BIOLOGICAL INTEGRITY OF THE REDWATER RIVER IN THE VICINITY OF CIRCLE, MONTANA BASED ON THE COMPOSITION AND STRUCTURE OF THE BENTHIC ALGAE COMMUNITY

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SUMMARY

Composite periphyton samples were collected from natural substrates at four sites on the Redwater River near Circle, Montana. Samples were collected following standard operating procedures of the Montana Department of Environmental Quality, processed and analyzed using standard methods for periphyton, and evaluated following modified USEPA rapid bioassessment protocols for wadeable streams.

A large siltation index for a prairie stream and an unusually large number of deformed diatom cells indicated **moderate impairment, fair biological integrity, and partial support of aquatic life uses** at the site (RW-3) immediately below the Circle wastewater lagoons, confirming the assessment made at this site in May 1999. The likely cause of the deformed cells was ammonia.

Several indicators point to increased organic nitrogen loading from above to below the Circle lagoons: (1) a decrease in nitrogen-fixing cyanobacteria; (2) an increase in *Euglena* (a pollution tolerant alga); (3) a decrease in the diatom pollution index; and (4) a decrease in the percentage of diatoms in the family Epithemiaceae. The diatom flora below the Circle lagoons was quite different from the floras at adjacent stations.

Although deformed diatoms were also observed at the other sites on the Redwater River, they accounted for less than half the percentage of deformed cells observed below the Circle lagoons. With decreased flows in summer and decompositon of algae and aquatic macrophytes, some of the abnormalities observed in the Redwater River may have been caused by ammonia generated from internal organic loading. The percentage of deformed cells at three of the four stations (1.56% to 1.73%) may be within the normal range for prairie streams in late summer.

INTRODUCTION

This report evaluates the biological integrity, support of aquatic life uses, and probable causes of impairment to those uses at four stations on the Redwater River near the town of Circle, Montana. The purpose of this report is to provide information that will help the State of Montana and other stakeholders determine whether the Redwater River is water-quality limited and in need of TMDLs.

The federal Clean Water Act directs states to develop water pollution control plans (Total Maximum Daily Loads or TMDLs) that set limits on pollution loading to water-quality limited waters. Water-quality limited waters are lakes and stream segments that do not meet water-quality standards, that is, that do not fully support their beneficial uses. The Clean Water Act and USEPA regulations require each state to (1) identify waters that are water-quality limited, (2) prioritize and target waters for TMDLs, and (3) develop TMDL plans to attain and maintain water-quality standards for all water-quality limited waters.

The evaluations in this report are based on the structure and species composition of the periphyton or phytobenthos community. The periphyton community is a basic biological component of all aquatic ecosystems. Periphyton accounts for much of the primary production and biological diversity of Montana streams (Bahls et al. 1992).

Periphyton is a diverse assortment of simple photosynthetic organisms called algae, and other microorganisms that live attached to or in close proximity of the stream bottom. Many algae, such as the diatoms, are microscopic. Diatoms are distinguished by having a cell wall composed of opaline glass--hydrated amorphous silica. Diatoms often carpet a stream bottom with a slippery brown film.

Some algae, such as the filamentous greens, are conspicuous and their excessive growth may be aesthetically displeasing. Algae may also deplete dissolved oxygen, interfere with fishing and fish spawning, clog water filters and irrigation intakes, create tastes and odors in drinking water, and generate toxins that may be lethal to livestock and other animals.

Plafkin et al. (1989) and Stevenson and Bahls (1999) list several advantages for using periphyton in biological assessments of streams:

- Algae are universally present in large numbers in all streams and unimpaired periphyton assemblages typically support a large number (>30) of species;
- Algae have rapid reproduction rates and short life cycles, making them useful indicators of short-term impacts;
- As primary producers, algae are most directly affected by physical and chemical factors, such as temperature, nutrients, dissolved salts, and toxins;
- Sampling is quick, easy and inexpensive, and causes minimal damage to resident biota and their habitat;
- Standard methods and criteria exist for evaluating the composition, structure, and biomass of algal associations;
- Identification to species is straightforward for the diatoms, for which there is a large body of taxonomic and ecological literature;
- Excessive algae growth in streams is often correctly perceived as a problem by the public.
- Periphyton and other biological communities reflect the *biological integrity*[1] of waterbodies; restoring and maintaining the biological integrity of waterbodies is a goal of the federal Clean Water Act;
- Periphyton and other biological communities integrate the

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[1] *Biological integrity* is defined as "the ability of an aquatic ecosystem to support and maintain a balanced, integrated, adaptive community of organisms having a species composition, diversity, and functional organization comparable to that of natural habitats within a region" (Karr and Dudley 1981).

effects of different stressors and provide a measure of their aggregate impact; and

- Periphyton and other biological communities may be the only practical means of evaluating impacts from non-point sources of pollution where specific ambient criteria do not exist (e.g., impacts that degrade habitat or increase nutrients).

PROJECT AREA AND SAMPLING SITES

The project area is located near the town of Circle in east central Montana. The Redwater River begins in southern McCone County at an elevation of about 3,000 feet and flows northeast for about 80 miles, joining the Missouri River south of Poplar, Montana. Circle (pop. 716) and Brockway (pop. 75) are the only communities on the Redwater River. Brockway is located about 12 miles upstream from Circle.

The watershed of the Redwater River is situated within the Northwestern Glaciated Plains Ecoregion (Woods et al. 1999). The surface geology of the watershed is composed of the Fort Union Formation, a coal-bearing sedimentary deposit of Paleocene age (Renfro and Feray 1972). Upland vegetation is predominantly mixed grassland (USDA 1976). The main land use is livestock grazing with some dryland farming.

Periphyton samples were collected at four sites, two above and two below the Circle wastewater treatment ponds (Map 1, Table 1). Elevations at the sampling sites are about 2,400 feet. The Redwater River is classified C-3 in the Montana Surface Water Quality Standards.

METHODS

Periphyton samples were collected following standard operating procedures of the Planning, Prevention, and Assistance Division of the Montana Department of Environmental Quality. Using appropriate tools, microalgae were scraped, brushed, and/or sucked from natural substrates in proportion to the rank of those substrates at the study site. Macroalgae were picked by hand in proportion to their abundance at the site. All collections of microalgae and macroalgae were pooled into a common container and preserved with Lugol's solution (APHA 1998).

Samples were examined to estimate the relative abundance of cells and rank by biovolume of diatoms and genera of soft (non-diatom) algae according to the method described in Bahls (1993). Soft algae were identified using Dillard (1999), Prescott (1978), Smith (1950), and Whitford and Schumacher (1984). These books also served as references on the ecology of the soft algae, along with Palmer (1977).

After the identification of soft algae, raw periphyton samples were cleaned of organic matter using sulfuric acid, and permanent diatom slides were prepared using Naphrax, a high refractive index mounting medium, following *Standard Methods for the Examination of Water and Wastewater* (APHA 1998). For each slide, between 405 and 453 diatom cells (810 to 906 valves) were counted at random and identified to species. The following were used as the main taxonomic and autecological references for the diatoms: Krammer and Lange-Bertalot 1986, 1988, 1991a, 1991b; Patrick and Reimer 1966, 1975.

Lowe (1974) was also used as an ecological reference for the diatoms. Bahls et al. (1984) provide autecological information on important diatom species that live in the Fort Union Region of Montana, including many of the diatom species found in the Redwater River.

The diatom proportional counts were used to generate an array of diatom association metrics (Table 2). A metric is a characteristic of the biota that changes in some predictable way with increased human influence (Barbour et al. 1999).

Metric values for the Redwater River were compared to numeric biocriteria developed for streams in the Great Plains Ecoregions of Montana (Table 3). These criteria are based on metric values measured in least-impaired reference streams (Bahls et al. 1992) and on metric values measured in streams that are known to be impaired by various sources and causes of pollution (Bahls 1993).

The criteria in Table 3 distinguish among four levels of impairment and three levels of aquatic life use support: no impairment or only minor impairment (full support); moderate impairment (partial support); and severe impairment (nonsupport). These impairment levels correspond to excellent, good, fair, and poor *biological integrity*, respectively.

QUALTIY ASSURANCE

Several steps were taken to assure that the study results are accurate and reproducible.

Upon receipt of the samples, station and sample information were recorded in a laboratory notebook and samples were assigned a unique number compatible with the Montana Diatom Database, e.g., 2008-01. The first part of this number (2008) designates the sampling site (Redwater River upstream of the McCone County Fairgrounds); the second part of the number (01) designates the number of periphyton samples that have been collected at this site to date for which data have been entered into the Montana Diatom Database.

Sample observations and analyses of soft (non-diatom) algae were recorded in a lab notebook. A portion of the raw sample was used to make duplicate diatom slides.

On completion of the project, station information, sample information, and diatom proportional count data will be entered into the Montana Diatom Database. One set of diatom slides will be deposited in the University of Montana Herbarium in Missoula. The other set of slides will be retained by *Hannaea* in Helena.

RESULTS AND DISCUSSION

Results are presented in Tables 4 and 5, located near the end of this report following the Literature Cited section. Spreadsheets containing completed diatom proportional counts, with species' pollution tolerance classes and percent abundances, are attached as Appendix A.

SAMPLE NOTES

Redwater River above Fairgrounds (RW-1). Trichomes of *Oscillatoria* in this sample did not have a visible sheath, but they did form clumps several millimeters wide. The trichomes were red in color and very thick. In mats, the trichomes were oriented parallel to one another, or they were interwoven. The *Lyngbya* in this sample had discoidal cells and a definite sheath; no branching was observed.

Redwater River below Circle lagoons (RW-3). At least two species of *Anabaena* were observed in this sample, one with straight trichomes and the other with coiled trichomes.

NON-DIATOM ALGAE

The Redwater River near Circle supported mostly a mix of green algae, diatoms, and cyanobacteria (primitive algae of the Phylum Cyanophyta, formerly called blue-green algae) (Table 4). Diatoms were the most abundant algae at all sites except above the Fairgrounds (RW-1). Each site supported from 10 to 12 genera of non-diatom algae, which is about average for prairie streams in Montana (Bahls 1993).

Cyanobacteria, which are capable of fixing atmospheric or molecular nitrogen, dominated the sample collected above the McCone Co. Fairgrounds, where they ranked first in biovolume (Table 4). Cyanobacteria ranked second in biovolume above the Circle lagoons, but dropped to fourth below the lagoons. This would indicate some nitrogen enrichment in the reach between RW-2 and RW-3. Diatoms and green algae have a growth advantage over cyanobacteria when supplemental sources of nitrogen are provided. Cyanobacteria resumed a rank of two below Horse Creek.

An occasional cell of *Euglena* at RW-3 also indicates moderate nutrient enrichment and organic loading below the Circle lagoons. These cells may have originated within the lagoons, as they are common in such habitats (Palmer 1977). *Euglena*, a good indicator of organic loading, was absent at the two upstream sites and rare at the downstream site below Horse Creek (RW-5).

Anabaena was one of several nitrogen-fixing cyanobacteria in the Redwater River. Under certain conditions, *Anabaena* can produce waterblooms that release neurotoxins into the water. These toxins can be lethal to livestock, pets, and wildlife. However, *Anabaena* was not abundant enough in the Redwater River to pose a problem for livestock producers.

DIATOMS

The major diatom species in the Redwater River were either sensitive to organic loading (Pollution Tolerance Class 3) or somewhat tolerant of organic loading (PTC 2). The sensitive species declined in abundance from above to below the Circle lagoons, resulting in a lower pollution index. However, the decline was not dramatic, indicating that discharges from the lagoons did not have a major effect on water quality (Table 5).

With few exceptions, diatom association metrics indicated good to excellent biological integrity at all sites (Table 5). The siltation index, which is based on the percentage of motile diatoms in the diatom association, indicated moderate impairment and partial support of aquatic life uses at RW-3 below the Circle lagoons. The increase in motile diatoms here was due mainly to a preponderance of *Nitzschia* species. Species of *Nitzschia* are generally tolerant of organic loading.

An unusually large number of teratological cells were observed at all sites, resulting in ratings of fair biological integrity and partial support of aquatic life uses (Table 5). Teratological cells were observed in diatom species that are sensitive to pollution (*Achnanthes minutissima, Cocconeis placentula, Fragilaria brevistriata*), somewhat tolerant of pollution (*Mastogloia smithii, Nitzschia frustulum, Nitzschia inconspicua, Nitzschia paleacea, Nitzschia valdestriata, Synedra fasciculata*), and very tolerant of pollution (*Navicula duerrenbergiana*).

The percentage of teratological cells more than doubled at RW-3 below the Circle lagoons. *Nitzschia inconspicua* was the dominant diatom at RW-3 and contributed eighty percent of the teratological cells at this site. *Nitzschia inconspicua* is characteristic of the mesosaprobic zone, where oxidation of the organic load is proceeding and where nitrogen is present as ammonia (Krammer and Lange-Bertalot 1988). *Nitzschia inconspicua* (misidentified as *N. epiphytica*) was the dominant diatom species in the East Gallatin River immediately below the old Bozeman wastewater treatment plant in the late 1960s (Bahls 1973).

It is unlikely that the *Nitzschia inconspicua* cells found in the Redwater River, including the teratological cells of this species, originated in the Circle lagoons. This species requires oxygenated waters where the breakdown of organic nitrogen is nearly complete. Unlike *Nitzschia palea* and other species of *Nitzschia* that thrive under anaerobic conditions and heavy organic loading, *Nitzschia inconspicua* has not been reported from sewage treatment ponds (Palmer 1969).

Moreover, the Circle lagoons are flushed in the spring, well before the samples for the present study were collected (in August). Any algae released in the flush would have been carried downstream or decomposed. Diatoms that might have been living in the Circle lagoons would not survive for long in the Redwater River, at least not in large numbers. In alkaline waters such as the Redwater River, dead (empty) diatom frustules likely would not persist for more than a few weeks.

In May of 1999, a large number of teratological cells of *Diatoma tenue* resulted in a similar rating of partial support in the Redwater River below the Circle lagoons (Bahls 1999). At that time, *Diatoma tenue* was the dominant diatom species both above and below the Circle wastewater effluent. No teratological cells of this species were observed above the wastewater lagoons. *Diatoma tenue* is a cool season diatom (Bahls et al. 1984); it tolerates about the same amount of organic loading as *Nitzschia inconspicua* (Lowe 1974); and it has not been reported from sewage stabilization ponds (Palmer 1977).

A number of factors are known to cause abnormalities in diatom cells, including heavy metals (McFarland et al. 1997). Salinity and ammonia are other possible causes. Salinity is not the likely cause of teratological cells in the Redwater River because most of the major diatom species that are present (Table 5) indicate fresh to somewhat brackish water. Heavy metals are also an unlikely cause. Deformities caused by heavy metals have been reported only from poorly buffered circumneutral waters (McFarland et al. 1997).

Although abnormal cells were observed at all stations in this study, they were more than twice as common below the Circle lagoons than at any other site. In May of 1999, problematic numbers of abnormal cells were observed *only* below the Circle lagoons. In August 2000, the percentage of deformed cells below the Circle lagoons (3.97%) exceeded the percentage observed at other sites on the Redwater River by a ratio of two to one.

In summer, with decreased flows, higher temperatures, and decompositon of algae and aquatic macrophytes, a certain amount of internal organic loading may occur. Some of the abnormalities observed in August may have been caused by ammonia generated from this internal organic loading. Thus, the percentage of deformed cells at three of the four stations (1.56% to 1.73%) may be within the normal range for prairie streams in summer.

Two additional diatom metrics show a significant change in water quality below the Circle lagoons. The minimum value for the percentage of diatoms in the family Epithemiaceae indicates nitrogen enrichment at RW-3. Diatoms in this family typically harbor nitrogen-fixing cyanobacteria within their cells and are adapted to living in waters that are nitrogen-poor. Similarity index values between adjacent stations in this reach of the Redwater River indicate that the greatest change in diatom species composition, hence water quality, occurred between sites

RW-2 and RW-3, which had only 30% of their diatom floras in common (Table 5.)

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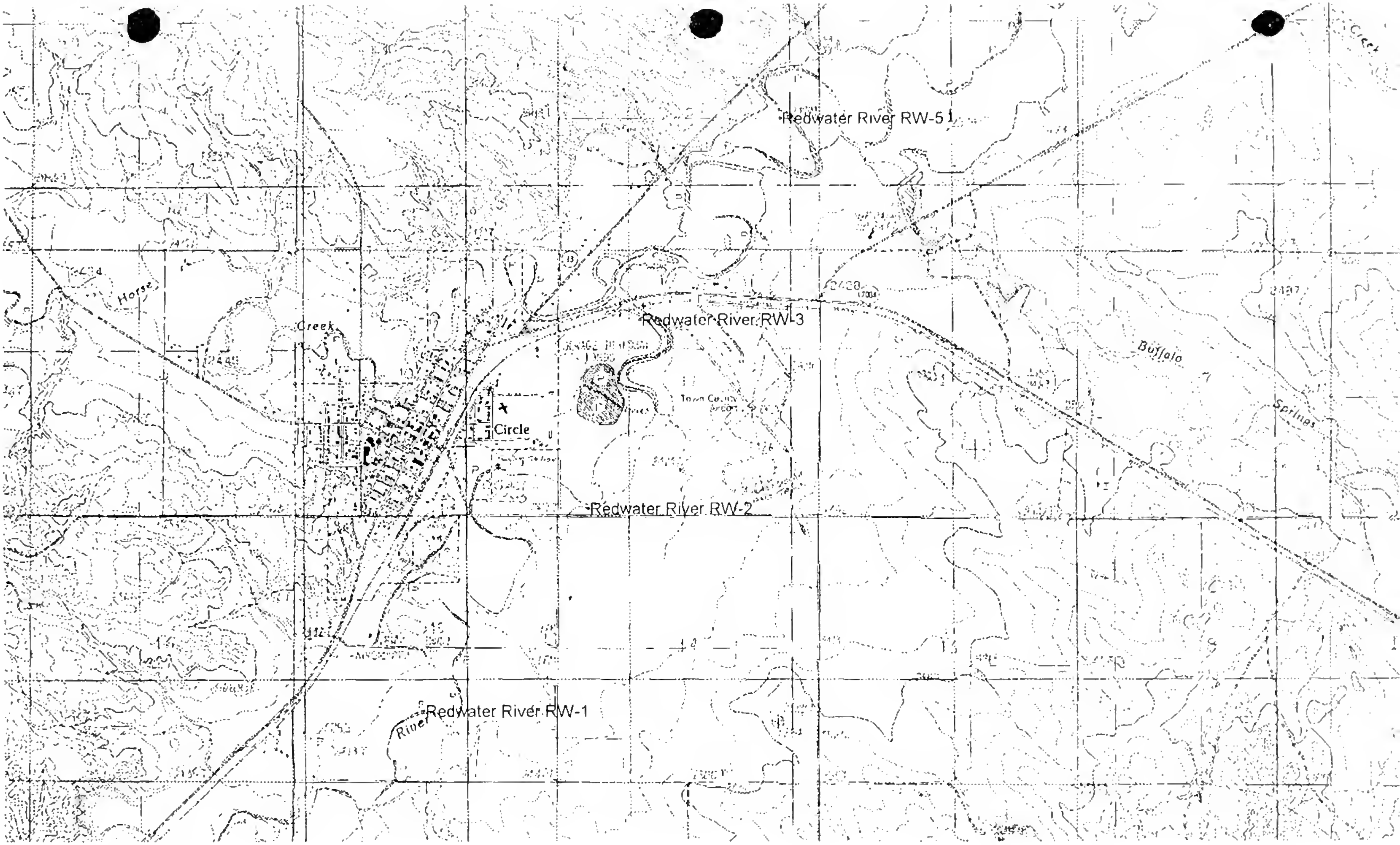

Map 1.

'Circle; MT'; Scale: 1" = 0.511Mi 822Mt 2,696Ft, 1 Mi = 1.958" , 1 cm = 323Mt

Table 1. Location of periphyton stations on the Redwater River sampled August 17, 2000: Station codes, sample numbers in the Montana Diatom Database, and latitudes and longitudes. Stations are listed in order from upstream to downstream.

Location	Station Code	Sample Number	Latitude/ Longitude
Redwater River upstream of McCone Co. Fairgrounds	RW-1	2008-01	47 24 08 N 105 35 12 W
Redwater River upstream of Circle STP lagoons	RW-2	2009-01	47 24 49 N 105 34 24 W
Redwater River downstream of Circle STP lagoons	RW-3	2010-01	47 25 25 N 105 34 09 W
Redwater River downstream of Horse Creek	RW-5	2011-01	47 26 05 N 105 33 28 W

Table 2. Diatom association metrics used to evaluate biological integrity in Montana streams: reference, range of values in Montana streams, and expected direction of metric response to increasing anthropogenic perturbation or natural stress.

Metric	Reference	Range of Values	Expected Response
Shannon Species Diversity	Bahls 1979	0.00-5.00+	Decrease[1]
Pollution Index[2]	Bahls 1993	1.00-3.00	Decrease
Siltation Index[3]	Bahls 1993	0.00-90.0+	Increase
Disturbance Index[4]	Barbour *et al.* 1999	0.00-100.0	Increase
No. Species Counted	Bahls 1979, 1993	0-100+	Decrease[1]
Percent Dominant Species	Barbour *et al.* 1999	5.0-100.0	Increase
Percent Abnormal Cells	McFarland *et al.* 1997	0.0-20.0+	Increase
Similarity Index	Whittaker 1952	0.0-80.0+	Decrease
Percent Epithemiaceae	Stevenson & Pan 1999	0.0-80.0+	Decrease
Percent Aerophiles	Johansen 1999	0.0-100	Increase

[1] Shannon diversity and species richness may increase somewhat in naturally nutrient-poor mountain streams in response to slight to moderate increases in nutrients or sediment.

[2] Composite numeric expression of the pollution tolerances assigned by Lange-Bertalot (1979) to the common diatom species.

[3] Sum of the percent abundances of all species in the genera *Navicula*, *Nitzschia*, and *Surirella*, plus the species *Cymbella sinuata*.

[4] Percent abundance of *Achnanthes minutissima*.

Table 3. Criteria for rating levels of biological integrity, environmental impairment or natural stress, and aquatic life use support in wadeable **plains** streams of Montana using selected metrics for benthic diatom associations. The lowest rating for any one metric is the overall rating for the study site.

Biological Integrity/ Impairment or Natural Stress/Use Support	Diversity Index (Shannon)	Pollution Index	Siltation Index	Disturbance Index	Number of Species Counted	Percent Dominant Species	Percent Abnormal Cells	Similarity Index[1]
Excellent None/Full Support	>3.99	>2.25	<50.0	<25.0	>39	<25.0	0.0	>59.9
Good/Minor Full Support	3.00-3.99	1.76-2.25	50.0-69.9	25.0-49.9	30-39	25.0 49.9	>0.0-<1.0	40.0-59.9
Fair/Moderate Partial Support	2.00-2.99	1.25-1.75	70.0-89.9	50.0-74.9	20-29	50.0-74.9	1.0-9.9	20.0-39.9
Poor/Severe Nonsupport	<2.00	<1.25	>89.9	>74.9	<20	>74.9	>9.9	<20.0

[1] The Similarity Index or Percent Community Similarity (Whittaker 1952) may be used to compare a study site to an unimpaired upstream control site on the same stream. This metric measures the degree of floristic similarity between diatom associations at the two sites and is the sum of the smaller of the two percent abundance values for each species that is common to both sites. Adjacent riffles on the same stream, without intervening tributaries or environmental perturbations, will generally have at least 60% of their diatom floras in common (Bahls 1993). PCS may also be used to guage the relative amount of impairment or recovery that occurs between adjacent study sites: >59.9% = very similar floras, no change; 40.0-59.9% = somewhat similar floras, minor change; 20.0-39.9% = somewhat dissimilar floras, moderate change; <20.0% = very dissimilar floras, major change.

Table 4. Relative abundance of cells and rank by biovolume of diatoms and genera of non-diatom algae in periphyton samples collected from the Redwater River: d = dominant; a = abundant; f = frequent; c = common; o = occasional; r = rare.

Taxa	Relative Abundance and Rank (in parentheses)			
	RW-1	RW-2	RW-3	RW-5
Chlorophyta				
Ankistrodesmus		c (9)	c (11)	
Bulbochaete		c (6)		
Cosmarium				r (12)
Hormidium				o (8)
Mougeotia		o (10)		
Oedogonium	o (4)	f (3)	o (2)	f (3)
Protoderma			o (13)	
Rhizoclonium	o (5)	o (8)	o (3)	o (5)
Scenedesmus	o (10)	o (11)	o (10)	o (9)
Spirogyra				c (4)
Euglenophyta				
Euglena			o (9)	r (11)
Chrysophyta				
Diatoms	a (2)	a (1)	a (1)	a (1)
Pyrrophyta				
Glenodinium		r (14)		
Cyanophyta				
Anabaena	o (11)	o (13)	o (8)	o (10)
Calothrix	o (9)	c (7)	o (6)	f (7)
Gloeothece		c (5)		
Lyngbya	o (6)			
Merismopedia		o (12)		
Oscillatoria	d (1)		o (7)	
Phormidium	o (7)	f (4)	c (4)	f (6)
Rivularia	f (3)	f (2)	o (5)	a (2)
Spirulina	c (8)		c (12)	

Table 5. Percent abundance of major diatom species[1] and values of selected diatom association metrics for periphyton samples collected from the Redwater River on August 17, 2000.

Species/Metric (Pollution Tolerance Class)[3]	Percent Abundance/Metric Values[2]			
	RW-1	RW-2	RW-3	RW-5
Amphora pediculus (3)	13.46	23.78	14.35	13.96
Cymbella hustedtii (3)	0.12	14.00	0.11	
Fragilaria brevistriata (3)	14.32	3.00	0.22	0.34
Nitzschia frustulum (2)	0.99	0.89	18.65	3.89
Nitzschia inconspicua (2)		0.22	25.94	5.84
Nitzschia reversa (2)	2.96	1.67	14.35	14.65
Nitzschia valdestriata (2)	15.68	19.89	2.10	5.95
Cells Counted	405	450	453	437
Total Species	72	46	49	63
Species Counted	68	41	41	55
Disturbance Index	0.86	3.78	1.10	0.57
Percent Dominant Species	15.68	23.78	<u>25.94</u>	14.65
Species Diversity Index	4.67	<u>3.68</u>	<u>3.43</u>	4.69
Pollution Index	<u>2.18</u>	2.35	<u>2.07</u>	<u>2.04</u>
Siltation Index	40.25	33.00	**79.14**	<u>67.96</u>
Percent Abnormal Valves	**1.73**	**1.56**	**3.97**	**1.58**
Percent Epithemiaceae	2.84	3.00	0.88	2.52
Similarity Index		52.22	30.08	55.54

[1] A major diatom species is here considered to be one that accounts for 10% or more of the cells in one or more samples of a sample set.

[2] <u>Underlined values</u> indicate good biological integrity, minor impairment, and full support of aquatic life uses; **bold values** indicate fair biological integrity, moderate impairment, and partial support of aquatic life uses; all other values indicate excellent biological integrity, no impairment, and full support of aquatic life uses when compared to diatom criteria for plains streams in Table 3.

[3] 3 = sensitive to pollution; 2 = tolerant of pollution; 1 = most tolerant of pollution.

APPENDIX A: DIATOM PROPORTIONAL COUNTS

Sample	Genus/Species/Variety	Pollution Tolerance Class	Count	Percent
200801	Achnanthes delicatula	2	9	1.11
200801	Achnanthes lanceolata	2	1	0.12
200801	Achnanthes minutissima	3	7	0.86
200801	Amphipleura pellucida	2	2	0.25
200801	Amphora coffeaeformis	1	7	0.86
200801	Amphora inariensis	3	1	0.12
200801	Amphora libyca	3	5	0.62
200801	Amphora pediculus	3	109	13.46
200801	Amphora veneta	1	7	0.86
200801	Caloneis bacillum	2	3	0.37
200801	Caloneis sp.	3	2	0.25
200801	Caloneis tenuis	3	8	0.99
200801	Cocconeis neothumensis	3	12	1.48
200801	Cyclotella meneghiniana	2	3	0.37
200801	Cymbella hustedtii	3	1	0.12
200801	Cymbella muelleri	2	2	0.25
200801	Cymbella pusilla	1	34	4.20
200801	Denticula tenuis	3	11	1.36
200801	Diploneis puella	2	3	0.37
200801	Entomoneis alata	2	1	0.12
200801	Entomoneis paludosa	2	1	0.12
200801	Epithemia adnata	2	1	0.12
200801	Epithemia argus	2	4	0.49
200801	Fragilaria brevistriata	3	116	14.32
200801	Fragilaria construens	3	17	2.10
200801	Fragilaria elliptica	2	48	5.93
200801	Hantzschia amphioxys	2	1	0.12
200801	Mastogloia elliptica	2	8	0.99
200801	Mastogloia grevillei	2	9	1.11
200801	Mastogloia smithii	2	19	2.35
200801	Navicula arvensis	1	2	0.25
200801	Navicula canalis	1	2	0.25
200801	Navicula capitata	2	3	0.37
200801	Navicula caterva	2	9	1.11
200801	Navicula cincta	1	3	0.37
200801	Navicula circumtexta	1	15	1.85
200801	Navicula cryptotenella	2	1	0.12
200801	Navicula duerrenbergiana	1	21	2.59
200801	Navicula erifuga	2	2	0.25
200801	Navicula goersii	2	4	0.49
200801	Navicula gregaria	2	13	1.60
200801	Navicula halophila	2	1	0.12
200801	Navicula peregrina	2	3	0.37
200801	Navicula phyllepta	2	1	0.12
200801	Navicula pygmaea	2	2	0.25
200801	Navicula salinarum	1	11	1.36
200801	Navicula subminuscula	1	0	0.00

Sample	Genus/Species/Variety	Pollution Tolerance Class	Count	Percent
200801	Navicula tenera	1	3	0.37
200801	Navicula veneta	1	18	2.22
200801	Nitzschia amphibia	2	5	0.62
200801	Nitzschia angustatula	2	1	0.12
200801	Nitzschia apiculata	2	3	0.37
200801	Nitzschia calida	2	0	0.00
200801	Nitzschia filiformis	2	8	0.99
200801	Nitzschia frustulum	2	8	0.99
200801	Nitzschia hungarica	2	0	0.00
200801	Nitzschia incognita	2	2	0.25
200801	Nitzschia liebetruthii	2	6	0.74
200801	Nitzschia microcephala	1	9	1.11
200801	Nitzschia palea	1	12	1.48
200801	Nitzschia reversa	2	24	2.96
200801	Nitzschia supralitorea	2	5	0.62
200801	Nitzschia valdecostata	2	2	0.25
200801	Nitzschia valdestriata	2	127	15.68
200801	Pinnularia krookii	2	3	0.37
200801	Pinnularia viridis	3	2	0.25
200801	Pleurosigma delicatulum	2	17	2.10
200801	Rhoicosphenia curvata	3	2	0.25
200801	Rhopalodia brebissonii	1	0	0.00
200801	Rhopalodia gibba	2	2	0.25
200801	Rhopalodia operculata	1	5	0.62
200801	Synedra fasciculata	2	1	0.12

Sample	Genus/Species/Variety	Pollution Tolerance Class	Count	Percent
200901	Achnanthes delicatula	2	3	0.33
200901	Achnanthes minutissima	3	34	3.78
200901	Achnanthes taeniata	3	4	0.44
200901	Amphora coffeaeformis	1	3	0.33
200901	Amphora delicatissima	1	2	0.22
200901	Amphora inariensis	3	3	0.33
200901	Amphora libyca	3	0	0.00
200901	Amphora pediculus	3	214	23.78
200901	Amphora veneta	1	0	0.00
200901	Caloneis tenuis	3	1	0.11
200901	Chaetoceros muelleri	1	1	0.11
200901	Cocconeis placentula	3	1	0.11
200901	Cyclotella meneghiniana	2	5	0.56
200901	Cymbella hustedtii	3	126	14.00
200901	Cymbella pusilla	1	20	2.22
200901	Cymbella silesiaca	2	1	0.11
200901	Diploneis puella	2	10	1.11
200901	Epithemia argus	2	20	2.22
200901	Fragilaria brevistriata	3	27	3.00
200901	Fragilaria construens v. venter	3	2	0.22
200901	Fragilaria elliptica	2	9	1.00
200901	Mastogloia elliptica	2	6	0.67
200901	Mastogloia smithii	2	33	3.67
200901	Navicula capitata	2	1	0.11
200901	Navicula circumtexta	1	1	0.11
200901	Navicula cryptotenella	2	0	0.00
200901	Navicula duerrenbergiana	1	7	0.78
200901	Navicula goersii	2	1	0.11
200901	Navicula recens	2	1	0.11
200901	Navicula tenera	1	0	0.00
200901	Navicula veneta	1	8	0.89
200901	Nitzschia acicularis	2	2	0.22
200901	Nitzschia amphibia	2	19	2.11
200901	Nitzschia filiformis	2	3	0.33
200901	Nitzschia frustulum	2	8	0.89
200901	Nitzschia incognita	2	1	0.11
200901	Nitzschia inconspicua	2	2	0.22
200901	Nitzschia liebetruthii	2	0	0.00
200901	Nitzschia microcephala	1	25	2.78
200901	Nitzschia palea	1	23	2.56
200901	Nitzschia reversa	2	15	1.67
200901	Nitzschia valdecostata	2	1	0.11
200901	Nitzschia valdestriata	2	179	19.89
200901	Rhopalodia gibba	2	3	0.33
200901	Rhopalodia operculata	1	4	0.44
200901	Synedra fasciculata	2	71	7.89

Sample	Genus/Species/Variety	Pollution Tolerance Class	Count	Percent
201001	Achnanthes delicatula	2	1	0.11
201001	Achnanthes minutissima	3	10	1.10
201001	Amphora inariensis	3	5	0.55
201001	Amphora libyca	3	2	0.22
201001	Amphora pediculus	3	130	14.35
201001	Cyclotella meneghiniana	2	8	0.88
201001	Cymbella hustedtii	3	1	0.11
201001	Cymbella pusilla	1	1	0.11
201001	Cymbella silesiaca	2	2	0.22
201001	Diploneis puella	2	4	0.44
201001	Entomoneis alata	2	2	0.22
201001	Entomoneis paludosa	2	0	0.00
201001	Epithemia turgida	3	0	0.00
201001	Fragilaria brevistriata	3	2	0.22
201001	Fragilaria elliptica	2	7	0.77
201001	Mastogloia elliptica	2	0	0.00
201001	Mastogloia smithii	2	3	0.33
201001	Navicula capitata	2	15	1.66
201001	Navicula caterva	2	6	0.66
201001	Navicula cincta	1	1	0.11
201001	Navicula circumtexta	1	3	0.33
201001	Navicula duerrenbergiana	1	0	0.00
201001	Navicula gregaria	2	0	0.00
201001	Navicula novaesiberica	2	4	0.44
201001	Navicula peregrina	2	1	0.11
201001	Navicula recens	2	1	0.11
201001	Navicula salinarum	1	0	0.00
201001	Navicula veneta	1	25	2.76
201001	Nitzschia agnita	1	6	0.66
201001	Nitzschia amphibia	2	6	0.66
201001	Nitzschia apiculata	2	0	0.00
201001	Nitzschia calida	2	2	0.22
201001	Nitzschia filiformis	2	3	0.33
201001	Nitzschia frustulum	2	169	18.65
201001	Nitzschia gracilis	2	1	0.11
201001	Nitzschia incognita	2	4	0.44
201001	Nitzschia inconspicua	2	235	25.94
201001	Nitzschia leistikowii	2	45	4.97
201001	Nitzschia liebetruthii	2	5	0.55
201001	Nitzschia microcephala	1	7	0.77
201001	Nitzschia palea	1	27	2.98
201001	Nitzschia paleacea	2	2	0.22
201001	Nitzschia reversa	2	130	14.35
201001	Nitzschia solita	1	0	0.00
201001	Nitzschia valdestriata	2	19	2.10
201001	Rhoicosphenia curvata	3	1	0.11
201001	Rhopalodia gibba	2	5	0.55
201001	Rhopalodia operculata	1	3	0.33
201001	Synedra fasciculata	2	2	0.22

Sample	Genus/Species/Variety	Pollution Tolerance Class	Count	Percent
201101	Achnanthes delicatula	2	3	0.34
201101	Achnanthes hauckiana	2	0	0.00
201101	Achnanthes minutissima	3	5	0.57
201101	Achnanthes taeniata	3	0	0.00
201101	Amphora coffeaeformis	1	2	0.23
201101	Amphora libyca	3	3	0.34
201101	Amphora pediculus	3	122	13.96
201101	Bacillaria paradoxa	2	11	1.26
201101	Caloneis bacillum	2	21	2.40
201101	Cocconeis placentula	3	1	0.11
201101	Cyclotella atomus	2	17	1.95
201101	Cyclotella meneghiniana	2	24	2.75
201101	Cymbella pusilla	1	3	0.34
201101	Denticula subtilis	2	2	0.23
201101	Diploneis puella	2	2	0.23
201101	Entomoneis alata	2	0	0.00
201101	Epithemia adnata	2	0	0.00
201101	Epithemia argus	2	1	0.11
201101	Epithemia sorex	3	2	0.23
201101	Epithemia turgida	3	0	0.00
201101	Fragilaria brevistriata	3	3	0.34
201101	Fragilaria elliptica	2	12	1.37
201101	Hantzschia amphioxys	2	2	0.23
201101	Mastogloia elliptica	2	9	1.03
201101	Mastogloia smithii	2	10	1.14
201101	Navicula angusta	2	1	0.11
201101	Navicula capitata	2	18	2.06
201101	Navicula caterva	2	5	0.57
201101	Navicula circumtexta	1	5	0.57
201101	Navicula erifuga	2	2	0.23
201101	Navicula minima	1	6	0.69
201101	Navicula novaesiberica	2	6	0.69
201101	Navicula phyllepta	2	1	0.11
201101	Navicula pygmaea	2	3	0.34
201101	Navicula recens	2	26	2.97
201101	Navicula salinarum	1	8	0.92
201101	Navicula tenelloides	1	6	0.69
201101	Navicula tenera	1	4	0.46
201101	Navicula veneta	1	34	3.89
201101	Nitzschia agnita	1	6	0.69
201101	Nitzschia amphibia	2	29	3.32
201101	Nitzschia apiculata	2	1	0.11
201101	Nitzschia calida	2	1	0.11
201101	Nitzschia filiformis	2	43	4.92
201101	Nitzschia frustulum	2	34	3.89
201101	Nitzschia hungarica	2	5	0.57
201101	Nitzschia incognita	2	16	1.83

Sample	Genus/Species/Variety	Pollution Tolerance Class	Count	Percent
201101	Nitzschia inconspicua	2	51	5.84
201101	Nitzschia leistikowii	2	10	1.14
201101	Nitzschia liebetruthii	2	24	2.75
201101	Nitzschia microcephala	1	0	0.00
201101	Nitzschia palea	1	18	2.06
201101	Nitzschia paleacea	2	43	4.92
201101	Nitzschia reversa	2	128	14.65
201101	Nitzschia siliqua	2	0	0.00
201101	Nitzschia solita	1	3	0.34
201101	Nitzschia valdecostata	2	5	0.57
201101	Nitzschia valdestriata	2	52	5.95
201101	Pinnularia appendiculata	3	7	0.80
201101	Rhopalodia gibba	2	6	0.69
201101	Rhopalodia operculata	1	11	1.26
201101	Synedra fasciculata	2	1	0.11
201101	Synedra pulchella	2	0	0.00