

SUPPORT OF AQUATIC LIFE USES
IN BIG SPRING CREEK, FERGUS COUNTY, MONTANA
BASED ON THE COMPOSITION AND STRUCTURE
OF THE BENTHIC ALGAE COMMUNITY

Prepared for:

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SUMMARY

In May and August 2001, periphyton samples were collected at six stations on Big Spring Creek near Lewistown, Montana for the purpose of assessing whether the creek is water-quality limited and in need of TMDLs. The samples were collected following DEQ standard operating procedures, processed and analyzed using standard methods for periphyton, and evaluated following modified USEPA rapid bioassessment protocols for wadeable streams.

Results from the samples collected in May could not be compared to biological criteria Montana streams because these samples were collected outside of the summer sampling "window". The sample collected below Lewistown in May contained an unusually large number of motile, mesotrophic to eutrophic diatoms that may indicate siltation and nutrient problems.

In August, the site just below Big Springs supported a cold-water flora that is typical of a spring creek. The moderate impairment noted here is caused by natural thermal stress.

Diatom metrics for site 02 above Lewistown indicated minor impairment from disturbance and possible low levels of toxic chemicals. Aquatic life uses were fully supported. Site 03 below Lewistown also provided full support of aquatic life uses, with minor impairment indicated from siltation and toxics.

Diatom metrics for site 05 near the mouth of Big Spring Creek indicated **moderate impairment and partial support of aquatic life uses**. The probable cause of this impairment is nutrient enrichment. The bioassessment ratings for sites 03 and 05 would be the same if diatom metrics for these sites were compared to criteria for prairie streams instead of mountain streams.

INTRODUCTION

This report evaluates the biological integrity, support of aquatic life uses, and probable causes of impairment to those uses, in Big Spring Creek near Lewistown, Montana. The purpose of this report is to provide information that will help the State of Montana determine whether Big Spring Creek 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.

Evaluation of use support in this report is based on the species composition and structure of the periphyton (benthic algae, phytobenthos) community at six stream sites that were sampled in May and August, 2001. The periphyton community is a basic biological component of all aquatic ecosystems. Periphyton accounts for much of the primary production and biological diversity in Montana streams (Bahls et al. 1992).

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

- 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 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).

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. Most 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.

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).

Some algae, such as the filamentous greens, are conspicuous and their excessive growth may be aesthetically displeasing, deplete dissolved oxygen, interfere with fishing and fish spawning, clog water filters and irrigation intakes, create tastes and odors in drinking water, and cause other problems.

PROJECT AREA AND SAMPLING SITES

The project area is located in Fergus County near the city of Lewistown (pop. 6,368) in central Montana. The Big Springs, seven miles southeast of Lewistown, generate most of the streamflow in Big Spring Creek. From Big Springs, Big Spring Creek flows northwesterly for about 30 miles through Lewistown and on to its confluence with the Judith River.

Major tributaries of Big Spring Creek, including aquifers feeding the Big Springs, head in the Big Snowy Mountains (maximum elevation 8,730 feet), an outlier of the Northern Rockies Ecoregion (Woods et al. 1999). Although the segment of Big Spring Creek addressed here is located in the Montana Valley and Foothill Prairies Ecoregion, water quality is dominated by the Big Springs. In the Montana Surface Water Quality Standards, the stream is classified B-1 above Lewistown and B-2 below Lewistown.

The surface geology of the Big Spring Creek watershed consists of Big Snowy dolomite and limestone in the headwaters, metamorphic rocks of the Kootenai Formation in the middle reach, and shales of the Colorado Group in the lower reach (Renfro and Feray 1972). Vegetation is alpine tundra and spruce-fir forest in the headwaters, mixed forest and grassland in the middle reach, and mixed grassland at lower elevations (USDA 1976).

Periphyton samples were collected at 2 sites on May 25 and at another 4 sites on August 20 and 22, 2001. The 2 sites

sampled on May 25 bracket the City of Lewistown (Map 1, Table 1). The four sites sampled in August span the reach from just below Big Springs to near the mouth of the stream (Maps 2 and 3, Table 1). Elevations at the sampling sites range from 4250 feet below Big Springs to 3475 feet near the mouth of the creek.

Recreation, fish and aquatic life uses in upper Big Spring Creek are threatened by land development and discharges from a fish hatchery (MDEQ 1998). Below Lewistown, aquatic life, fish and recreational uses are partially impaired by agriculture, channelization, on-site domestic wastewater, the outfall from the Lewistown wastewater treatment plant, stormwater runoff, animal confinement facilities, and silviculture (MDEQ 1998).

Three previous reports on Big Spring Creek and tributaries have been prepared for MDEQ by this consultant (Bahls 1999a, 1999b, and 2001).

METHODS

Periphyton samples were collected in May by Tom Pick, NRCS/DNRC, and in August by Rebecca Ridenour, MDEQ Monitoring and Data Management Bureau, following standard operating procedures of the MDEQ Planning, Prevention, and Assistance Division.

Using appropriate tools, microalgae were scraped, brushed, 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.

The samples were examined to estimate the relative abundance and rank by biovolume of diatoms and genera of soft (non-diatom)

algae according to the method described by 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 (1969, 1977).

After the identification of soft algae, the raw periphyton samples were cleaned of organic matter using sulfuric acid and potassium dichromate, 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).

Between 410 and 458 diatom cells (820 to 916 valves) were counted at random and identified to species. The four volume series by Krammer and Lange-Bertalot (1986, 1988, 1991a, 1991b) was used as the main taxonomic and autecological reference for the diatoms. Lowe (1974), Bahls et al. (1984), Van Dam et al. (1994), and Lange-Bertalot (1996) were also used as ecological references for the diatoms.

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 generated from the August samples were compared to numeric biocriteria or threshold values developed for streams in the Rocky Mountain Ecoregion 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 biocriteria in Table 3 do not apply to the May samples, which were collected outside of the summer sampling "window".

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.

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

Replicate periphyton samples were collected from site 03. Both samples from this site were analyzed by *Hannaea* and the results were compared side-by-side (Tables 4 and 5). Diatom metrics generated from the replicate samples resulted in identical bioassessment ratings for this site: minor impairment and full support of aquatic life uses.

Upon receipt of the samples, station and sample information were recorded in a laboratory notebook and the samples were assigned a unique number compatible with the Montana Diatom Database, e.g., 2250-01. The first part of this number (2250) designates the sampling site (Big Spring Creek at County Farm below Lewistown); the second part of this 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 along with station and sample information provided by MDEQ. A portion of the raw sample was used to make duplicate diatom slides. After completing the diatom proportional count, the slide used for the count will be deposited in the University of Montana Herbarium in Missoula.

The be retained by *Hanna* in Helena.

Station information, sample information, and diatom proportional count data have been entered into the Montana Diatom Database, maintained on a PC by *Hanna* in Microsoft Access.

RESULTS AND DISCUSSION

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

SAMPLE NOTES

BS-1. The *Cladophora* in this sample was senescent. *Oscillatoria* was present both as an epiphyte and as free-living trichomes.

BS-2. Sample is silty; contains *Elodea* and other plant material.

M22BSPRC01. Some moss is present.

M22BSPRC02. Visible mats of *Phormidium* are present.

M22BSPRC05. Sample is silty.

NON-DIATOM ALGAE

Diatoms dominated periphyton samples collected from the

Burleigh and County Farm sites in May (Table 4). Green algae and cyanobacteria were also common at both sites. The Burleigh site above Lewistown supported more than twice the number of algal genera than did the County Farm site below Lewistown.

In August, diatoms (*Diatoma hyemalis*) dominated the site above Castle Creek just below the Big Springs. Two other chrysophycean algae--*Tribonema* and *Vaucheria*--ranked 2nd and 3rd in biomass here. All of these algae are typical of cold, spring-fed streams. Green algae were absent from this site.

The mat-forming cyanobacterium *Phormidium* dominated the algal flora below Pike Creek in August (Table 4). Diatoms were common here and ranked second. Green algae were also absent at this site, which supported only 2 genera of non-diatom algae.

The filamentous and potentially nuisance-forming green alga *Cladophora* dominated the algal flora of Big Spring Creek at the two sites below Lewistown that were sampled in August (Table 4). Diatoms ranked 2nd in biovolume at both sites and cyanobacteria were also present, especially as epiphytes on *Cladophora* at BSPRC05. Replicate samples from BSPRC03 generated very similar results (Table 4).

DIATOMS

May Samples. Since they were collected outside of the summer sampling "window", diatom metrics generated from the May samples cannot be compared with the biocriteria in Table 3. This is because samples collected during the cool seasons in Montana tend to be dominated by a single species.

Nitzschia fonticola dominated the diatom assemblages at both sites sampled in May (Table 5). This is a cool-season, but clean

water diatom that indicates meso-eutrophic conditions (Van Dam et al. 1994). *N. fonticola* is also motile and is adapted to living on unstable substrates.

The two periphyton samples that were collected in May from above and below Lewistown shared 64% of their diatom floras (Table 5). This indicates that the two sites were very similar, floristically, and that little or no environmental change occurred between them. However, the two sites both supported a handful of teratological diatom valves, which may indicate the presence of toxic chemicals in the water.

August Samples. The sample collected just below Big Springs in August was dominated by *Diatoma hyemalis* (Table 5). This oligotrophic diatom indicates cold waters that do not vary in temperature by more than a few degrees seasonally. Although the Shannon species diversity and percent dominant species metrics for this site both indicated moderate impairment and partial support of aquatic life uses, the thermal stresses that cause this "impairment" may be considered natural and result from the discharge of the Big Springs.

Diatom metrics at sites 02 and 03 indicated minor impairment but full use support in August (Table 5). At site 02, a slightly elevated disturbance index and percent dominant species were due to an abundance of *Achnantheidium minutissimum*. Site 02 only shared about 12% of its flora with site 01, indicating that the flora of Big Spring Creek had changed from a spring flora to more of a stream flora at this point.

A somewhat elevated number of motile diatoms at site 03 indicated minor impairment from siltation here (Table 5). Sites 02 and 03 shared only about a quarter of their floras, indicating that a moderate amount of environmental change had occurred between them. These sites bracket the City of Lewistown and the

outfall from the Lewistown wastewater treatment plant.

The two replicate samples from site 03 generated a percent community similarity index of 84% (Table 5). In addition, diatom metrics from both sites yielded the same bioassessment rating: minor impairment but with full support of aquatic life uses. These results are acceptable from a quality assurance standpoint.

A large percent dominant species indicated moderate impairment and partial support of aquatic life uses at BSPRC05 (Table 5). The dominant species here was the eutrophic diatom *Cocconeis pediculus*. This diatom has a concave valve surface, by which it is adapted to living as an epiphyte on *Cladophora* and other filamentous algae. Its abundance at site 05 is directly related to dominance at this site by *Cladophora* (Table 4). *Cladophora* and *Cocconeis* indicate that the probable cause of impairment at this site is nutrient enrichment.

Sites 03 and 05 shared only about a third of their diatom floras, indicating that a moderate amount of environmental change (degradation) occurred between them. Both sites are downstream from the outfall of the Lewistown wastewater treatment plant.

All of the sites sampled in August, except the one just below Big Springs, had a few teratological diatom valves. This may indicate small concentrations of toxic chemicals, including heavy metals. None of the sites sampled in May and August supported a large number of diatoms in the family Epithemiaceae. This indicates that phosphorus, not nitrogen, was likely the limiting nutrient in Big Spring Creek.

The overall impairment ratings for Big Spring Creek would not change if metrics for sites 03 and 05 in the E-2 segment below Lewistown were compared to criteria for plains streams instead of mountain streams. Site 03 would still have good

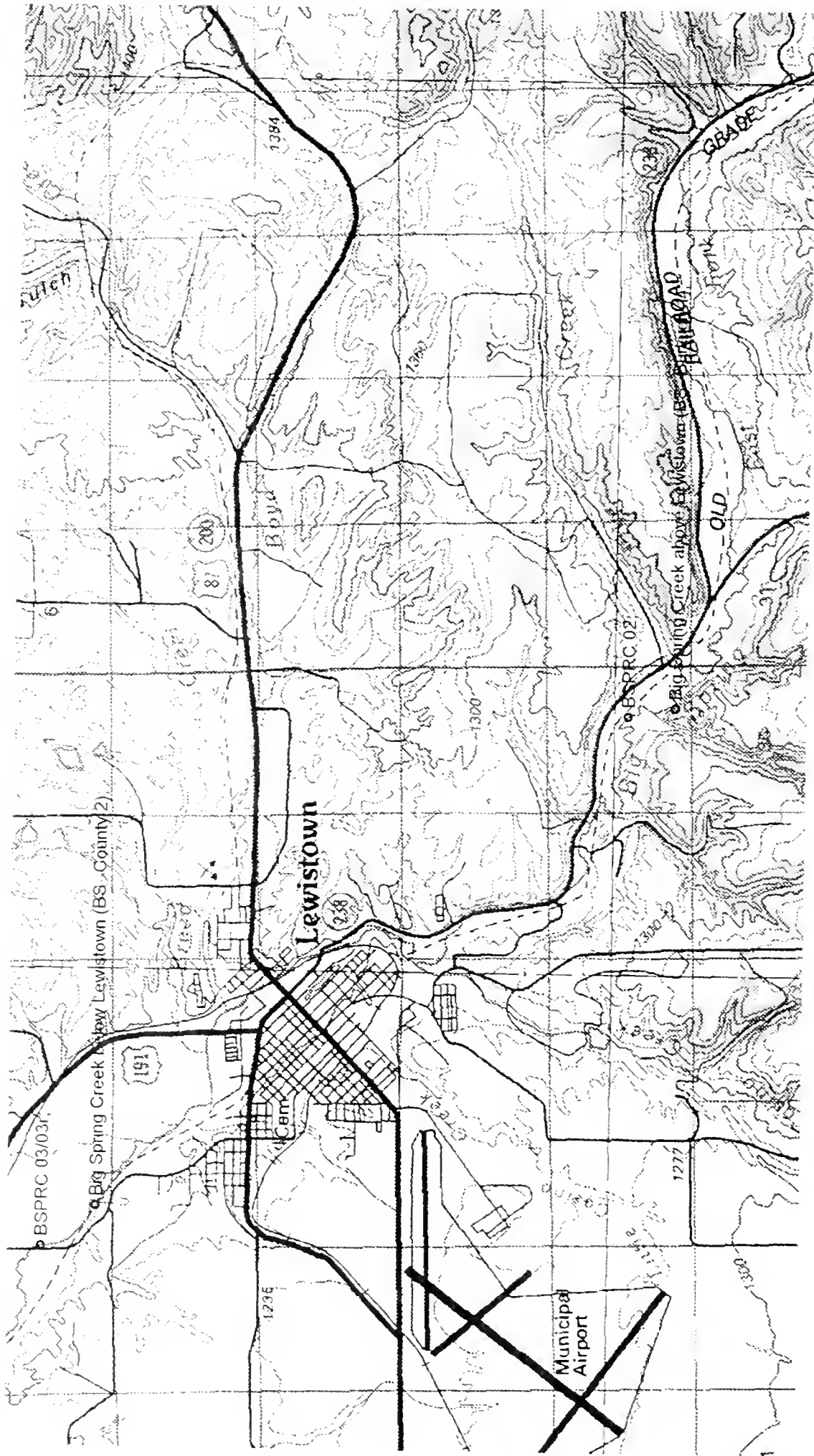
biological integrity (minor impairment due to teratological cells) and site 05 would have fair biological integrity due to elevated percent dominant species and depressed diatom diversity.

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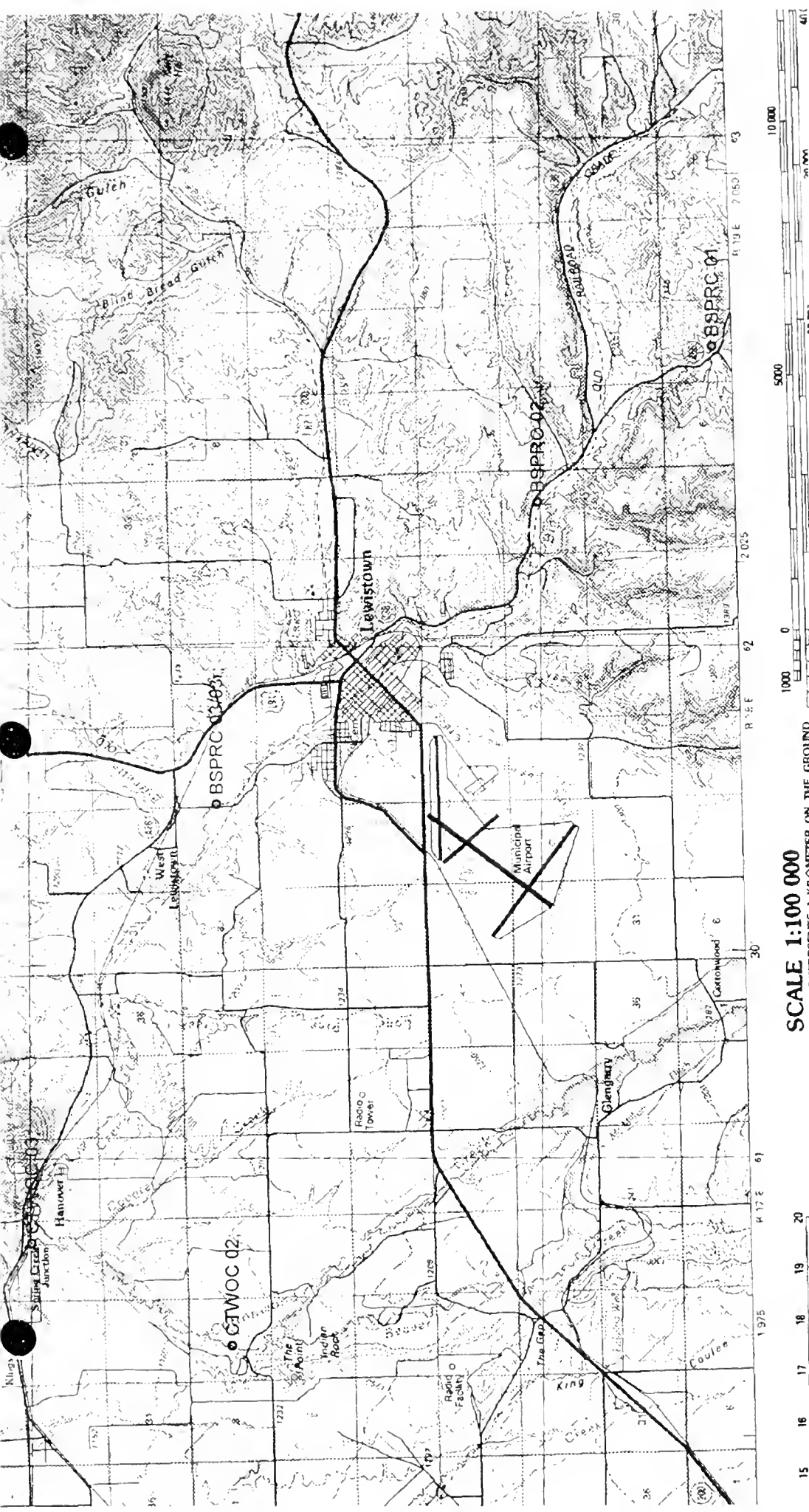
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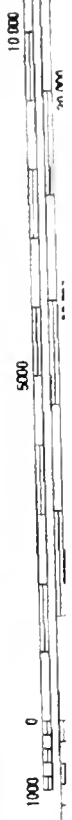
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Map 1



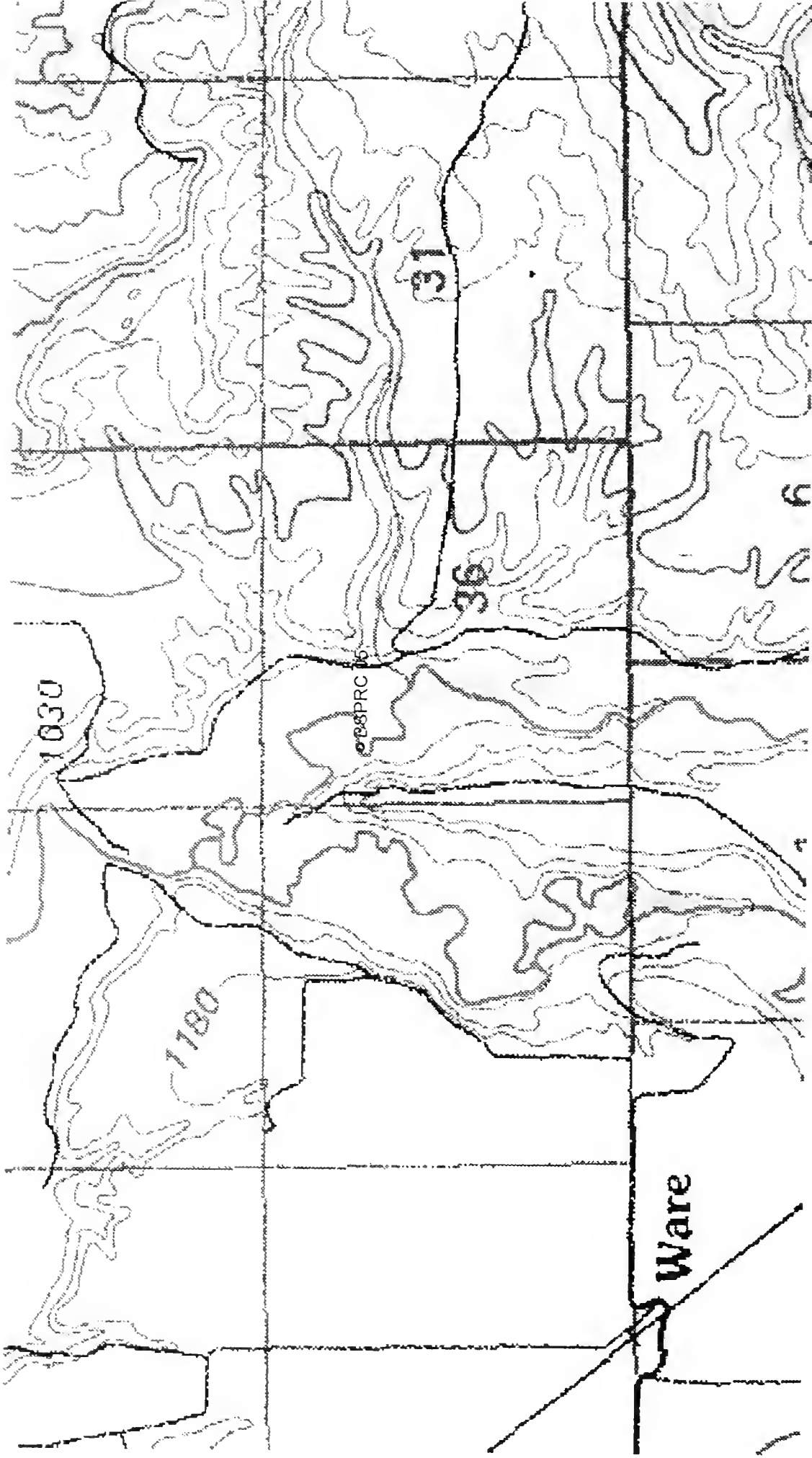
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Map 2



Map 3

Table 1. Location of periphyton sampling stations on Big Spring Creek in 2001.

Station	Station Code	Sample Number	Latitude/ Longitude	Sample Date
Big Spring Creek at Burleigh above Lewistown	BS-1	2251-01	47 01 40 109 22 53	5/25/01
Big Spring Creek at County Farm below Lewistown	BS-2	2250-01	47 05 10 109 27 13	5/25/01
Big Spring Creek headwaters above Castle Creek	M22BSPRC01	1038-03	47 00 12 109 20 40	8/20/01
Big Spring Creek below Pike Creek	M22BSPRC02	1756-02	47 01 57 109 22 57	8/20/01
Big Spring Creek below Lewistown	M22BSPRC03	1822-02	47 05 30 109 27 34	8/20/01
Big Spring Creek below Lewistown (replicate)	M22BSPRC03r	1822-03	47 05 30 109 27 34	8/20/01
Big Spring Creek near mouth in section 36	M22BSPRC05	1759-02	47 11 46 109 38 02	8/22/01

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*.

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 **mountain** 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 Cells	Percent Abnormal Cells	Percent Similarity Index ¹
Excellent None/Full Support	>2.99	>2.50	<20.0	<25.0	>29	<25.0	0.0	>59.9
Good/Minor Full Support	2.00- 2.99	2.01- 2.50	20.0- 39.9	25.0- 49.9	20- 29	25.0- 49.9	>0.0- <1.0	40.0- 59.9
Fair/Moderate Partial Support	1.00- 1.99	1.50- 2.00	40.0- 59.9	50.0- 74.9	10- 19	50.0- 74.9	1.0- 9.9	20.0- 39.9
Poor/Severe Nonsupport	<1.00	<1.50	>59.9	>74.9	<10	>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 unimpacted 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 florae in common (Bahls 1993). PCS may also be used to gauge the relative amount of impairment or recovery that occurs between adjacent study sites: >59.9% = very similar florae, no change; 40.0-59.9% = somewhat similar florae, minor change; 20.0-39.9% = somewhat dissimilar florae, moderate change; <20.0% = very dissimilar florae, major change.

Table 4. Relative abundance of cells and ordinal rank by biovolume of diatoms (Class Bacillariophyceae) and genera of non-diatom algae in periphyton samples collected from Big Spring Creek in 2001: d = dominant; a = abundant; f = frequent; c = common; o = occasional; r = rare.

Taxa	Station						
	BS-1	BS-2	BSPPC01	BSPRC02	BSPRC03	BSPRC03r	BSPRC05
Chlorophyta (green algae)							
Ankistrodesmus							
Cladophora	o/7	o/3			o/5	o/6	o/8
Hormidium	o/10				d/1	d/1	d/1
Mougeotia	o/8						
Oedogonium	a/2	o/5			a/3	a/3	o/7
Scenedesmus							
Spirogyra	f/3						
Ulothrix	o/9	c/4					
Zygonium	c/6						
Chrysophyta (golden algae)							
Bacillariophyceae	a/1	d/1		c/2	d/2	d/2	f/2
Tribonema	c/5		d/1				
Vaucheria			c/3				
			o/2				
Cyanophyta (cyanobacteria)							
Calothrix							
Chamaesiphon			o/6				a/3
Chroococcus							a/4
Oscillatoria	f/4	f/2	o/4	c/3	c/4	o/4	o/6
Phormidium			o/5	a/1		o/5	c/5
No. of Non-Diatom Genera	9	4	5	2	4	5	7

Table 5. Percent abundance of major diatom species and values of selected diatom association metrics for periphyton samples collected from Big Spring Creek in 2001. Underlined values indicate full support of aquatic life uses with minor impairment; **bold values** indicate partial support and moderate impairment; **underlined and bold values** indicate nonsupport and severe impairment; all other values indicate full support of aquatic life uses and no impairment when compared to criteria for mountain streams in Table 3.

Species/Metric (Trophic State ¹)	Station						
	BS-1	BS-2	BSPRC01	BSPRC02	BSPRC03	BSPRC03r	BSPRC05
<i>Achnanthes biasolettianum</i> (m)	2.02	0.23	0.60	8.10	0.66	0.33	
<i>Achnantheidium minutissimum</i> (t)	17.02	5.25	2.28	46.31	4.29	6.54	9.63
<i>Amphora pediculus</i> (e)	2.26	1.82	1.20	2.38	5.05	6.65	4.63
<i>Cocconeis pediculus</i> (e)	0.00		0.00		6.04	4.58	50.85
<i>Cymbella affinis</i> (e)	2.74	1.37		10.60	0.99	0.87	0.24
<i>Diatoma hyemalis</i> (o)	0.00		70.98				
<i>Diatoma vulgare</i> (m-e)	1.07	2.05	0.48	0.24	9.45	10.47	
<i>Encyonopsis microcephala</i> (m-e)	1.19	0.46	0.00	4.88	0.88	1.85	7.68
<i>Gomphonema minutum</i> (e)	0.24	2.51	0.36		12.31	12.76	0.12
<i>Navicula cryptotenella</i> (t)	7.14	2.05		1.55	2.86	1.64	2.20
<i>Navicula tripunctata</i> (e)	4.29	6.84	0.48	0.95	8.24	3.93	1.22
<i>Nitzschia fonticola</i> (m-e)	35.71	52.45	6.35	4.29	12.75	13.41	0.24
<i>Nitzschia palea</i> (e)	0.71	7.30		0.48		0.65	0.85
<i>Rhoicosphenia curvata</i> (e)		0.80	0.00		15.27	11.89	2.07
Number of Cells Counted	420	438	417	420	455	458	410
Shannon Species Diversity	3.65	3.06	1.94	3.13	4.01	4.17	<u>2.95</u>
Pollution Index	2.79	2.73	2.89	2.80	2.80	2.78	2.78
Siltation Index	59.40	78.90	6.84	8.69	<u>33.52</u>	<u>29.12</u>	17.68
Disturbance Index	17.02	5.25	2.28	<u>46.31</u>	4.29	6.54	9.63
Number of Species Counted	46	41	<u>22</u>	32	37	43	38
Percent Dominant Species	35.71	52.45	70.98	<u>46.31</u>	15.27	13.41	50.85
Percent Abnormal Cells	0.95	0.68	0.00	<u>0.24</u>	<u>0.33</u>	<u>0.11</u>	<u>0.98</u>
Percent Epithemiaceae	0.00	0.23	0.00	0.48	0.22	0.00	0.49
Similarity Index	63.70			12.07	26.62	83.61	33.01

¹ e = eutraperthentic; m = mesotraperthentic; m-e = meso-eutraperthentic; o = oligotraperthentic; t = tolerant of a wide range of trophic state from oligotraperthentic to eutraperthentic.

APPENDIX A: DIATOM PROPORTIONAL COUNTS

Sample	Genus/Species/Variety	Pollution Tolerance Class	Count	Percent
225101	Achnanthes lanceolata	2	3	0.36
225101	Achnanthidium biasolettianum	3	17	2.02
225101	Achnanthidium minutissimum	3	143	17.02
225101	Amphipleura pellucida	2	1	0.12
225101	Amphora libyca	3	1	0.12
225101	Amphora pediculus	3	19	2.26
225101	Aulacoseira distans	3	1	0.12
225101	Cocconeis pediculus	3	0	0.00
225101	Cocconeis placentula	3	16	1.90
225101	Cymbella affinis	3	23	2.74
225101	Cymbella cistula	3	1	0.12
225101	Diatoma hyemalis	3	0	0.00
225101	Diatoma moniliformis	2	7	0.83
225101	Diatoma vulgare	3	9	1.07
225101	Encyonema silesiacum	2	11	1.31
225101	Encyonopsis microcephala	2	10	1.19
225101	Frustulia vulgaris	2	2	0.24
225101	Gomphonema angustatum	2	1	0.12
225101	Gomphonema minutum	3	2	0.24
225101	Gomphonema olivaceum	3	9	1.07
225101	Gomphonema parvulum	1	2	0.24
225101	Gyrosigma acuminatum	3	1	0.12
225101	Melosira varians	2	8	0.95
225101	Navicula acceptata	2	0	0.00
225101	Navicula capitata	2	2	0.24
225101	Navicula capitatoradiata	2	11	1.31
225101	Navicula cryptotenella	2	60	7.14
225101	Navicula pupula	2	2	0.24
225101	Navicula reichardtiana	2	10	1.19
225101	Navicula tripunctata	3	36	4.29
225101	Nitzschia acicularis	2	5	0.60
225101	Nitzschia apiculata	2	2	0.24
225101	Nitzschia dissipata	3	34	4.05
225101	Nitzschia fonticola	3	300	35.71
225101	Nitzschia frustulum	2	2	0.24
225101	Nitzschia heufleriana	3	8	0.95
225101	Nitzschia linearis	2	1	0.12
225101	Nitzschia palea	1	6	0.71
225101	Nitzschia paleacea	2	2	0.24
225101	Nitzschia sociabilis	2	14	1.67
225101	Nitzschia vermicularis	2	1	0.12
225101	Pseudostaurosira brevistriata	3	8	0.95
225101	Reimeria sinuata	3	2	0.24
225101	Staurosira construens	3	2	0.24
225101	Staurosirella leptostauron	3	28	3.33
225101	Staurosirella pinnata	3	6	0.71
225101	Surirella minuta	2	3	0.36
225101	Synedra nana	3	4	0.48
225101	Synedra ulna	2	4	0.48

Sample	Genus/Species/Variety	Pollution Tolerance Class	Count	Percent
225001	<i>Achnanthydium biasolettianum</i>	3	2	0.23
225001	<i>Achnanthydium minutissimum</i>	3	46	5.25
225001	<i>Amphipleura pellucida</i>	2	1	0.11
225001	<i>Amphora pediculus</i>	3	16	1.82
225001	<i>Cocconeis placentula</i>	3	2	0.23
225001	<i>Cymbella affinis</i>	3	12	1.37
225001	<i>Denticula tenuis</i>	3	2	0.23
225001	<i>Diatoma mesodon</i>	3	1	0.11
225001	<i>Diatoma moniliformis</i>	2	8	0.91
225001	<i>Diatoma vulgaris</i>	3	18	2.05
225001	<i>Encyonopsis microcephala</i>	2	4	0.46
225001	<i>Gomphonema minutum</i>	3	22	2.51
225001	<i>Gomphonema olivaceum</i>	3	25	2.85
225001	<i>Melosira varians</i>	2	2	0.23
225001	<i>Navicula acceptata</i>	2	2	0.23
225001	<i>Navicula capitatoradiata</i>	2	9	1.03
225001	<i>Navicula cryptotenella</i>	2	18	2.05
225001	<i>Navicula gregaria</i>	2	2	0.23
225001	<i>Navicula halophila</i>	2	2	0.23
225001	<i>Navicula menisculus</i>	2	3	0.34
225001	<i>Navicula pelliculosa</i>	1	2	0.23
225001	<i>Navicula reichardtiana</i>	2	5	0.57
225001	<i>Navicula tripunctata</i>	3	60	6.84
225001	<i>Navicula trivialis</i>	2	2	0.23
225001	<i>Nitzschia acicularis</i>	2	9	1.03
225001	<i>Nitzschia archibaldii</i>	2	4	0.46
225001	<i>Nitzschia dissipata</i>	3	24	2.74
225001	<i>Nitzschia fonticola</i>	3	460	52.45
225001	<i>Nitzschia heufferiana</i>	3	3	0.34
225001	<i>Nitzschia linearis</i>	2	4	0.46
225001	<i>Nitzschia palea</i>	1	64	7.30
225001	<i>Nitzschia pusilla</i>	1	10	1.14
225001	<i>Nitzschia recta</i>	3	1	0.11
225001	<i>Nitzschia sociabilis</i>	2	6	0.68
225001	<i>Nitzschia vermicularis</i>	2	2	0.23
225001	<i>Reimeria sinuata</i>	3	3	0.34
225001	<i>Rhoicosphenia curvata</i>	3	7	0.80
225001	<i>Staurosirella leptostauron</i>	3	7	0.80
225001	<i>Staurosirella pinnata</i>	3	2	0.23
225001	<i>Synedra nana</i>	3	1	0.11
225001	<i>Synedra ulna</i>	2	4	0.46

Sample	Genus/Species/Variety	Pollution Tolerance Class	Count	Percent
103803	<i>Achnanthes lanceolata</i>	2	11	1.32
103803	<i>Achnanthidium biasolettianum</i>	3	5	0.60
103803	<i>Achnanthidium minutissimum</i>	3	19	2.28
103803	<i>Amphora pediculus</i>	3	10	1.20
103803	<i>Cocconeis pediculus</i>	3	0	0.00
103803	<i>Cocconeis placentula</i>	3	7	0.84
103803	<i>Cyclotella distinguenda</i>	2	3	0.36
103803	<i>Cymbella cistula</i>	3	2	0.24
103803	<i>Cymbella falaisensis</i>	3	36	4.32
103803	<i>Denticula tenuis</i>	3	0	0.00
103803	<i>Diatoma hyemalis</i>	3	592	70.98
103803	<i>Diatoma mesodon</i>	3	5	0.60
103803	<i>Diatoma vulgare</i>	3	4	0.48
103803	<i>Encyonema silesiacum</i>	2	2	0.24
103803	<i>Encyonopsis microcephala</i>	2	0	0.00
103803	<i>Fragilaria capucina</i>	2	8	0.96
103803	<i>Fragilaria vaucheriae</i>	2	0	0.00
103803	<i>Gomphonema angustatum</i>	2	37	4.44
103803	<i>Gomphonema minutum</i>	3	3	0.36
103803	<i>Gomphonema parvulum</i>	1	13	1.56
103803	<i>Karayevia clevei</i>	3	2	0.24
103803	<i>Meridion circulare</i>	3	6	0.72
103803	<i>Navicula capitatoradiata</i>	2	0	0.00
103803	<i>Navicula decussis</i>	3	0	0.00
103803	<i>Navicula lundii</i>	2	0	0.00
103803	<i>Navicula minima</i>	1	0	0.00
103803	<i>Navicula tripunctata</i>	3	4	0.48
103803	<i>Nitzschia fonticola</i>	3	53	6.35
103803	<i>Rhoicosphenia curvata</i>	3	0	0.00
103803	<i>Staurosirella leptostauron</i>	3	0	0.00
103803	<i>Staurosirella pinnata</i>	3	5	0.60
103803	<i>Synedra ulna</i>	2	7	0.84

Sample	Genus/Species/Variety	Pollution Tolerance Class	Count	Percent
175602	<i>Achnanthydium biasolettianum</i>	3	68	8.10
175602	<i>Achnanthydium minutissimum</i>	3	389	46.31
175602	<i>Amphipleura pellucida</i>	2	0	0.00
175602	<i>Amphora inariensis</i>	3	4	0.48
175602	<i>Amphora pediculus</i>	3	20	2.38
175602	<i>Aulacoseira distans</i>	3	3	0.36
175602	<i>Cocconeis placentula</i>	3	23	2.74
175602	<i>Cymbella affinis</i>	3	89	10.60
175602	<i>Cymbella cistula</i>	3	2	0.24
175602	<i>Denticula kuetzingii</i>	3	2	0.24
175602	<i>Denticula tenuis</i>	3	2	0.24
175602	<i>Diatoma moniliformis</i>	2	31	3.69
175602	<i>Diatoma vulgare</i>	3	2	0.24
175602	<i>Encyonema brehmii</i>	2	6	0.71
175602	<i>Encyonema silesiacum</i>	2	22	2.62
175602	<i>Encyonopsis microcephala</i>	2	41	4.88
175602	<i>Fragilaria vaucheriae</i>	2	4	0.48
175602	<i>Gomphonema angustatum</i>	2	4	0.48
175602	<i>Gomphonema olivaceum</i>	3	2	0.24
175602	<i>Gomphonema parvulum</i>	1	3	0.36
175602	<i>Gomphonema pumilum</i>	3	9	1.07
175602	<i>Navicula acceptata</i>	2	0	0.00
175602	<i>Navicula capitatoradiata</i>	2	4	0.48
175602	<i>Navicula cryptotenella</i>	2	13	1.55
175602	<i>Navicula pupula</i>	2	0	0.00
175602	<i>Navicula reichardtiana</i>	2	2	0.24
175602	<i>Navicula tripunctata</i>	3	8	0.95
175602	<i>Nitzschia acicularis</i>	2	2	0.24
175602	<i>Nitzschia dissipata</i>	3	2	0.24
175602	<i>Nitzschia fonticola</i>	3	36	4.29
175602	<i>Nitzschia palea</i>	1	4	0.48
175602	<i>Nitzschia sociabilis</i>	2	2	0.24
175602	<i>Reimeria sinuata</i>	3	11	1.31
175602	<i>Staurosirella leptostauron</i>	3	7	0.83
175602	<i>Synedra ulna</i>	2	23	2.74

Sample	Genus/Species/Variety	Pollution Tolerance Class	Count	Percent
182202	<i>Achnanthes lanceolata</i>	2	2	0.22
182202	<i>Achnantheidium biasolettianum</i>	3	6	0.66
182202	<i>Achnantheidium minutissimum</i>	3	39	4.29
182202	<i>Amphora inariensis</i>	3	2	0.22
182202	<i>Amphora pediculus</i>	3	46	5.05
182202	<i>Aulacoseira islandica</i>	3	1	0.11
182202	<i>Cocconeis pediculus</i>	3	55	6.04
182202	<i>Cocconeis placentula</i>	3	31	3.41
182202	<i>Cymbella affinis</i>	3	9	0.99
182202	<i>Denticula kuetzingii</i>	3	2	0.22
182202	<i>Diatoma moniliformis</i>	2	18	1.98
182202	<i>Diatoma vulgare</i>	3	86	9.45
182202	<i>Encyonema silesiacum</i>	2	27	2.97
182202	<i>Encyonopsis microcephala</i>	2	8	0.88
182202	<i>Gomphoneis herculeana</i>	3	1	0.11
182202	<i>Gomphonema minutum</i>	3	112	12.31
182202	<i>Gomphonema olivaceum</i>	3	2	0.22
182202	<i>Gomphonema parvulum</i>	1	4	0.44
182202	<i>Gyrosigma attenuatum</i>	3	2	0.22
182202	<i>Karayevia clevei</i>	3	2	0.22
182202	<i>Melosira varians</i>	2	1	0.11
182202	<i>Navicula acceptata</i>	2	2	0.22
182202	<i>Navicula capitatoradiata</i>	2	30	3.30
182202	<i>Navicula cryptotenella</i>	2	26	2.86
182202	<i>Navicula gregaria</i>	2	3	0.33
182202	<i>Navicula menisculus</i>	2	4	0.44
182202	<i>Navicula pupula</i>	2	2	0.22
182202	<i>Navicula reichardtiana</i>	2	31	3.41
182202	<i>Navicula tripunctata</i>	3	75	8.24
182202	<i>Nitzschia capitellata</i>	2	0	0.00
182202	<i>Nitzschia dissipata</i>	3	8	0.88
182202	<i>Nitzschia fonticola</i>	3	116	12.75
182202	<i>Nitzschia inconspicua</i>	2	2	0.22
182202	<i>Nitzschia sociabilis</i>	2	2	0.22
182202	<i>Nitzschia supralitorea</i>	2	4	0.44
182202	<i>Rhoicosphenia curvata</i>	3	139	15.27
182202	<i>Staurosirella leptostauron</i>	3	2	0.22
182202	<i>Staurosirella pinnata</i>	3	0	0.00
182202	<i>Synedra ulna</i>	2	8	0.88

Sample	Genus/Species/Variety	Pollution Tolerance Class	Count	Percent
182203	<i>Achnanthes lanceolata</i>	2	2	0.22
182203	<i>Achnantheidium biasolettianum</i>	3	3	0.33
182203	<i>Achnantheidium minutissimum</i>	3	60	6.54
182203	<i>Amphipleura pellucida</i>	2	0	0.00
182203	<i>Amphora inariensis</i>	3	2	0.22
182203	<i>Amphora pediculus</i>	3	61	6.65
182203	<i>Cocconeis pediculus</i>	3	42	4.58
182203	<i>Cocconeis placentula</i>	3	45	4.91
182203	<i>Cymbella affinis</i>	3	8	0.87
182203	<i>Diatoma moniliformis</i>	2	14	1.53
182203	<i>Diatoma vulgare</i>	3	96	10.47
182203	<i>Encyonema auerswaldii</i>	2	1	0.11
182203	<i>Encyonema brehmii</i>	2	5	0.55
182203	<i>Encyonema silesiacum</i>	2	16	1.74
182203	<i>Encyonopsis microcephala</i>	2	17	1.85
182203	<i>Fragilaria capucina</i>	2	2	0.22
182203	<i>Fragilaria vaucheriae</i>	2	2	0.22
182203	<i>Gomphoneis herculeana</i>	3	2	0.22
182203	<i>Gomphonema minutum</i>	3	117	12.76
182203	<i>Gomphonema olivaceum</i>	3	8	0.87
182203	<i>Gomphonema parvulum</i>	1	9	0.98
182203	<i>Gomphonema pumilum</i>	3	2	0.22
182203	<i>Gomphonema truncatum</i>	3	2	0.22
182203	<i>Navicula acceptata</i>	2	6	0.65
182203	<i>Navicula capitatoradiata</i>	2	29	3.16
182203	<i>Navicula cryptotenella</i>	2	15	1.64
182203	<i>Navicula lundii</i>	2	3	0.33
182203	<i>Navicula menisculus</i>	2	2	0.22
182203	<i>Navicula recens</i>	2	2	0.22
182203	<i>Navicula reichardtiana</i>	2	39	4.25
182203	<i>Navicula stroemii</i>	2	2	0.22
182203	<i>Navicula tripunctata</i>	3	36	3.93
182203	<i>Nitzschia capitellata</i>	2	2	0.22
182203	<i>Nitzschia dissipata</i>	3	0	0.00
182203	<i>Nitzschia fonticola</i>	3	123	13.41
182203	<i>Nitzschia palea</i>	1	6	0.65
182203	<i>Nitzschia sociabilis</i>	2	2	0.22
182203	<i>Reimeria sinuata</i>	3	2	0.22
182203	<i>Rhoicosphenia curvata</i>	3	109	11.89
182203	<i>Stauroneis smithii</i>	2	2	0.22
182203	<i>Stausira construens</i>	3	2	0.22
182203	<i>Stausirella leptostauron</i>	3	11	1.20
182203	<i>Stausirella pinnata</i>	3	3	0.33
182203	<i>Synedra acus</i>	2	2	0.22
182203	<i>Synedra ulna</i>	2	3	0.33

Sample	Genus/Species/Variety	Pollution Tolerance Class	Count	Percent
175902	Achnanthydium minutissimum	3	79	9.63
175902	Amphora inariensis	3	2	0.24
175902	Amphora libyca	3	0	0.00
175902	Amphora pediculus	3	38	4.63
175902	Aulacoseira canadensis	3	0	0.00
175902	Aulacoseira distans	3	1	0.12
175902	Aulacoseira islandica	3	2	0.24
175902	Cocconeis pediculus	3	417	50.85
175902	Cocconeis placentula	3	27	3.29
175902	Cyclotella meneghiniana	2	1	0.12
175902	Cymbella affinis	3	2	0.24
175902	Encyonema silesiacum	2	3	0.37
175902	Encyonopsis microcephala	2	63	7.68
175902	Epithemia sorex	3	2	0.24
175902	Gomphonema minutum	3	1	0.12
175902	Gomphonema parvulum	1	10	1.22
175902	Gyrosigma attenuatum	3	1	0.12
175902	Navicula capitata	2	0	0.00
175902	Navicula capitatoradiata	2	26	3.17
175902	Navicula cryptotenella	2	18	2.20
175902	Navicula menisculus	2	5	0.61
175902	Navicula peregrina	2	2	0.24
175902	Navicula perpusilla	2	2	0.24
175902	Navicula radiosa	3	1	0.12
175902	Navicula reichardtiana	2	14	1.71
175902	Navicula tripunctata	3	10	1.22
175902	Navicula veneta	1	0	0.00
175902	Nitzschia acicularis	2	2	0.24
175902	Nitzschia capitellata	2	0	0.00
175902	Nitzschia dissipata	3	39	4.76
175902	Nitzschia fonticola	3	2	0.24
175902	Nitzschia frustulum	2	6	0.73
175902	Nitzschia heufleriana	3	2	0.24
175902	Nitzschia palea	1	7	0.85
175902	Nitzschia paleacea	2	4	0.49
175902	Nitzschia perminuta	3	2	0.24
175902	Nitzschia sigmoidea	3	1	0.12
175902	Nitzschia sociabilis	2	2	0.24
175902	Reimeria sinuata	3	1	0.12
175902	Rhoicosphenia curvata	3	17	2.07
175902	Rhopalodia gibba	2	2	0.24
175902	Staurosirella leptostauron	3	2	0.24
175902	Staurosirella pinnata	3	4	0.49

