and the Academy of Natural Sciences of Philadelphia) following the 400 cell count (800 valve) protocol described in DEQ contract (MT DEQ 2004). Methods used for other samples varied, but our best profession judgment is that the consequence of this to interpretation using Increaser Taxa lists is minor. Increaser Taxa are, by definition, commonly occurring taxa. Teply (2010) would typically encounter 350 to 400 distinct taxa in any random draw of 40 samples used in analysis. However, less than about one-fifth of taxa would be considered common and of these, about one-quarter would be observed to increase in impaired samples. These taxa are well-known and should be readily and reliably identified. State protocols for sampling and analysis are recommended for use of Increaser Taxa; however, this guidance should be robust to other protocols which deviate slightly from this standard.

Discriminant Models

Increaser Taxa were evaluated using discriminant analysis which tested the ability of total percent relative abundance (PRA) of taxa on the Increaser Taxa list to discriminate between impaired and non-impaired streams. Discriminant models were evaluated based on their statistical significance (preferably $p<0.05$), and overall classification accuracy (preferably grater than 65%) and false positive rates (preferably less than

30%) in the validation data set. Some discretion was used to consider the influence of outliers and unusual sample circumstances. We also considered diatom autoecology in published sources (e.g., van Dam et al. 1994) and through professional judgment.

An advantage of discriminant models (e.g., versus ANOVA or percentile-based threshold setting) is that discriminant models can be used to directly calculate the probability of class membership (i.e., the probability that a sample represents impaired conditions). For instance, for a given PRA of Increaser Taxa, discriminant models permit us to state the probability that the sample is from an impaired stream. This improves the State's ability to assess naturally flowing streams and was an attraction to selecting the approach. Meaningful application of discriminant models in this way assumes randomness and sample independence and normality and equal variance in metric values among impaired and non-imparied samples. In general, we found that these assumptions were not saliently violated and that bias was minimal.

It is important to note that the probability yielded by discriminant models is just that – a probability. Independent validation of these models indicates an average overall classification accuracy of just over 65% - this is an accuracy level that would be considered marginally statistically significantly better than a flip of a coin. Generally, this uncertainty is reduced as the probability of impairment increases with increasing PRA of Increaser Taxa. Generally, this error is not uniformly distributed either. In application, users will likely find these Increaser Taxa to perform remarkably well throughout a watershed in about two-thirds of water quality assessments. The rest of the time (about one-third of water quality assessments), predictions indicated by Increaser Taxa will have greater uncertainty. This error is not insubstantial, but it is a dramatic improvement compared to classification accuracy using existing biocriteria.

Procedures

Four steps are presented below for using Increaser Taxa for interpretation of periphyton samples from wadeable streams. Step I re-states rules for determining the stream group

associated with the diatom sample. Step II outlines simple procedures for calculating PRA of Increaser Taxa. Step III guides the reader through the determination of probability of impairment using this value. Step IV provides suggested language for stating sample results and interpretation of impairment.

Step I – Determine Stream Group

To classify a sample according to stream group, use rules based on MT DEQ fisheries classification (cold or warm) and Level III ecoregions presented in **Figure 1**. Within these strata, there is a high degree of similarity among taxa assemblages; this is critical to the use of Increaser Taxa. To classify streams into cold or warm water fisheries, use rules implemented by the State based on Level III and IV ecoregions (see **Appendix A**). A GIS database of Level III and IV ecoregion boundaries can be found through the Montana State Library, Natural Resource Information System. In some instances, there will be a stream segment that straddles two stream groups. This occurs frequently along the Rocky Mountain front and streams originating in the isolated mountain ranges in the plains. Professional judgment is required to choose one, or both, groups that best apply.

Step II – Calculate Assessment Metrics

Two metrics are required for interpretation of sample results. The Number of Taxa on the Increaser Taxa list is self-explanatory. Refer to Increaser Taxa lists in **Appendix B** for each stream group and impairment cause. PRA of taxa on the Increaser Taxa list is calculated as the sum of PRAs for each taxon on the Increaser Taxa list that is counted in the sample. PRAs are calculated by dividing the number of valves counted for each taxon by the total number of valves counted in the sample. These values would be calculated independently, of course, for each impairment cause with an Increaser Taxa list reported for the associated stream group. The Warm Water Fisheries and Northern/Canadian Rockies have two sets of Increaser Taxa – for sediment and nutrients – the Middle Rockies only has one – that is for sediment.

Step III – Determine Probability of Impairment

The probability that the sample represents a stream impaired due to either sediment or nutrients can be determined via tables in **Appendix C**. These tables translate PRA values into an associated probability of impairment. Although the underlying probability distribution is normal, these tables are organized by incremental values of probability of impairment. To determine the probability of impairment, simply find the tabled PRA greater than and less than the PRA of Increaser Taxa (determined above) and interpret the associated probability as a range. For very low or very high PRA values, the probability would be interpreted to be less than 5% or greater than 95%, respectively.

As an example, if the percent relative abundance of taxa on the Warm Water Fisheries, Sediment Increaser Taxa list is 23%, the probability that the sample represents a stream impaired by sediment can be interpreted to be about 55 to 65%. If a finer interpretation is desired, the probability of impairment can be interpolated accordingly; a straight-line interpolation is adequate for water quality assessments. If the PRA were 10% less – 13% – then the probability decreases to 35 to 45%. A 10% higher PRA – 33% – would be interpreted to be about 65 to 75%. The 50% probability occurs at about 17.92 PRA; this is the threshold for sediment impairment reported by Teply (2010).

Step IV – Interpret the Sample Result

The purpose of this step is to interpret results within the context of empirical evidence presented by Teply (2010). The following suggested language seeks to minimize inadvertent mis-interpretation or mis-representation of results using Increaser Taxa lists. First, all interpretations using Increaser Taxa should begin with the following statement, clarifying the basis for the interpretation to follow:

> *"Sample diatom taxa counts were evaluated to determine the probability of [Impairment Cause] impairment using the [Impairment Cause] Increaser Taxa List for the [Stream Group]."*

The investigator would then describe the Increaser Taxa appearing in the sample and their autecological importance as indicators of stress due to the impairment cause:

> *"[Number of Increaser Taxa] of [Total Increaser Taxa on the List] diatom taxa on the [Impairment Cause] Increaser Taxa list were counted, representing a total percent relative abundance of [PRA of Increaser Taxa]. These taxa have autecological affinities that make them suitable indicators of [Impairment Cause]."*

Finally, statements regarding the probability of impairment, as determined above, are made as follows:

> *"This indicates that the sample represents a stream that has about a [Probability of Impairment] percent probability of being impaired due to [Impairment Cause] under 303(d) guidelines. This probability is based on past evidence of taxa associated with [Impairment Cause]-impaired streams in [Stream Group] streams. [Impairment Cause] Increaser Taxa do not discriminate other causes of impairment and this result does not indicate whether the stream may or may not be impaired due to other causes."*

Interpretations that are made following this guidance do not preclude interpretations made following other biocriteria (e.g., Bahls 1993) nor does this guidance expressly endorse their use. It is simply provided to facilitate interpretation of diatom samples using Increaser Taxa identified by Teply (2010). The reader is strongly urged to review the discussion of uncertainty associated with Increaser Taxa on Page 4 of this guidance document. It is important that they understand the probabilistic nature of these impairment determinations and the associated error rates. Overall, it is strongly suggested that any interpretations be made by a qualified diatomist and/or aquatic ecologist experienced with Montana streams.

References

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Appendix A: Stream Fisheries Classifications

Cold Water Fisheries

Table 3.1. Ecoregions in which Nutrient-impairment Assessment Methodologies Described in this Section Best Apply.

Ecoregion Scale	Ecoregion Name	Ecoregion Number
Level III	Northern Rockies	15
Level III	Idaho Batholith	16
Level III	Middle Rockies	17
Level III	Canadian Rockies	41
Level IV	Sweetgrass Uplands	42l
Level IV	Milk River Pothole Upland	42n
Level IV	Rocky Mountain Front Foothill Potholes	42q
Level IV	Foothill Grassland	42r
Level IV	Unglaciated Montana High Plains	43o
Level IV	Non-calcareous Foothill Grassland	43s
Level IV	Shields-Smith Valleys	43t
Level IV	Limy Foothill Grassland	43u
Level IV	Pryor-Bighorn Foothills	43v

Warm Water Fisheries

Table 4.1. Level III Ecoregions in which Nutrient-impairment Assessment Methodologies in this Section Best Apply, along with Specified Level IV Ecoregions for which the Methodologys do not Apply.

Ecoregion Scale			
Level III	Level III	Ecoregion Name	Ecoregion Number
Level III		**Northwestern Glaciated Plains**	**42**
Level IV ecoregions in the Northwestern Glaciated Plains for which the methods do not apply:			
	Level IV	Sweetgrass Uplands	42l
	Level IV	Milk River Pothole Upland	42n
	Level IV	Rocky Mountain Front Foothill Potholes	42q
	Level IV	Foothill Grassland	42r
Level III		**Northwestern Great Plains**	**43**
Level IV ecoregions in the Northwestern Great Plains for which the methods do not apply:			
	Level IV	Unglaciated Montana High Plains	43o
	Level IV	Non-calcareous Foothill Grassland	43s
	Level IV	Shields-Smith Valleys	43t
	Level IV	Limy Foothill Grassland	43u
	Level IV	Pryor-Bighorn Foothills	43v

Appendix B: Final Increaser Taxa Lists

Sediment Increaser Taxa

Warm Water Fisheries - Sediment
Amphora pediculus
Caloneis bacillum
Cocconeis placentula
Diatoma moniliformis
Epithemia sorex
Gomphonema minutum
Navicula capitatoradiata
Navicula cryptotenella
Navicula gregaria
Navicula reichardtiana
Nitzschia inconspicua
Nitzschia liebethruthii
Nitzschia linearis
Reimeria sinuata
Surirella brebissonii kuetzingii

Middle Rockies - Sediment
Amphora inariensis
Cocconeis pediculus
Cocconeis pseudolineata
Eolimna minima
Geissleria acceptata
Gomphonema drutelingense
Meridion circulare
Navicula gregaria
Navicula lanceolata
Navicula tripunctata
Nitzschia recta
Planothidium frequentissimum
Planothidium lanceolatum
Reimeria sinuata
Sellaphora pupula
Staurosirella leptostauron

Northern Rockies - Sediment
Achnanthidium deflexum
Aulacoseira italica
Eolimna minima
Gomphonema minutum
Gomphonema pumilum
Gomphonema rhombicum
Gomphosphenia sp.
Melosira varians
Meridion circulare
Navicula cryptocephala
Navicula radiosa
Nitzschia archibaldii
Nitzschia palea
Nitzschia perminuta
Planothidium frequentissimum
Pseudostaurosira brevistriata
Reimeria sinuata
Rhopalodia gibba
Staurosira construens

Nutrient Increaser Taxa

Warm Water Fisheries - Nutrients
Amphora pediculus
Gomphonema parvulum
Navicula cryptotenella
Navicula libonensis
Navicula tripunctata
Nitzschia acicularis
Nitzschia amphibia
Nitzschia archibaldii
Nitzschia fonticola
Nitzschia gracilis
Nitzschia inconspicua
Nitzschia linearis

Northern Rockies - Nutrients
Achnanthes nodosa
Achnanthidium deflexum
Adlafia minuscula
Eolimna minima
Geissleria acceptata
Gomphonema minutum
Gomphonema pumilum
Gomphosphenia sp.
Meridion circulare
Navicula cryptocephala
Nitzschia fonticola
Nitzschia inconspicua
Nitzschia perminuta
Planothidium frequentissimum
Synedra rumpens

Appendix C: Impairment Probabilities

Sediment Increaser Taxa

Warm Water Fisheries - Sediment	
Percent Relative Abundance	Probability of Impairment
0.00	5%
0.00	10%
0.00	15%
2.14	20%
5.28	25%
8.09	30%
10.70	35%
13.17	40%
15.56	45%
17.92	50%
20.28	55%
22.67	60%
25.14	65%
27.75	70%
30.56	75%
33.70	80%
37.35	85%
41.94	90%
48.75	95%

Middle Rockies - Sediment	
Percent Relative Abundance	Probability of Impairment
0.00	5%
0.00	10%
2.21	15%
4.67	20%
6.79	25%
8.69	30%
10.46	35%
12.13	40%
13.75	45%
15.34	50%
16.93	55%
18.55	60%
20.22	65%
21.99	70%
23.89	75%
26.01	80%
28.47	85%
31.58	90%
36.18	95%

Northern/Canadian Rockies - Sediment	
Percent Relative Abundance	Probability of Impairment
0.00	5%
0.00	10%
0.73	15%
4.07	20%
6.93	25%
9.50	30%
11.88	35%
14.14	40%
16.33	45%
18.48	50%
20.63	55%
22.82	60%
25.08	65%
27.46	70%
30.03	75%
32.89	80%
36.23	85%
40.43	90%
46.65	95%

Nutrient Increaser Taxa

Warm Water Fisheries - Nutrients	
Percent Relative Abundance	Probability of Impairment
0.00	5%
0.00	10%
0.00	15%
0.44	20%
2.58	25%
4.50	30%
6.28	35%
7.97	40%
9.60	45%
11.21	50%
12.82	55%
14.45	60%
16.14	65%
17.92	70%
19.84	75%
21.98	80%
24.48	85%
27.61	90%
32.26	95%

Northern/Canadian Rockies - Nutrients	
Percent Relative Abundance	Probability of Impairment
0.00	5%
0.00	10%
0.00	15%
2.01	20%
4.63	25%
6.98	30%
9.15	35%
11.22	40%
13.21	45%
15.18	50%
17.15	55%
19.14	60%
21.21	65%
23.38	70%
25.73	75%
28.35	80%
31.39	85%
35.23	90%
40.91	95%