

AQUATIC BIOLOGY OPERATIONS MANUALS
MINNESOTA ENVIRONMENTAL QUALITY BOARD
REGIONAL COPPER-NICKEL STUDY

April 27, 1977

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1. INTRODUCTION AND SCOPE OF STUDY

1.1. Introduction

Ecological communities are dynamic systems which are in a continual state of flux. The distribution and abundance of component species varies in response to complex changes in their environment. These fluctuations in community structure may be brought about by natural environmental changes such as those caused by seasonal and annual variation in climate, or may be man-induced such as changes resulting from the construction and operation of a mine.

Copper-nickel mining and associated development in northeastern Minnesota could have a major impact on the region's aquatic ecosystems. As a result of this potential for environmental impact, a regional environmental impact study and monitoring system was instituted for the purpose of:

- 1) characterizing the environmental conditions in the study area;
- 2) predicting the potential regional impacts of copper-nickel mining and associated developments upon the present environmental conditions;
- 3) collecting baseline data for future assessment of regional environmental changes induced by copper-nickel mining and associated development.

The aquatic biology study had the responsibility for collecting information on the study area's aquatic biota (except fishes). An aquatic biology monitoring system and survey was implemented to provide data for

the regional characterization, impact prediction, and establishment of a baseline for future assessments of impacts on aquatic ecosystems.

1.1.1. Regional Characterization

Regional ecosystems will be described by combining aquatic biological data with water quality and fisheries data to provide the following information:

- 1) describe interrelationships within the regional aquatic ecosystems;
- 2) categorize the aquatic ecosystems with respect to community structure and species associations;
- 3) identify impacted regional aquatic ecosystems;
- 4) identify aquatic ecosystems which should not be subjected to any disturbances;
- 5) determine relative assimilative capacities of aquatic ecosystems.

1.1.2. Impact Analysis

Based on information provided by the mining technology group, a list of potential effluents, land uses, and population changes will be developed. Applying bioassay data and information from the literature to the regional aquatic biology characterization, determinations can be made on the relative effect of potential effluents, land uses, and population changes to aquatic ecosystems. By using the ecosystem approach, impacts upon one portion of the biota can be shown to directly or indirectly affect other portions of the biota.

No quantitative estimates of the impact of copper-nickel mining or associated development can be expected because of complex interactions within aquatic ecosystems. Determination of impacts to the region's aquatic ecosystems will be based upon the professional judgement of the aquatic biology staff and staff advisors, and their knowledge of the systems involved.

1.1.3. Baseline Monitoring

The monitoring system provides the basis for assessing the actual regional impacts of copper-nickel mining and its associated developments in the future. Because aquatic organisms integrate the total environment, aquatic biological monitoring is important in determining the regional impacts on water quality and in providing data for developing models of interrelationships within aquatic ecosystems. The regional monitoring program does not replace site specific monitoring, but provides the capability to assess cumulative impacts of development within watersheds and serves as a model for future site specific studies.

1.2. Scope of Study

The study provides quantitative and qualitative baseline data on the aquatic ecosystems within the potential copper-nickel development region. Because of the region's size, much of the sampling is qualitative. At key points, monitoring of a quantitative nature is undertaken along with the qualitative survey work. The remainder of this manual describes the

stream sampling program. The lake sampling program is described in a separate manual.

1.2.1. Periphyton

Periphyton (attached algae) are essentially the only primary producers in streams, and are of prime importance to stream ecosystems. Periphyton can be sampled to provide a measure of primary production (chlorophyll a), and to determine the community composition in a given stream. Because periphyton grow attached to the substrate, they can provide a valuable biological monitor to detect subtle changes in water quality.

1.2.2. Macrophytes

Macrophytes were assigned a low sampling priority in the stream studies because they are of limited ecological importance in these streams. Their regional distribution is limited, they are difficult to sample quantitatively, and they lack prime importance in stream food chains. For this study, the macrophytes' most important function is as heavy metals monitors because they concentrate metals in their tissues.

1.2.3. Benthic Invertebrates

Benthic invertebrates play an important role in the transfer of energy from primary producers (periphyton) and allochthonous materials to fish. Therefore, the benthic community is sampled to facilitate detection of biological changes which might indicate changing environmental conditions.

1.2.4. Heavy Metals

Increases in heavy metals concentrations in streams may be reflected in the tissue of aquatic organisms. Analysis for heavy metals in periphyton, macrophytes, and invertebrate tissues may be valuable in developing models of heavy metals movement through the ecosystem and in selecting organisms to serve as biological monitors of long term changes in heavy metals concentrations.

1.3. Time Schedule Chart

A time schedule for the overall aquatic biology study appears in Table S-1.

2. STUDY AREA AND SAMPLING STATIONS

2.1. Study Area

The study area encompassed approximately 700 square miles. Ely was on the northern edge of the area while Aurora and Hoyt Lakes were situated on the southern edge. Sampling was designed to monitor the region's major watersheds, which include areas to be directly affected by copper-nickel mining, areas affected by related development, and unaffected areas.

2.2. Sampling Stations

Sampling stations were selected to closely coincide with the water quality sampling stations, thus facilitating correlation of physical and biological data. Exact locations were determined during on-site inspections. An attempt was made to locate sample sites in riffle

areas upstream from any bridge or dam structures; although sometimes this was not possible. Sampling station locations and designations are listed in Table S-2. Justification for each station is listed in Table S-3.

Primary, secondary, and tertiary station designations were assigned to indicate sampling priority (see Table S-3). Primary sites monitored over-all conditions of major watersheds. There were four quantitative and qualitative sampling periods at primary sites (Table S-4). Priority was assigned to the analysis of the first and third sets of samples. Semi-annual qualitative surveys were made at the tertiary sites. Although tertiary sites have the possibility of impact at a later date, they were considered less important in the overall region. Other qualitative samples were collected in small tributaries as time permitted.

2.2.1. Regional Maps

A regional map showing locations of sampling sites appears in Appendix S-1.

3. METHODOLOGY

3.1. Techniques

3.1.1. Justification of Techniques

There is no standard quantitative method for sampling aquatic ecosystems; therefore, artificial substrate samplers were selected as the primary quantitative method for periphyton and benthic invertebrate collection. These samplers were designed to standardize and simplify quantitative field sampling, and used because traditional quantitative devices are

susceptible to human bias and cannot be used under all conditions. Artificial substrates equalize sampling effort at all stations and reduce the variability within quantitative data under most conditions.

The major drawback in using artificial substrates is their selective nature. No matter what type of substrate is used, colonization by all organisms found in a given ecosystem will not occur. However, even natural substrates are selective, and unless a quantitative bottom sample can be collected from each natural substrate type, bottom samples would not produce a more reliable estimate of the natural community structure.

Artificial substrates, which may or may not mimic the natural substrates, are colonized by aquatic organisms. These colonizing organisms are subjected to all the fluctuations in chemical and most physical parameters. Because the sampling effort at each station is similar, changes in water quality, as related to the biota, may be observed. Beak et al. (1973) reviewed the uses of artificial substrates for biological monitoring.

Various types of artificial substrate samplers have been developed for sampling benthic invertebrates. These substrates include concrete slabs (Moon, 1935), hardware cloth brush boxes (Wene and Wickliff, 1970), multiple plate samplers (Hester and Dendy, 1962; Fallner, 1971), and rock filled baskets (Henson, 1965). Artificial substrates for collecting periphyton generally consist of glass or plexiglass plates, although there have been experiments with styrofoam and polyethylene strips. A

variety of methods have been proposed for suspending these substrate materials in lakes and streams. The literature on artificial substrate sampling for periphyton has been reviewed by Sladeckova (1962).

Drifting benthic invertebrates were collected also. Predation and population estimates (Elliot, 1970), and secondary production (Waters, 1962) may be derived from drift data. Although heavily studied (see Waters, 1972 for literature review), drift has rarely been used in biological monitoring. Because invertebrate drift originates from various upstream habitats, a wide variety of organisms are captured; thus giving drift promise as a valuable monitoring tool in the future. Biologists may use these data for faunal surveys or as an additional monitoring method for detecting biological changes (Larimore, 1974; Wallace and Hynes, 1975). A complete review of drift methods and their applications was done by Elliot (1970).

We carried out qualitative sampling to: develop complete species lists; survey areas that time constraints prevent sampling quantitatively; and sample those species not collected in quantitative samples due to the selectivity of artificial substrates and drift nets. Weber (1973) fully outlines the rationale and methods for qualitative sampling.

3.1.2. Procedures

General field and laboratory procedures are outlined in Appendix S-2. An outline of sampling techniques and analyses used in the aquatic

biology monitoring system follows. Details of procedures are found in appendixes.

3.1.2.1. Periphyton

Artificial substrates consisting of glass slides were used to quantitatively sample periphyton (Weber, 1973; American Public Health Association, 1975). Three racks which hold four slides were suspended approximately 30 cm below the water surface for a three-week colonization period. In addition, a qualitative survey of the naturally occurring periphyton was carried out. All natural substrates were observed and any growth collected. Sampling frequencies are indicated in Table S-4. Complete field and laboratory procedures are outlined in Appendix S-3.

3.1.2.2. Macrophytes

Macrophytes were qualitatively sampled in the streams during the peak growth period which normally occurs in August. Macrophytes were collected for identification and for heavy metals tissue analysis at selected stations (see Section 3.1.2.4.). Samples were collected by pulling macrophytes from the bottom substrate and placing them in plastic bags. Whole plants were collected including flowers and seed pods; and roots, rhizomes, or tubers if present. In addition, in an effort to collect both common and rare species, all habitat types were sampled. No semi-aquatic plants were collected during this survey.

3.1.2.3. Benthic Invertebrates

Modified Hester-Dendy samplers (Weber, 1973) were the artificial substrate samplers employed for benthic invertebrates collection. Unlike most other types of artificial substrates, these samplers provide a known area of colonization. Sampling with Hester-Dendy samplers is easy because of collection and processing simplicity. Three samplers were suspended in the streams at approximately mid-depth for a six-week colonization period (Weber, 1973). In cases of low water, samplers were placed on the stream bottom. Three replicate samplers were placed at each primary and secondary site as reported in the sample schedule in Table S-5. A standard #40 sieve was employed to separate benthic invertebrates from fine debris in all sample collections.

Drift nets with an upstream opening of 0.025 m^2 , length of 2.4 m, and 440 μm mesh were placed in the streams for 24 hours, beginning during daylight hours. Because maximum drift occurs between sunset and sunrise, and is extremely low during the day, the exact starting time was not critical. Sampling followed the same schedule as the Hester-Dendy sampling outlined in Table S-5.

Because Hester-Dendy samplers and drift nets are selective in their sampling, qualitative invertebrate samples were collected to compile a species list of the entire natural community. Sampling consisted of examining, for two man-hours, all the various stream habitat types such as pools and riffles, various size rocks, logs, and silt. Quali-

tative sampling followed the schedule listed in Table S-5. Complete field collection and laboratory methods for benthic invertebrate samples are included in Appendix S-4.

3.1.2.4. Heavy Metals Tissue Analysis

Periphyton, macrophytes, and invertebrates were collected for heavy metals analysis in conjunction with the qualitative survey work. These samples were collected in August at the primary stations and secondary stations KC-1, BB-1, F-1, and B1-1. At the laboratory, samples were split with the first portion remaining at the field station for identification and the remainder shipped for analysis. Macrophytes and periphyton were shipped in water from the sampling station, while invertebrates were frozen. Complete procedures for collection and processing of heavy metals samples are included in Appendix S-5.

3.1.2.5. Permanent Archive

A reference collection of the aquatic invertebrates and aquatic macrophytes found in the study region has been made. This collection contains representative individuals of each species found. Invertebrates were preserved in 70 percent ethanol, except for chironomids which were mounted on slides using CNCP-10 mounting media. In addition to the reference collection, each sample was preserved in 70 percent ethanol for possible inclusion in the archives. Hyrax diatom mounts and CNCP-10 mounts made directly on periphyton slides may also

be included in the archives.

Portions of samples analyzed for heavy metals were preserved for future reference. Macrophyte and periphyton samples were dried, ground, and stored in Teflon vials. Invertebrate samples were freeze-dried and stored in vials.

3.1.3. Information Inputs From Other Programs

Information and data from the water quality and fisheries studies were necessary to analyze aquatic biology data for the regional characterization. Water quality data were obtained through the Operations and Data Coordination staff. Fisheries data were obtained directly from the Fish Study Project group. Data from leaching and bioassay programs were needed for impact analysis, and also obtained through the Operations and Data Coordination staff.

3.2. Sampling Frequency

Sampling frequencies are given in Table S-4. For secondary samples, collection occurs at the same frequency as primary samples. However, the even set of secondary samples (second, fourth, etc.) will not be analyzed unless time and budget permit.

3.3. Equipment

An equipment list is given in Appendix S-6. Drawings of the Hester-Dendy sampler, periphyton sampler, and anchor for samplers appear in Figure S-1.

3.4. Quality Control Provisions

3.4.1. General

All aquatic biology personnel underwent a field training program before regular sampling began. This program was used to demonstrate the collection techniques employed in the aquatic biology sampling program. All personnel had a working knowledge of all procedures even though they were not directly involved with each phase of the project.

Personnel were required to follow procedures outlined in this operations manual. Because the aquatic biology study was expected to evolve, field personnel were encouraged to develop new techniques, procedures, and parameters to be sampled. These new techniques were approved by the head of the aquatic biology program before implementation. Under unusual field conditions, senior personnel were expected to take appropriate action. All changes in sampling routines were recorded in the field notebook. Compliance checks with field and laboratory procedures were made by the project head to ensure that sampling was carried out efficiently and accurately.

To ensure sample integrity, samples were recorded as they were collected. Additional checks were made at the time samples were placed into storage and analyzed. Data sheets were developed for following the movement of samples from stream to data within the final document.

To ensure taxonomic quality, selected periphyton and invertebrate samples were sent to consulting taxonomists for analysis. Further verification of reference species was carried out by experts in the various taxonomic areas.

For invertebrate samples, these experts were: P.A. Lewis, USEPA, Cincinnati, Ohio (Stenonema and Stenacron); W. McCafferty, Purdue University (Ephemeroptera); Unzicker, Illinois Natural History Survey (Trichoptera); W. Hilsenhoff, University of Wisconsin-Madison (Plecoptera); E.F. Cook, University of Minnesota (Diptera, Odonata, and Hemiptera); R.W. Gundersen, St. Cloud State University (Coleoptera); and William Beck, Florida A and M University (Chironomidae). Selected periphyton slides were sent to Dr. Charles Reimer, Philadelphia Academy of Natural Sciences. In addition, some periphyton samples were exchanged between project taxonomists, and slides reanalyzed to detect errors.

3.4.2. Calibration, Maintenance, and Testing of Equipment

Equipment manuals are found in the Hennepin Square and Kawishiwi Field Lab offices. These manuals give procedures for operation, maintenance, and calibration of equipment.

4. RECORD KEEPING AND DATA TRANSMISSION

No specific data notebook was kept. Instead, numbered sheets were used and bound in a notebook upon completion of the monitoring program. A spiral-bound notebook was kept for following the movement of samples from stream to laboratory to analysis to final report. Pencil was used to make all recordings in the field. Abbreviations are explained in Appendix S-7. Sample data sheets also appear in Appendix S-7. Aquatic biology data were transmitted to the Operations and Data Coordination staff on a monthly basis.

5. RESULTS, ANALYSIS, AND REPORTS

5.1. Numerical Analysis

The following numerical measures may be used for analysis for both periphyton and benthic invertebrate data:

- 1) species list--identification to the lowest possible taxonomic level
- 2) density--individuals/unit area calculated as a total and for individual species; this could also be expressed as numbers/sampler
- 3) relative abundances
- 4) diversity index, $\bar{d} = - \sum (Ni/N) \log Ni/N$ (from Wilhm and Dorris, 1968)
- 5) redundancy; $r = \frac{\bar{d}_{max} - \bar{d}}{\bar{d}_{max} - \bar{d}_{min}}$ (from Wilhm and Dorris, 1968)
- 6) coefficients of similarity

a) $C = \frac{2 (\sum P_j)}{\sum P_i + \sum P_j}$ (from Burlington, 1962)

where:

P (prominence value) = density x frequency at all stations

$\sum P_j$ = sum of lower of two P values the two stations have in common

$\sum P_i$ = sum of P values at ith station

$\sum P_j$ = sum of P values at jth station

b) $B = 1/k \sum_{i=1}^k \frac{\min (X_{ia}, X_{ib})}{\max (X_{ia}, X_{ib})}$ (from Pinkham, 1976)

where:

k = number of comparisons between two stations

X_{ia}, X_{ib} = number of individuals in the ith taxon at stations a and b

$$c) \quad CC = \frac{P}{P + M} \quad (\text{from Roback et al, 1969})$$

where:

P = number of matches

M = number of taxon present at one station and absent at another

7) Chlorophyll a (ug/m²)

Depending on the magnitude of the variability in the parameters listed above, statistical and graphical interpretation methods may be employed to characterize the streams.

5.2. Progress and Final Reports

Quarterly reports were written about the aquatic biology study's status. In addition, monthly reports of data were provided when available.

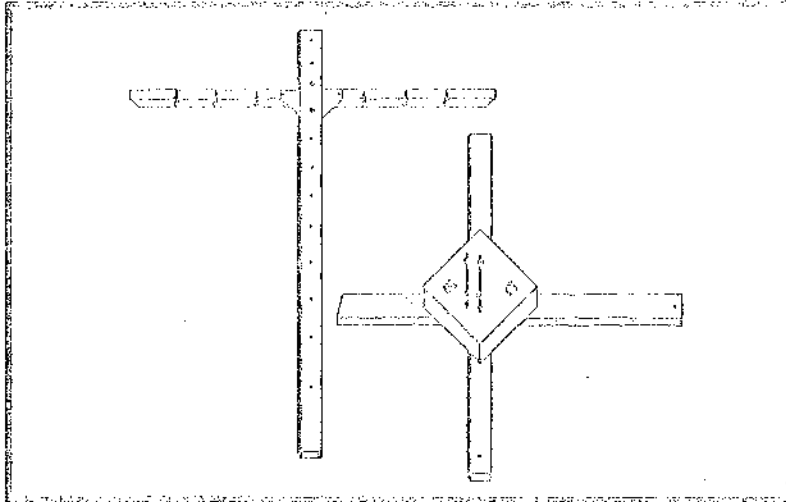
6. STUDY STAFF

Resumes of the aquatic biology staff are in Appendix S-8.

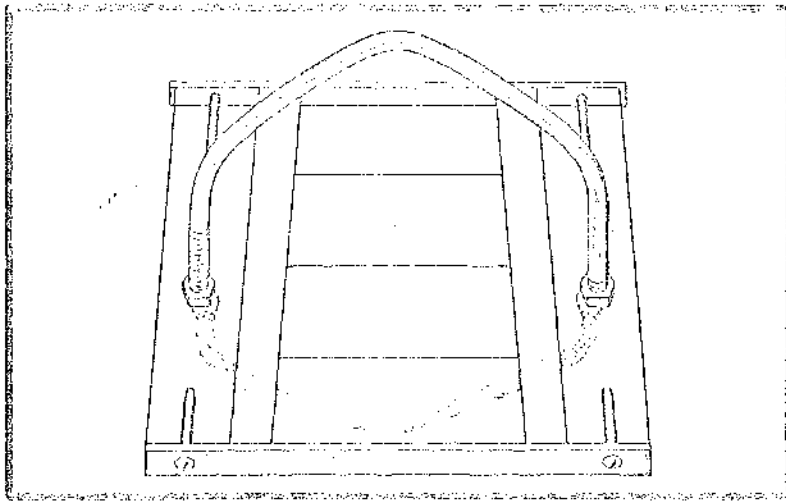
7. ASSOCIATED PERSONNEL

Dr. Thomas Waters of the University of Minnesota, Department of Entomology, Fish, and Wildlife was the only advisor to the stream aquatic biology program for the 1976 field program.

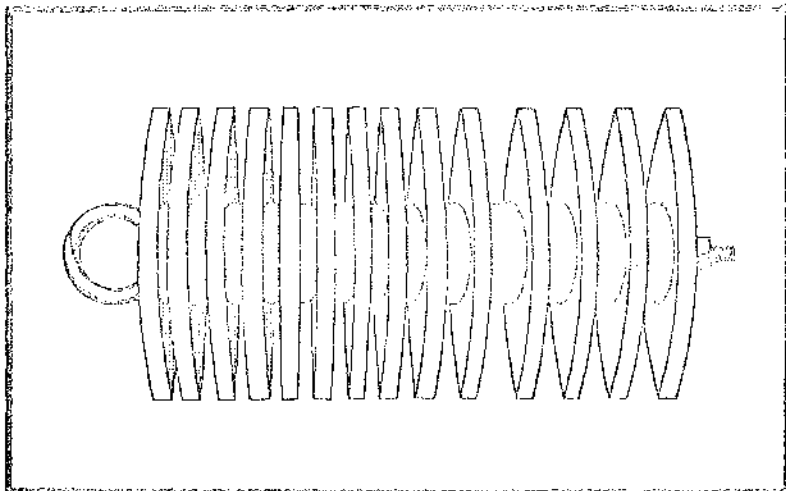
FIG. 3-1 Sampling Equipment



A.
ANCHORING
DEVICE



B.
PERIPHYTON
SAMPLER



C.
HESTER-
DERBY
SAMPLER

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Table S-1. Tentative Schedule for Aquatic Biology Study
(As of April 1977)

<u>Date</u>	<u>Items</u>
April and May 1976	-complete monitoring program design -hire staff
May 17, 1976	-start field monitoring program
October 1, 1976	-all heavy metal identifications complete -1976 field work essentially complete -taxonomic analysis of qualitative samples
December 1, 1976	-qualitative samples finished -begin preliminary report on qualitative data
December 15, 1976	-preliminary report completed -literature search completed -quantitative analysis begins
January 17, 1977	-January workshop -begin developing plans for 1977 monitoring program
February 1977	-winter field monitoring
April 1, 1977	-complete quantitative analysis -begin final draft of overall report and data interpretation for 1976 sampling
May 1, 1977	-complete report on overall 1976 monitoring program -complete plans for 1977 monitoring program
May 15, 1977	-begin 1977 field sampling -continue taxonomic analysis work
October 1, 1977	-complete field work -aquatic biology staff moves to Minneapolis -qualitative and quantitative analysis work continues -all literature review work completed
November 1, 1977	-data interpretation in progress -all field samples analyzed
April 1, 1978	-staff reduction -begin final preparation of aquatic biology section of environmental impact study and monitoring reports

Table S-1. Tentative Schedule for Aquatic Biology Study
(As of January 1977). (contd.)

<u>Date</u>	<u>Item</u>
July 1, 1978	-draft environmental impact study report completed -staff reduction
December 1, 1978	-final environmental impact study report completed
January 1, 1979	-Regional Copper-Nickel Study complete

Table S-2. Regional Biological Monitoring Stations.

<u>Dr-WI Designated Watershed Name</u>	<u>#</u>	<u>Location</u>	<u>Township</u>	<u>Range</u>	<u>Section</u>	<u>Rank</u>	<u>Station Designation</u>
Fall Lake	3	Outflow from Fall Lake, 100 yards upstream of Newton Lake	63N	11W	5	Primary	K-1
		Squaw Creek, north of Winton, between Cedar, Bass, and Fall Lakes	63N	11W	18	(a)	S-1
Shagawa	4	Outflow from Shagawa Lake, downstream 100 yards from Highway 88 crossing	63N	12W	26	Secondary	R-2
Kawishiwi	5	Outflow from Garden Lake, at Highway 169	63N	11W	20	Tertiary	K-3
		Outflow from White Iron Lake, at Highway 16	63N	11W	32	Tertiary	K-4
		Gaging station, Kawishiwi River	63N	9W	24	Tertiary	K-6
		Gaging station, South Kawishiwi River	62N	11W	23	Tertiary	K-7
		South Kawishiwi River, approximately 200 yards inside Boundary Waters Canoe Area boundary	62N	10W	6	Primary	K-8

(a) Additional station, not part of regular monitoring program.

Table S-2. Regional Biological Monitoring Stations. (contd.)

<u>Co-NI Designated Watershed Name</u>	<u>#</u>	<u>Location</u>	<u>Township</u>	<u>Range</u>	<u>Section</u>	<u>Rank</u>	<u>Station Designation</u>
Beck Island	6	Gaging station at Highway 1 (downstream from bridge)	62N	12W	23	Secondary	BI-1
Isabella	7	Gaging station at inflow to Bald Eagle Lake	61N	9W	6	Tertiary	I-1
		Little Isabella River at East Tomahawk Road, USFS Road 424	61N	9W	29	(a)	LIR-1
Filson Creek	8	Mouth of Filson Creek approximately one-eighth mile upstream from confluence with Kawishiwi	62N	11W	24	Secondary	F-1
Birch Lake	9	Birch River at Highway 21	61N	13W	27	(a)	BR-1
		Gaging station inflow to White Iron Lake, 150 yards downstream from Birch Lake dam	62N	11W	19	Secondary	K-3
Unnamed Creek	9a	Inflow to Bob Bay (Birch Lake)	61N	12W	36	Secondary	UB-1
Kesley Creek	9b	Mouth of Kesley Creek (Birch Lake)	61N	11W	17	Secondary	KC-1

(a) Additional station, not part of regular monitoring program.

Table S-2. Regional Biological Monitoring Stations. (contd.)

<u>Co-MI Designated Watershed Name</u>	<u>#</u>	<u>Location</u>	<u>Township</u>	<u>Range</u>	<u>Section</u>	<u>Rank</u>	<u>Station Designation</u>
Dunka	10	Gaging station adjacent to Highway 112	60N	12W	9	Primary	D-1
		Upstream; same as ABAX upstream site	60N	12W	27	Tertiary	D-2
Stony	11	Adjacent to Roaring Stony Resort	61N	11W	30	Primary	SR-1
		Gaging station at USFS Road 424	60N	11W	8	Tertiary	SR-2
		Located 0.8 miles upstream from Highway 1 crossing	60N	10W	28	Secondary	SR-3
		At USFS Road 106	60N	9W	31	Tertiary	SR-4
		Outflow from Greenwood Lake, about one mile downstream	59N	10W	21	Tertiary	SR-5
		Between Stations SR-3 and SR-4, close to Lake County Road 2	60N	10W	--	(a)	SR-6
St. Louis	13	Gaging station at Highway 100	58N	15W	21	Primary	SL-1
		At USFS Road 133	58N	15W	30	Tertiary	SL-2
		Immediately upstream from Reserve Railroad Crossing	58N	12W	22	Secondary	SL-3

(a) Additional station, not part of regular monitoring program.

<u>On-It Designated Watershed Name</u>	<u>#</u>	<u>Location</u>	<u>Township</u>	<u>Range</u>	<u>Section</u>	<u>Rank</u>	<u>Station Designation</u>
Partridge	14	Gaging station at County Road 110 (downstream from bridge)	58N	14W	13	Primary	P-1
		County Road 110, one mile NE of Hoyt Lakes (upstream from bridge, upstream of Colby Lake)	58N	14W	9	Secondary	P-2
		Colvin Creek at USFS Road 420	58N	13W	9	Tertiary	P-3
		South Branch of Partridge River at USFS Road 113	59N	13W	25	Tertiary	P-4
		Downstream from Erie Railroad crossing	58N	12W	6	Secondary	P-5
		First and Second creeks, north of confluence with Partridge River	58N	15W	12	(a)	P&S-1
		Wyman Creek, north of Hoyt Lakes at County Road 110	58N	14W	4	(a)	W-2
Embarrass	15	Upstream from Embarrass Bridge	60N	15W	25	Primary	E-1
		At Highway 104 crossing	60N	13W	18	Tertiary	E-2

(a) Additional station, not part of regular monitoring program.

<u>Station Rank</u>	<u>#</u>
Primary	7
Secondary	10
Tertiary	13
Additional	6
Total	36

Note: All primary and secondary sites are in riffle areas unless otherwise specified.
Tertiary sites include all habitat types.

Table S-3. Selection of Biological Monitoring Stations.

<u>Watershed</u>	<u>Station</u>	<u>Rank</u>	<u>Justification</u>
Fall Lake	K-1	Primary	Overall biological conditions; inflow to BWCA; cumulative effect of all development north of Laurentian Divide.
Shagawa	K-2	Secondary	Potential population growth in Ely, existing impact from Ely sewage treatment plant.
Kawishiwi	K-3	Tertiary	Summation of North and South Kawishiwi.
Kawishiwi	K-4	Tertiary	Background information; summation of Bear Island and South Kawishiwi rivers.
Kawishiwi	K-6	Tertiary	Background information in control area.
Kawishiwi	K-7	Tertiary	Background information; potential effect from INCO site.
Kawishiwi	K-8	Primary	Upstream control station; monitors outflow from BWCA.
Bear Island	BI-1	Secondary	Overall biological conditions of drainage basin with low potential impact.
Isabella	I-1	Tertiary	Background information in control area.
Filson Creek	F-1	Secondary	Potential site of INCO open pit, drainage of INCO site into South Kawishiwi.
Birch Lake	K-5	Secondary	Summation of South Kawishiwi; downstream of proposed mining and Birch Lake outflow (potential and present development).
Unnamed Creek	BB-1	Secondary	Only known significant heavy metals input within study area.
Keeley Creek	KC-1	Secondary	Potential runoff from INCO tailing basin.

Table S-3. Selection of Biological Monitoring Stations. (contd.)

<u>Watershed</u>	<u>Station</u>	<u>Rank</u>	<u>Justification</u>
Dunka	D-1	Primary	Overall conditions of drainage basin; downstream of present and potential mining.
Dunka	D-2	Tertiary	Background information on upstream area of Dunka River and check on AMAX monitoring.
Stony	SR-1	Primary	Overall biological conditions of drainage basin, proposal for tailing basin upstream.
Stony	SR-2	Tertiary	Background information.
Stony	SR-3	Secondary	Upstream area of watershed below confluence of Stony and Greenwood rivers.
Stony	SR-4	Tertiary	Background information.
Stony	SR-5	Tertiary	Background information.
St. Louis	SL-1	Primary	Describes biological conditions in the Partridge and St. Louis systems, and the interaction between those systems.
St. Louis	SL-2	Tertiary	Background information.
St. Louis	SL-3	Secondary	Background conditions for headwaters area; potential tailing disposal site upstream.
Partridge	P-1	Primary	Overall biological conditions in Partridge River; potential for population increase in Aurora and Hoyt Lakes.
Partridge	P-2	Secondary	Potential mining development upstream; also upstream monitoring station for Aurora and Hoyt Lakes.
Partridge	P-3	Tertiary	Background information in area of potential tailing basin.
Partridge	P-4	Tertiary	Background information in area of potential tailing basin.

Table S-3. Selection of Biological Monitoring Stations. (contd.)

<u>Watershed</u>	<u>Station</u>	<u>Rank</u>	<u>Justification</u>
Partridge	P-5	Secondary	Upstream monitoring station for Partridge watershed.
Embarrass	E-1	Primary	Potential tailing disposal site located upstream; overall monitoring station for Embarrass watershed.
Embarrass	E-2	Tertiary	Background information in upstream area of watershed.

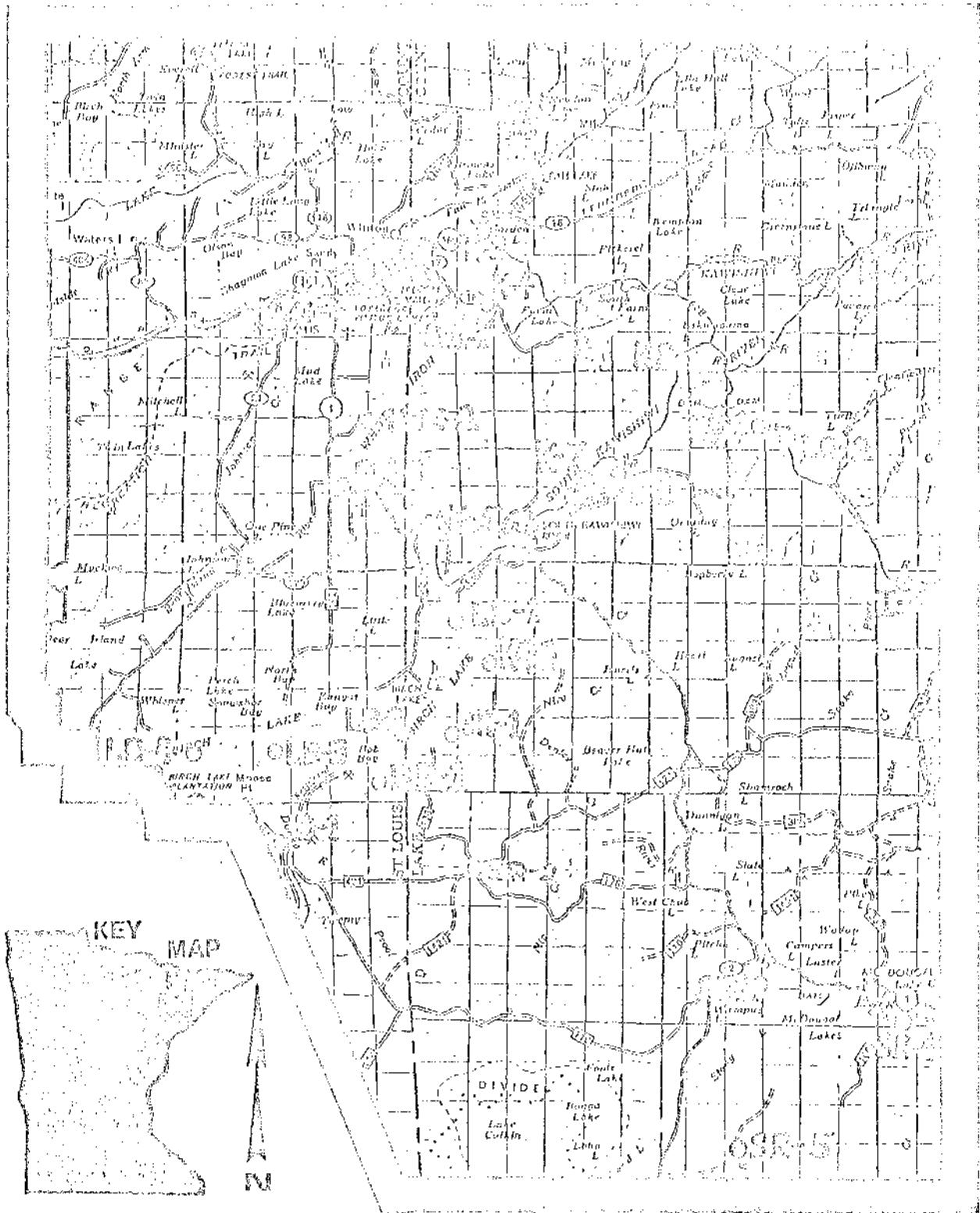
Table S-4. Frequency of Sampling: Periphyton, Benthic Invertebrates, and Macrophytes.

<u>Type of Collection</u>	RANK					
	<u>Primary</u> <u>No.</u> <u>Interval</u>	<u>Secondary</u> <u>No.</u> <u>Interval</u>	<u>Tertiary</u> <u>No.</u> <u>Interval</u>	<u>No.</u> <u>Interval</u>	<u>No.</u> <u>Interval</u>	<u>No.</u> <u>Interval</u>
Periphyton (glass slides)						
summer-fall	6	3 weeks	6	3 weeks	0	---
winter-spring	1	---	0	---	0	---
Periphyton (qualitative)						
summer-fall	4	6 weeks	4	6 weeks	2	12 weeks
winter-spring	1	---	0	---	0	---
Benthic invertebrates (Hester/Dendy)						
summer-fall	3	6 weeks	3	6 weeks	0	---
winter-spring	1	---	0	---	0	---
Benthic invertebrates (drift)						
summer-fall	4	6 weeks	3	6 weeks	0	---
winter-spring	1	---	0	---	0	---
Benthic invertebrates (qualitative)						
summer-fall	3	6 weeks	2	12 weeks	2	12 weeks
winter-spring	1	---	0	---	0	---
Macrophytes						
summer-fall	1	---	1	---	1	---
winter-spring	0	---	0	---	0	---
Heavy metals						
summer-fall	1	---	0	---	0	---
winter-spring	0	---	0	---	0	---

Table S-5. Field Sampling Schedule for Aquatic Biology--Stream Monitoring (Summer-Fall 1976).

<u>Week #</u>	<u>Sampling Schedule</u>
1 and 2	<ol style="list-style-type: none"> 1) Install samplers at stations. 2) Collect drift samples at primary stations.
3	Begin qualitative sampling at all stations.
4 and 5	<ol style="list-style-type: none"> 1) Collect periphyton from glass slides. 2) Install new glass slide samplers. 3) Do chlorophyll <u>a</u> analysis. 4) Continue qualitative sampling.
6	No sampling scheduled.
7 and 8	<ol style="list-style-type: none"> 1) Pick up Hester/Dendy samplers. 2) Collect periphyton from glass slides. 3) Install new Hester/Dendy and glass slide samplers. 4) Collect drift samples at primary and secondary stations. 5) Do chlorophyll <u>a</u> analysis.
9	<ol style="list-style-type: none"> 1) Collect qualitative samples at primary and secondary sites. 2) Begin macrophyte survey.
10 and 11	<ol style="list-style-type: none"> 1) Collect periphyton from glass slides. 2) Do chlorophyll <u>a</u> analysis. 3) Continue qualitative sampling at primary sites.
12	Complete macrophyte survey.
13 and 14	<ol style="list-style-type: none"> 1) Pick up Hester/Dendy samplers. 2) Collect periphyton from glass slides. 3) Install new Hester/Dendy and glass slide samplers. 4) Collect drift samples at primary and secondary stations. 5) Do chlorophyll <u>a</u> analysis.
15	<ol style="list-style-type: none"> 1) Begin second qualitative sampling at all stations. 2) Collect samples for heavy metals analysis.
16 and 17	<ol style="list-style-type: none"> 1) Collect periphyton from glass slides. 2) Install new glass slide samplers. 3) Do chlorophyll <u>a</u> analysis. 4) Continue qualitative sampling.
18	No sampling scheduled.
19 and 20	<ol style="list-style-type: none"> 1) Pick up Hester/Dendy samplers. 2) Collect periphyton from glass slides. 3) Collect drift samples at primary and secondary sites.

Aquatic Biology Monitoring Sites (Approximate Locations)



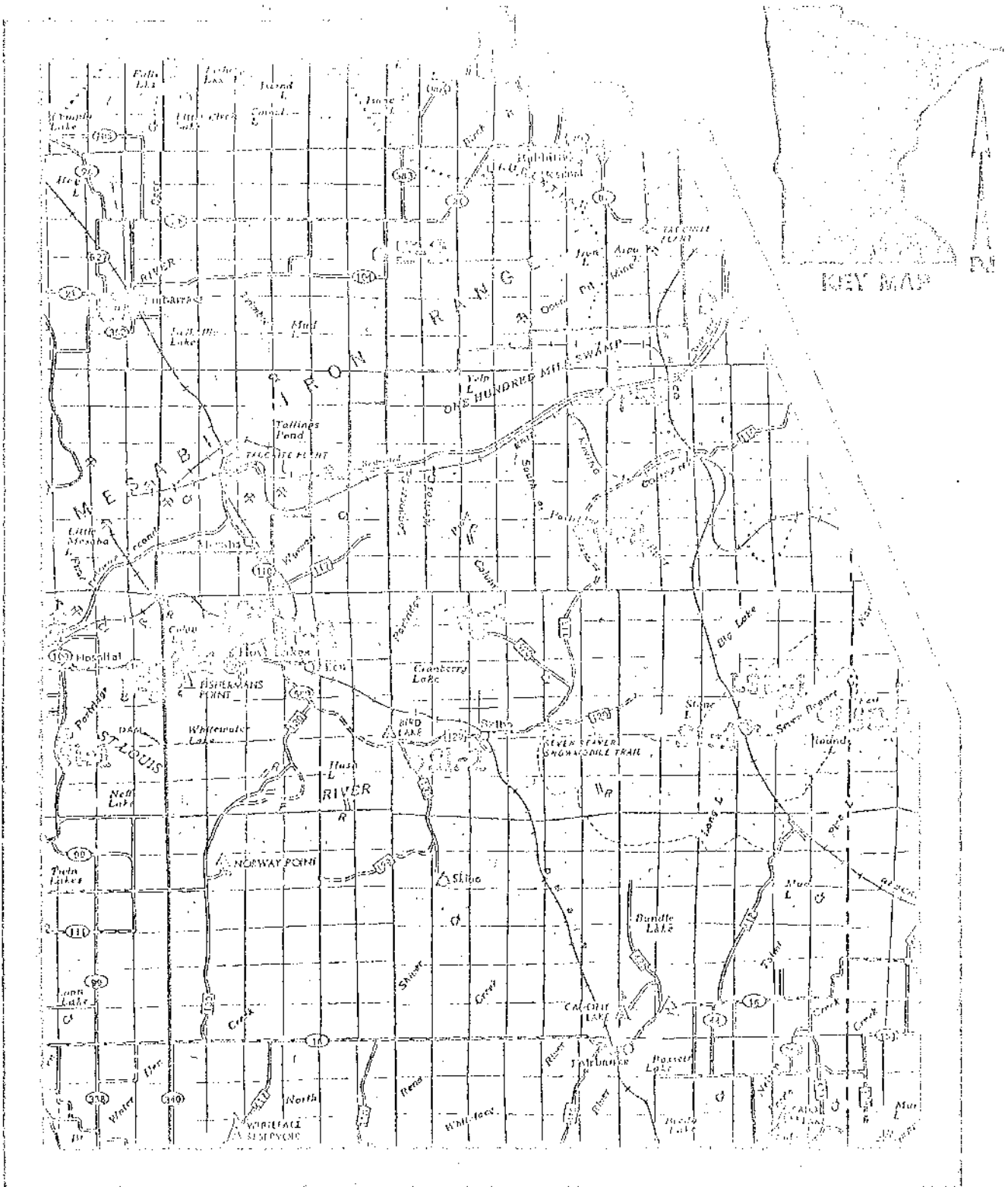
Primary
 Secondary
 Tertiary
 Lakes

1/4" = 1 Mile

0 1 2 3 4 5 6 7 8 Miles

Source: Superior National Forest Map
 Forest Service • USDA
 Duluth, Minn. 1972

Aquatic Biology Monitoring Sites (Approximate Locations)



Primary
 Secondary
 Tertiary
 Lakes

1/4" = 1 Mile



Source: Superior National Forest Map
 Forest Service · USDA
 Duluth, Minn. 1972

Appendix S-2. GENERAL FIELD AND LABORATORY PROCEDURES

PREPARATION FOR FIELD SAMPLING

- 1) Check equipment necessary for scheduled sampling.
- 2) Label all sample bottles and containers before leaving the laboratory. Labels should include collection date, sample station, type of sample, and replicate designation.
- 3) Prepare equipment for sample processing upon return from field.

FIELD SAMPLE COLLECTION

- 1) Approaching from downstream, remove artificial substrate samplers or drift nets, carefully note their condition, and log them into the field notebook. Be sure samples are placed in the correct container.
- 2) Follow prescribed methods for handling each type of sampling gear.
- 3) If applicable, attach new samplers to anchoring device.
- 4) Make the following field observations and measurements:
 - a) water temperature
 - b) air temperature
 - c) stream conditions (e.g., turbid, clear, high, low)
 - d) weather conditions (e.g., cloudy, rain, windy)
 - e) stream velocity (measured with Gurley current meter or equivalent).
- 5) Make any general observations which might aid in data interpretation.
- 6) If collecting qualitative samples, carefully note the collection time and exact collection location. Also note the various types of habitat samples and how sampled.

LABORATORY PROCEDURES

- 1) Log in all samples. All labels should be checked.
- 2) Process the samples according to prescribed procedures and store in designated areas.
- 3) When working on samples, sign them out of storage and keep them labeled as you work on them. No jars or vials should be left unlabeled.
- 4) When the sample has been completely analyzed, check it off the master list and see that all vials, slides, and data sheets relating to the sample have been correctly labeled.

Appendix S-3. FIELD AND LABORATORY METHODS--PERIOPHYTON

FIELD PROCEDURES

Sampler Installation

- 1) Suspend three slide racks containing four slides (25 x 75 mm) each, at each sample site (primary and secondary) in either a vertical or horizontal position depending on water level and siltation problems. Slide racks are identified by a letter (A through C) by facing the anchoring device and looking upstream. The sampler on the left is designated A, next to it B, and on the right C. Each slide in a slide rack has been given a numerical designation (1 through 4). These numbers are permanently enscribed on the slide rack.
- 2) Suspend the slide racks approximately 30 cm below the water surface. If the stream flow is low, the slide racks are placed just above the substrate, totally covering the slides with water.
- 3) Colonization periods are three weeks long.

Substrate Retrieval

- 1) Detach slide racks from the anchoring device.
- 2) Place into a plastic slide box, in numerical order, slide numbers 1, 3, and 4 from each slide rack. (Slide boxes were labeled in laboratory with site name and slide rack designation.) Place a little water in each slide box to prevent drying of the slides. Place the slide boxes into a cooler for transport to the field laboratory for further processing.

3) With forceps pick excess organic debris from slide 2, and place slide into a polypropylene bottle marked with the sample site and rack designation. Each bottle contains 10 ml of a 90 percent acetone solution saturated with $MgCO_3$. These bottles are also placed into the cooler for transport to the field laboratory for processing.

4) The cooler is dark inside and contains cold packs.

Collection of Qualitative Periphyton Samples

1) Collect composite samples by scraping rocks, logs, and local vegetation in riffles and pools. In addition, pipet algae found growing on silt and sand.

2) Place samples in an amber sample bottle (4 oz) containing 15-20 drops of Lugol's solution. Fill the remaining volume with distilled water. Actual sample volume should be 5-10 ml.

LABORATORY PROCEDURES

Chlorophyll a Processing

1) Scrap on both sides the slide from the polypropylene bottle using the edge of another microscope slide. Wash the residue into a 50 ml grinding tube with a small amount of acetone (90 percent $MgCO_3$ saturated). Using a small funnel to ensure minimum sample loss, rinse the solution from the polypropylene bottle into the grinding tube. Total volume should not exceed 20 ml, unless the algae growth was exceedingly dense, in which

case volume should not exceed 45 ml.

- 2) Masserate the tissue within the grinding tube for approximately one minute using a tissue grinder at high rpm.
- 3) After grinding, pour the liquid into a 25 or 50-ml volumetric flask and fill to the line with 90 percent acetone (MgCO₃ saturated).
- 4) Pour this solution into a 50 ml centrifugation tube and place into a centrifuge at 500 g for 25 min or 1000 g for 10 min.
- 5) After centrifuging the sample, measure optical density of a 4.2-ml aliquot with a spectrophotometer using the Trichromatic Method of Pigment Determination (UNESCO/SCOR), and Lorenzen Method of Phaeopigment Determination (American Public Health Association, 1976).

Preparation, Preservation, and Sample Analysis of Periphyton Species Composition

1) Preservation of Samples

- a) From the plastic slide box, preserve slide 1 as a permanent record of the existing flora on the substrate. To accomplish this, clean the side of least growth and one end of the other side. Mark the clean end with the date and code number. On the remaining area with growth, place enough Lugol's solution to cover the area. After one minute, wash off the excess Lugol's solution towards one end with distilled water, and place a small amount of CMCP-10 mounting medium on the slide. Put a large coverslip (No. 1 or 1½) over the area and allow the medium to spread out, applying

some light but steady pressure. Dry for several days and place in a slide box for storage.

b) From the plastic slide box remove slide 3. Place small funnel into a 4 oz bottle, and scrape both sides of the slide into the funnel. Wash the sample into the bottle with distilled water and add 15-20 drops of Lugol's solution; fill the remaining volume with distilled water. Secure the cap tightly and properly mark the bottle. Store the bottle in a cool dark area until analyzed.

c) From the slide box remove slide 4. Scrape both sides of the slide into a 25-ml beaker. Add 8 ml of 5 percent potassium persulfate oxidizing agent. Heat the beaker to 95°C for 30 minutes (do not boil). Cool the sample and transfer to a 50-ml centrifuge tube. Centrifuge at 1000 g for 10 minutes. Drain off the supernatant and add 25 ml of distilled water. Shake the solution, then centrifuge at 1000 g for 10 minutes. Repeat the washing process three times. After the last washing, use a disposable pipet to deliver several drops of the remaining solution onto a No. 1½ coverslip. Allow the solution to dry by placing the coverslip on a hot plate at 95°C. (Avoid overheating which will cause splattering and cross contamination.) Place a clean and appropriately marked standard microscope slide on a moderately warm hot plate (157°C) and add a drop of Hyrax mounting medium at the center of the slide. After the sample has dried, pick up the coverslip, invert, and place on top of the Hyrax drop. When bubbling ceases, remove the slide from the hot plate, and apply light pressure to the coverglass

until the Hyrax cools and hardens (one minute).

2) Preparation of Samples to be Counted

a) Shake the preserved sample solution by rotating about 30 times. Prepare for sedimentation counts by filling a 5-oz, 10-cc sedimentation chamber with the sample solution. If the sample is too dense to count, a dilution may be necessary and the appropriate dilution factor used in calculation of organisms present.

b) Fill the chambers and allow the contents to settle at the rate of four hours per 10 mm of height.

3) Sample Analysis

a) Sedimentation Counts---After settling, place the chamber on the inverted microscope and examine under 40x and 100x objectives. Strip counts are made until a minimum of 100 of the most abundant species are counted. Report data on the Periphyton Sedimentation Count bench sheet, along with other observations. Report quantitative determination of organisms as:

$$\text{number cells / mm}^2 = \frac{C \times 1000 \text{ mm}^3 \times V \times DF}{L \times W \times D \times S \times A}$$

where: C = number of cells counted (tally)
V = sample volume, ml
DF = dilution factor
L = length of a strip, mm
W = width of a strip (whipple grid image mm width)
D = depth of chamber, mm
S = number of strips counted
A = area of substrate scraped, mm²

Periphyton are placed into the following general groups during sedimentation counts:

- cocoid blue-green algae
- filamentous blue-green algae
- cocoid green
- filamentous green
- green flagellates
- other cocoid

b) Diatom Analysis Species Proportional Counts---Examine each slide with species being identified and enumerated. This procedure is done under high magnification by examining random lateral strips the width of the whipple grid, and identifying and counting all diatoms within the borders of the grid until 250 cells (500 half cells) are tallied. When the count is complete, total the tallies and calculate the percentages of the individual species. Report data on Periphyton Diatom Analysis bench sheet.

$$N = \text{percent in diatom count} \times \text{total count per mm}^2$$

Diatom species identifications, made during the proportional counts, place diatoms into the following general groups:

- dead centric diatoms
- live centric diatoms
- dead pennate diatoms
- live pennate diatoms

Qualitative Analysis

Process qualitative samples as quantitative samples, except only identifications are made.

Appendix S-4. FIELD AND LABORATORY METHODS FOR BENTHIC INVERTEBRATES

FIELD PROCEDURES: HESTER-DENDY SAMPLERS

Hester-Dendy Sampler Installation

- 1) Suspend modified Hester-Dendy samplers midway between the surface and bottom, except where the depth is greater than 2.5 meters, in which case the samplers should be suspended 1.2 meters below the surface.
- 2) Three replicate samplers are suspended vertically at all primary and secondary sites. Samplers are identified by a letter (A through C) by facing the anchoring device and looking upstream. The left-hand sampler is designated A, next to it B, and on the right C.

Hester-Dendy Sampler Retrieval

- 1) Place a dipnet under the sampler before removal from anchoring device to capture any organisms disrupted upon removal.
- 2) Detach sampler from anchoring device and place in a plastic tupperware container labeled with sample code number. Keep these containers cool until arrival at the laboratory.
- 3) Pick any organisms found in the dipnet and place in the sample container.
- 4) After the three samplers have been removed, attach new samplers to the anchoring device at applicable sites.

Hester-Dendy Sample Processing

- 1) At the laboratory dismantle the Hester-Dendy samplers and scrape the plates into a white enamel pan.
- 2) Pour the sample through a #40 standard sieve and rinse with water to remove all fine sediments.
- 3) Place the remaining sample into a bottle and preserve with five percent formalin.

FIELD PROCEDURES: DRIFT COLLECTION

Installation

- 1) Anchor draft nets with an upstream opening of 0.050 square meters, net length of 2.4 meters, a zipper 0.3 meters from the back, and 440- μ m mesh size in the stream.
- 2) Three replicate nets are used for each collection at primary and secondary sites.
- 3) Set the nets any time during the daylight hours and leave in the stream for 24 hours.

Retrieval of Nets

- 1) Remove nets from the anchoring device and rinse the contents into the bottom of the net.
- 2) Remove the end of the net at the zipper and place the contents into a

sample container identified with date and code number.

Sample Processing

- 1) At the laboratory, wash the sample through a #40 standard sieve.
- 2) Place the rinsed sample in a sample container and preserve with five percent formalin.

QUALITATIVE SAMPLING

- 1) Spend two man-hours per station collecting invertebrates from all possible habitats.
- 2) Sampling consists of kick-net samples; picking organisms from rocks, logs, debris, plants, and detritus; and dredge samples if deep areas are present. Examine riffles, pools, and backwaters.
- 3) Preserve all material collected in five percent formalin.
- 4) Note relative abundances.

LABORATORY PROCEDURES

Sample Preparation

- 1) Sieve and wash all samples to remove formalin.
- 2) Place samples in white enamel pan, pick organisms out of debris, and place in jars containing 70 percent alcohol.

- 3) If the sample size is extremely large, pick and analyze subsamples.
- 4) Preparation of Chironomid Larvae
 - a) To clear the head capsules, boil larvae in 5-10 percent KOH.
 - b) Carefully remove the head capsule from the body.
 - c) Mount both head capsule and body on a microscope slide using CMC-10 or equivalent mounting media. Head capsule should be ventral side up. Light steady pressure should be applied to the coverslip (round No. 1½) to flatten and spread the head. Allow the slide to dry overnight and store in slide box.

Sample Analysis

- 1) Identify organisms to the following levels:
Non-insects--class unless obvious (e.g., *Hyallella azteca*)
Insects--Odonata, Hemiptera, Neuroptera, Megaloptera, Coleoptera--family;
Diptera, Trichoptera, Ephemeroptera, Plecoptera--mature specimens to species where keys exist, when specimen is immature, identify to lowest possible level.
- 2) Separate all new taxa into vials for the reference collection and return remaining organisms to sample jar.

Data Handling

- 1) As the organisms in each sample are identified, record the data on invertebrate bench sheets.

Appendix S-4, Page 5.

- 2) After entire sample has been identified, record the total number of organisms and the total number of individuals of each species.
- 3) Qualitative samples are used to develop a species list only.

Appendix S-5. HEAVY METALS TISSUE ANALYSIS

COLLECTION PROCEDURE

- 1) During all procedures no metal tools or objects should come into contact with the sample.
- 2) Macrophytes
 - a) Collect plants by hand from the bottom substrate and place in plastic bags containing lake water from that location. (See Operations Manual; Aquatic Biology--Lakes for complete collection procedures.)
 - b) Each sample should contain a minimum of 20 grams wet weight of each species for heavy metals analysis and an additional specimen for identification.
 - c) All parts of the plant, roots, stems, leaves, and flowers are collected.
 - d) Place a waterproof label in each bag with standard site and date designation on it.
 - e) Samples are stored in a cooler with cool-paks until arrival at the laboratory.
 - f) At the laboratory separate the plants from each site by species. Place 20 grams wetweight of a species in a whirl-pak bag with lake or stream water from the collection site. Squeeze all air out of the bag and close it.
 - g) Assign a serial number (see Data Sheets, part 2) to each species and

include this on a waterproof label in the whirl-pak and on a tape label on the outside of the whirl-pak.

3) Periphyton

- a) Follow collection procedures outlined above for macrophytes except that individual species need not be separated. In most cases only filamentous blooms will provide enough volume to run heavy metals analysis.
- b) Preserve periphyton for identification in Lugol's as outlined in Appendix S-3.

4) Invertebrates

- a) Collection procedures outlined in Appendix S-4 are followed to obtain a representative sample of invertebrates.
- b) Teflon coated forceps and polyethylene sieve are substituted for comparable metal items.
- c) Snails, clams, leeches, crayfish, and insects are separated in the field as they are collected. Samples are placed in polypropylene bottles or polyethylene bags. As with the macrophytes, 20 grams wet weight is needed, plus specimens for identification.
- d) Use no preservatives in the field.
- e) At the laboratory, organisms are transferred to whirl-pak bags and frozen. The fleshy body is removed from the shells of clams and snails,

and the shells discarded. Whirl-pak bags contain a waterproof label with the assigned serial number.

DATA SHEETS

- 1) For each whirl-pak bag a data sheet (see Figure S5-1) must be completely filled out in duplicate.
- 2) The serial number on each data sheet is transferred to the sample labels. Serial numbers follow the format described below:

A 0000 X where A stands for Aquatic Biology, 0000 is numbered consecutively starting at 0001, and X stands for li (Macrophytes), P (Periphyton), and I (Invertebrates).

SHIPPING

- 1) Samples are packed in styrofoam coolers for shipment.
 - a) Include cool-paks with macrophyte and periphyton samples. These samples should arrive at the laboratory for processing within 24 hours after collection.
 - b) To keep invertebrate samples frozen, ship them in dry ice. Invertebrates may be stored frozen at the field station for several weeks before shipment.
- 2) Ship samples to :
Regional Copper-Nickel Study
138 Hennepin Square Building
2021 E. Hennepin Avenue
Minneapolis, MI 55413
Attention: Laura King

3) Upon arrival in the Twin Cities samples are transferred to the correct laboratory for processing and analysis.

CHAIN OF CUSTODY

1) Strict chain of custody procedures are followed throughout the entire process to ensure sample integrity.

2) A sample data form for the heavy metals tissue analysis is included as Figure S5-1. This includes a section for chain of custody.

Figure S5 1 Tissue Analysis Field Sheet.

COPPER-NICKEL REGIONAL STUDY
AQUATIC BIOLOGICAL MONITORING
TISSUE ANALYSIS

Serial No. _____

Field Data

Species _____ Date Collected _____ Station No. _____
Station Location _____
Weather Conditions _____ Temp. _____
Lake or Stream Conditions _____ Temp. _____
Substrate _____
Collection Method _____
Means of Preservation and Storage _____
Time of Collection _____ Time of Processing _____
Collected by _____
Comments _____

Laboratory Data

Arrival Date _____ Date of Analysis _____
Time Processing Begins _____ Method of Proc. _____
Analysis Method _____
Comments _____

Chain of Custody

From (Signature) Address _____ Received (Signature) Address _____ Date Sent _____ Date Rec. _____

- 1.
- 2.
- 3.
- 4.
- 5.
- 6.
- 7.
- 8.
- 9.
- 10.

Appendix 6. EQUIPMENT LIST (Does not include vehicles, boats, motors, etc.)

- 1) Wild .. 40 Inverted Microscope (no serial #)
Magnification range $\frac{10X}{90X}$, $\frac{40X}{350X}$, $\frac{100X}{900X}$ 6X eyepieces
 $150X$, $600X$, $1500X$ 10X eyepieces
- 2) Two PZO Stereoscope Microscopes
Magnification range 4X to 100X
Serial # 15052 and 15016
- 3) Beckman DU-2 Spectrophotometer, Serial # 109200-1000614
Power Supply, Serial # 331633
- 4) Clay-Adams Centrifuge, Serial # 19350
- 5) Anchoring Device (See Figure S-1)
approximately 36" high and 36" wide; 2x4 and 2x2 pine attached to
25-pound anchor--poured concrete; devise floats to provide constant
depth for samplers; various length uprights can be used also
- 6) Hester-Dendy Artificial Substrates (see Figure S-1)
4 mm-Masonite multiple plate sampler; 75 mm diameter plates;
variable spacing; 144 mm long; 0.75 m² area
- 7) Periphyton Glasslide Rack (see Figure S-1)
11 mm Plexiglass and brass; 112 mm x 115 mm; 4 slide capacity
- 8) Drift nets
0.025 m² mouth, 2.4 m length; 425 μ m mesh-niter netting
- 9) Gurley Current Meters (Full size and Pygmy)

Appendix S-7. SAMPLE LOG, DATA KEEPING, AND DATA SHEETS

Appendix S-7 includes examples of the sample log and data sheets for the Aquatic Biology, Streams program. Figure S7-1 is an example of a page from the sample log book in which all samples are logged in, and their process status followed and recorded. The "SAMPLE DESIGNATION" section includes the site (e.g., P-1, K-5); sample type (e.g., drift, periphyton); replicate (A, B, C); and date of sample. The "SAMPLE STATUS" section has blocks that are initialed and dated as a sample moves through processing. Notes relative to the sample are made under the "OTHER" column, or referred to the blank page section in the back of the log book. The "Field Collection Data" sheets are printed on waterproof paper and are numbered consecutively 1-500. Each sheet is logged, even when voided, damaged, or mutilated. The remaining sheets are bench analysis sheets for the specific samples.

Stream designations and sample type codes are given in Tables S7-1 and S7-2. Stream names are indicated by one or two-letter abbreviations. Station locations are indicated by numbers. Numbering begins at a downstream location and proceeds upstream (e.g., P-1 is the mouth of Partridge River and P-5 is the source). An example of a finished label is illustrated below:

6-1-76
SL-1-D-B

where 6-1-76 is the collection date, SL stands for St Louis River, 1 for Station 1, D for drift sample, and B for replicate B.

Appendix S-7

Table S7-1. Stream Designations.

<u>Stream Name</u>	<u>Abbrieviation</u>
Kawishiwi River	K
Filson Creek	F
Isabella River	I
Stony River	SR
Bear Island Creek	BI
Dunka River	D
Embarrass River	E
Bob Bay Creek (Unnamed Creek)	BB
Partridge River	P
St Louis River	SL
Keeley Creek	KC

Appendix S-7

Table S7-2. Sample Type Codes.

<u>Sample Type</u>	<u>Code</u>
Periphyton	P
Hester-Dendy	H
Drift	D
Phytoplankton	PP
Zooplankton	Z
Dredge	DR
Macrophyte	M
Qualitative periphyton	QP
Qualitative invertebrate	QI

Appendix S-7

Figure S7-1. Sample Log Sheet.

SAMPLE DESIGNATION				SAMPLE STATUS			
SITE	TYPE	REP	DATE	LABELED & SEALED	PICKED	PROCESSED & LOGGED	OTHER (lost, destroyed, etc.)

3a/76

Appendix S-7

Figure S7-2. Field Collection Data Sheet.

REGIONAL COPPER-NICHEL STUDY
AQUATIC BIOLOGICAL MONITORING
FIELD COLLECTION DATA

Date: Time: Station: Primary____ Secondary____ Tertiary____

Water Temp.: Air Temp.: Weather Conditions:

Stream Conditions: Stream Velocity:

Periphyton Sampler P-A:
 P-B:
 P-C:

Hester-Dendy Sampler H-A:
 H-B:
 H-C:

Drift Samplers D-A:
 D-B:
 D-C:

Periphyton Qualitative:

Invertebrate Qualitative:

General Comments:

PERIPHYTON SEDGWICK-RATHEE COUNT

River or Lake _____ Inclusive Dates _____
Station _____ Date Analyzed _____
State _____ Analyzed by _____

Table with columns: CODE, ORGANISM, Tally, μmm^2 . Rows include categories like Total coccoid blue-green algae, Total filamentous blue-green algae, Total coccoid green algae, Total filamentous green algae, Total green flagellates, Other coccoid algae, Other pigmented flagellates, Filamentous bacteria and fungi, and Protozoa.

Centrics μmm^2
Pennates μmm^2

Diatoms
Centric Shells, Live Centrics
Pennate Shells, Live Pennates

Preservative _____ S - R Factor _____ TOTAL (cells/ μmm^2)
No. Slides _____
Area Scraped _____ Remarks: _____
Scraps diluted to _____ mL
First check _____ Recorded _____

PLANKTON AND PERIPLHYTON
CHLOROPHYLL AND BIOMASS DATA

I. IDENTIFYING INFORMATION:

A. Station: _____

B. Date: _____

C. Method of Sample Collection and Handling: _____

II. SPECTROPHOTOMETRY DATA:

A. OPTICAL DENSITY MEASUREMENTS:

Instrument Used: _____

Rep.	Extract Volume	Dilution Factor	Optical Density Readings					663 b/a
			750	663b*	645	630	663a*	
1.	_____	_____	_____	_____	_____	_____	_____	
2.	_____	_____	_____	_____	_____	_____	_____	
3.	_____	_____	_____	_____	_____	_____	_____	
4.	_____	_____	_____	_____	_____	_____	_____	

* (b - before acidification; a = after acidification)

B. CHLOROPHYLL CALCULATIONS:

Rep.	Concentration of Chlorophyll in Extract (mg/l)			Sample area or volume (liters/m ³)	Chlorophyll content of sample (ug/l; mg/m ³)		
	Chl a	Chl b	Chl c		Chl a	Chl b	Chl c
1.	_____	_____	_____	_____	_____	_____	_____
2.	_____	_____	_____	_____	_____	_____	_____
3.	_____	_____	_____	_____	_____	_____	_____
4.	_____	_____	_____	_____	_____	_____	_____

III. FLUOROMETER DATA:

Instrument Used: _____

Rep.	Dilution Factor	Reading Before (b) Acidification		Reading After (a) Acidification		
		Reading R _b	Sens. Level (S)	R _a	(S)	R _a /R _b
1.	_____	_____	_____	_____	_____	_____
2.	_____	_____	_____	_____	_____	_____
3.	_____	_____	_____	_____	_____	_____
4.	_____	_____	_____	_____	_____	_____

IV. ORGANIC MATTER (ASH-FREE WEIGHT)

Rep.	Grc. No.	Empty Crucible Weight (A)	Weight with Dry Sample (B)	Weight After Firing	Sample Dry Weight	Ash Free Weight (B-C)	Organic Matter (pm/m ³)
1.	_____	_____	_____	_____	_____	_____	_____
2.	_____	_____	_____	_____	_____	_____	_____
3.	_____	_____	_____	_____	_____	_____	_____
4.	_____	_____	_____	_____	_____	_____	_____

V. REMARKS:

Appendix S-8. STAFF RESUMES

Resumes for the Aquatic Biology staff appear in this appendix. Included are resumes for Mark Johnson, Michael Mischuk, Thomas Lager, Steven Williams, and Jeffrey McCulloch. Figure S8-1 shows the relationship of the staff to each other.

RESUME FOR MARK D JOHNSON, SUPERVISOR, AQUATIC BIOLOGY STUDY GROUP
July 7, 1976

Home Address: 2344 Como Avenue Box 3268, Route 1
St Paul, MN 55108 or Ely, MN 55731

Office Address: Department of Natural Resources Kawishiwi Field Lab
Minerals Division P O Box 569
345 Centennial Building or Ely, MN 55731
St Paul, MN 55155 (218) 365-5034

Present Duties--Responsible for design and implementation of aquatic biology monitoring program.

Work Experience

- June 1975 - Field Biologist, Northern States Power Company. Worked
October 1975 on fish entrainment project at the Prairie Island Nuclear
Power Plant. Also participated in regular biological
monitoring programs studying the Mississippi River in
the vicinity of the plant.
- February 1975 - Research Biologist, Ecology Consultants, Inc, Ft Collins,
June 1975 Colorado. Participated in various environmental mon-
itoring programs and studies related to the power industry.
- March 1973 - Senior Biologist, Minnesota Pollution Control Agency.
December 1974 Participated in collection of baseline data in Voyageurs
National Park (1974), review of environmental impact
statements (1973, 1974), review of monitoring programs
at Northern States Power Company's nuclear power stations
(1973, 1974), study on the effects of pollution on
lichens along the north shore of Lake Superior (1973),
study on the effect of taconite tailings on periphyton
growth in Lake Superior (1973), and was a member of
the Henderson Power Plant EIS team (1974).
- July 1971 - Research Assistant, Colorado State University, Ft Collins,
December 1972 Colorado. Collected baseline aquatic biological data
at the site of the Ft St Vrain Nuclear Power Station
for use in the environmental impact statement. Partici-
pated in design of the environmental monitoring program
in the vicinity of the Ft St Vrain station.

RESUME FOR MARK D JOHNSON (contd)

Work Experience (contd)

June 1972 - Consultant, ROMCOE, Denver, Colorado. Participated
September 1972 in fish survey conducted for the preparation of an
environmental impact statement related to a new ski
resort development near Vail, Colorado.

Education

Master of Science, Department of Fishery and Wildlife Biology, Colorado
State University, March 1973. Thesis entitled, "Aquatic Biota of the
South Platte River and St Vrain Creek in the vicinity of the Ft St Vrain
Nuclear Generating Station."

Bachelor of Science, College of Biological Sciences, University of Minnesota,
St Paul, June 1971.

Also attended Environmental Protection Agency Water Training Course,
"Bioassay in Pollution Control and Analysis," Cincinnati, Ohio, October
1973.

Professional Affiliations

Member, American Fisheries Society

Member, Phi Kappa Phi

Publications

Everhart, W H and M D Johnson. 1971. Aquatic biota sampling program.
Pages 8-24 in Environmental Radiation Surveillance Program for the
Public Service Company of Colorado, Quarterly Report, July 1, 1971
- September 30, 1971, Ft St Vrain Nuclear Generating Station. Edited
by K J Schiager and S T Bard. Colorado State University, Ft Collins,
Colorado.

Ob cit. 1972. Aquatic biota sampling program. Pages 2-27 in Environmental
Radiation Surveillance Program for the Public Service Company of
Colorado, Annual Report, January 1, 1971 - December 31, 1971. Edited
by K J Schiager and S T Bard. Colorado State University, Ft Collins,
Colorado.

RESUME FOR MARK D JOHNSON (contd)

Publications (contd)

Ob cit. 1972. Aquatic biota sampling program. Pages 11-15 in Environmental Radiation Surveillance Program for the Public Service Company of Colorado, 1st and 2nd Quarterly Reports, January 1, 1972 - June 30, 1972. Edited by K J Schiager and S T Bard. Colorado State University, Ft Collins, Colorado.

RESUME FOR MICHAEL MISCHUK, AQUATIC BIOLOGIST, NATURAL RESOURCE
SPECIALIST III, June 23, 1976

Home Address: 1551 Holton Street
St Paul, MN 55108
(612) 644-5954

Office Address: Kawishiwi Field Lab
P O Box 569
Ely, MN 55731
(218) 365-5034

Present Duties--Responsible for periphyton collection, periphyton taxonomy, chlorophyll analysis, and field coordination problems for the Copper-Nickel aquatic biology program.

Work Experience

1972-1976

Graduate Assistant, St Cloud State University.

- 1) Assisted Dr Keith M Knutson in field investigation of algae populations in conjunction with the ecological monitoring program for Northern States Power Company's Monticello Nuclear Plant located on the Mississippi River. Responsibilities included sample collection, chlorophyll analysis and algae identification and enumeration.
- 2) Assisted in fish entrainment studies, both natural and induced. Responsibilities included equipment design and field sampling.
- 3) In September 1973, designed and started project "Primary Production on the Mississippi River at Monticello, Minnesota." Objectives of this study were to determine differences, if any, between the heated and ambient zones of the river. The study consisted of an upstream-downstream evaluation of the effects of increased temperatures on chemical and biological parameters. This work was done as part of requirements for a master's thesis.
- 4) Assisted Dr Knutson in sampling and subsequent identification of algae in the Escambia River, Pensacola, Florida for Gulf Power Company, in the vicinity of their electric generating plant (November, 1973).
- 5) Assisted in algae identification and enumeration of species taken from Lake Ontario, for NUS Corporation.
- 6) Assisted in an evaluation and update study of chemical and biological parameters of Clearwater Lake and Clearwater River system (Minnesota) for testimony given

RESUME FOR MICHAEL MISCHEK (contd)

Work Experience (contd)

- 1972-1976
(contd) before the Minnesota Water Resources Board on a proposed watershed district.
- Summer -
Fall 1972 Assistant to Mr George V Anderson. Worked with Mr Anderson on the limnology of Big Fish Lake in Stearns County, Minnesota. Assisted on removal and subsequent identification and mapping of aquatic vegetation around the lake. In the fall, assisted on age and growth work on fish taken from the lake.
- April 1972 Internship, National Water Quality Lab, Duluth, Minnesota. Assigned to an AL-6 team, Environmental Requirements of Fish, under the direction of Mr John H McCormick, principal investigator, and Mr Bernard A Jones, team coordinator. The objective of the study "Thermal Tolerance of the American Smelt, Osmerus mordax" was the evaluation of thermal tolerance of the three obligatory shallow water life stages of the American Smelt during the reproductive period. Responsibilities included: selection of appropriate test temperatures; equipment set up; compilation and evaluation of data; and a formal report.
- Fall 1971 -
Spring 1972 Assistant to Mr Robert Anderson. Studied the feeding habits of the shorthead redhorse, Moxostoma macrolepidotum (Lesueur), and the carp, Cyprinus Carpio L., which were collected near the Monticello Nuclear Power Plant. Assisted in field capture of specimens by electrofishing. After removal of the stomachs, identification of the contents were made, which consisted mainly of aquatic insects.
- Summer 1971 Studied productivity of Lake Sylvia in Wright County, Big Watab Lake in Stearns County, and East Quarry, an abandoned granite quarry near St Cloud State College. Project design and sampling were responsibilities.

Also have certificate as a SCUBA diver and hold senior diving certificate. Used this skill when doing fish surveys and general river work for Northern States Power Company.

RESUME FOR MICHAEL MTSCHNIK (cont'd)

Military Service

1967 - present Military Service, United States Army Reserves, Assistant Operations Sergeant. Basic training was at Fort Knox, Kentucky from November 1967 to April 1968. Received advanced individual training in communications, which consisted of radio mechanics school, vehicular and individual land wiring. Returned to Fort Snelling, Minnesota as a reservist in April 1968. Duties consisted of: 1) Field Radio Mechanic (first echelon repair of radios within a battalion); 2) Section Chief (supervised ten people in radio repair, radio installation, and installation of telephone land lines); 3) Communications Chief (primary duty, Platoon Sergeant, supervised 20 people, required to write training schedules for own personnel plus those within battalion requiring the same training, oversaw maintenance of all communications equipment within battalion, plus wrote and submitted status reports on equipment and training); 4) Other (served as instructor for Noncommissioned Officers Academy, which included writing training schedules, giving instruction, and evaluating status of students). Present duties (as Assistant Operations Sergeant) include assisting the writing of tactical plans, recording these plans in journals, and maintaining records of training status.

Education

Master of Arts with major in aquatic biology and emphasis on power plant ecology, St Cloud State University, 1976.

Bachelor of Arts with major in biology and special emphasis on aquatic studies, St Cloud State University (College), 1972.

RESUME F. L. THOMAS MICHAEL FAGER, AQUATIC BIOLOGIST, NATURAL RESOURCE
SPECIALIST III, June 23, 1976

Home Address: P O Box 524
Ely, MN 55731

Office Address: Kawishiwi Field Lab
P O Box 569
Ely, MN 55731
(218) 365-5034

Present Duties:--Responsible for invertebrate collection, invertebrate taxonomy, and qualitative sampling for the Copper-Nickel aquatic biology program.

Work Experience

- 1973-1975 Graduate Research Assistant at St Cloud State University on Northern States Power Company contract. Determined the effects of the heated water discharge from the NSP Monticello Nuclear Generating Plant on the macro-invertebrate populations in the Mississippi River. Duties included assisting in annual report writing; assisting with fish mark and recapture studies, artificial fish egg substrate studies, electro-fishing, seining, gill and trap netting, and taxonomy of fishes; and assisting with invertebrate drift, taxonomy, and algae studies. Supervised two to four assistants in the field and laboratory.
- 1974 Independent Research Project, St Cloud State University. Determined the effect of rapid temperature fluctuations on the respiratory rates of the stonefly, Paragnetina sp., utilizing a Gilson Respirometer.
- 1972 Summer Research Program, University of Minnesota Limnological Laboratory on Lake Superior. Duties included qualitative and quantitative analysis of lotic periphyton (algae) and quantitative analysis of chlorophyll; and studied nutrient uptake by the aquatic plant, Ceratophyllum demersum, in controlled artificial environments.

RESUME FOR THOMAS MICHAEL LAGER (contd)

Work Experience (contd)

1971-1972 Bemidji State University. Assisted in a lake shore survey to determine the influence of domestic pollutants on the water quality of lakes. Determined the temperature effects on the photosynthetic and respiration rates of plankton using a respirometer.

Education

Master of Arts in biology with emphasis on aquatic ecology and limnology (35 credits), St Cloud State University, 1976. Research for master's degree included the determination of effects of the heated water discharge from the NSP nuclear generating plant on the macroinvertebrate populations in the Mississippi River. The population quality, quantity, biomass, weight per organism, and life cycles were statistically analyzed between the heated and nonheated portions of the river and correlated with the preoperational study.

Summer institute work, University of Minnesota, Minneapolis, June-August 1972. "Water Quality Investigation and Research Techniques." General introduction to a wide variety of analytical techniques for the evaluation of chemical and biological parameters (12 credits).

Bachelor of Arts with major in biology and minor in chemistry, Bemidji State University, 1972. Emphasis was in aquatic ecology and limnology (22 credits).

Associate of Arts with major in biology and minor in chemistry, Rainy River State Junior College, International Falls, Minnesota, 1969.

Professional Affiliations and Journals

Member, North American Benthological Society

Member, Entomological Society of Washington

Member, The Ecological Society of America

Eatonia, A Newsletter for Ephemeropterists

RESUME FOR STEVEN N WILLIAMS, AQUATIC BIOLOGIST, NATURAL RESOURCES
SPECIALIST 1, June 23, 1976

Home Address: P O Box 569
Ely, MN 55731

Office Address: Kawishiwi Field Lab
P O Box 569
Ely, MN 55731
(218) 365-5034

Present Duties--Responsible for macrophyte collection, equipment maintenance, and sample processing for the Copper-Nickel aquatic biology program. Assist in invertebrate sample collection.

Work Experience

- April 1975 - September 1975 Colorado Division of Wildlife. Conducted initial six-month segment of five-year creel census on Shadow Mountain Lake and Lake Granby. Study was designed to evaluate angling pressure, success ratio, angler harvest, and success of stocking programs.
- June 1974 - March 1975 Ecology Consultants Incorporated. Duties included collection, identification, quantification, and biomass analysis of benthic macroinvertebrates. Project work included identification of insects from Minnesota (Northern States Power), Missouri (Kansas City Power and Light), Wyoming (Missouri Basin Power), Oklahoma (Public Service of Oklahoma), and Colorado (Sun Oil). Duties also included identification, quantification, impact assessment, and stomach analysis on fish populations, with particular emphasis on large river systems.
- 1972 - 1973 Colorado State University. Training included work with doctoral candidate in culture of northern pike, population assessment of fish in small farm ponds, and effects of northern pike introductions.
- 1972 Colorado Division of Wildlife. Work requirement in fulfillment of degree in Fishery Biology. Major emphasis placed on general survey of fish populations in the Colorado River within the State of Colorado. Work also included assessment of fish populations in both high

RESUME FOR STEVEN N WILLIAMS (contd)

Work Experience (contd)

1972 (contd) mountain lakes and man-made reservoirs in relation to setting minimum stream flows.

Education

Bachelor of Science with major in biological science, Colorado State University, Fort Collins, Colorado, 1974.

Associate of Arts and Science with major in preprofessional forestry, Paul Smith's College, New York, 1971.

RESUME FOR JEFFREY LEE MCCULLOCH, AQUATIC BIOLOGIST, NATURAL RESOURCES
SPECIALIST I, June 23, 1976

Home Address: 220 E Sheridan Street
Ely, MN 55731

Office Address: Kawishiwi Field Lab
P O Box 569
Ely, MN 55731
(218) 365-5034

Present Duties: Responsible for heavy metal sample collection, equipment inventory, and sample processing for the Copper-Nickel aquatic biology program. Assist in periphyton collection and processing.

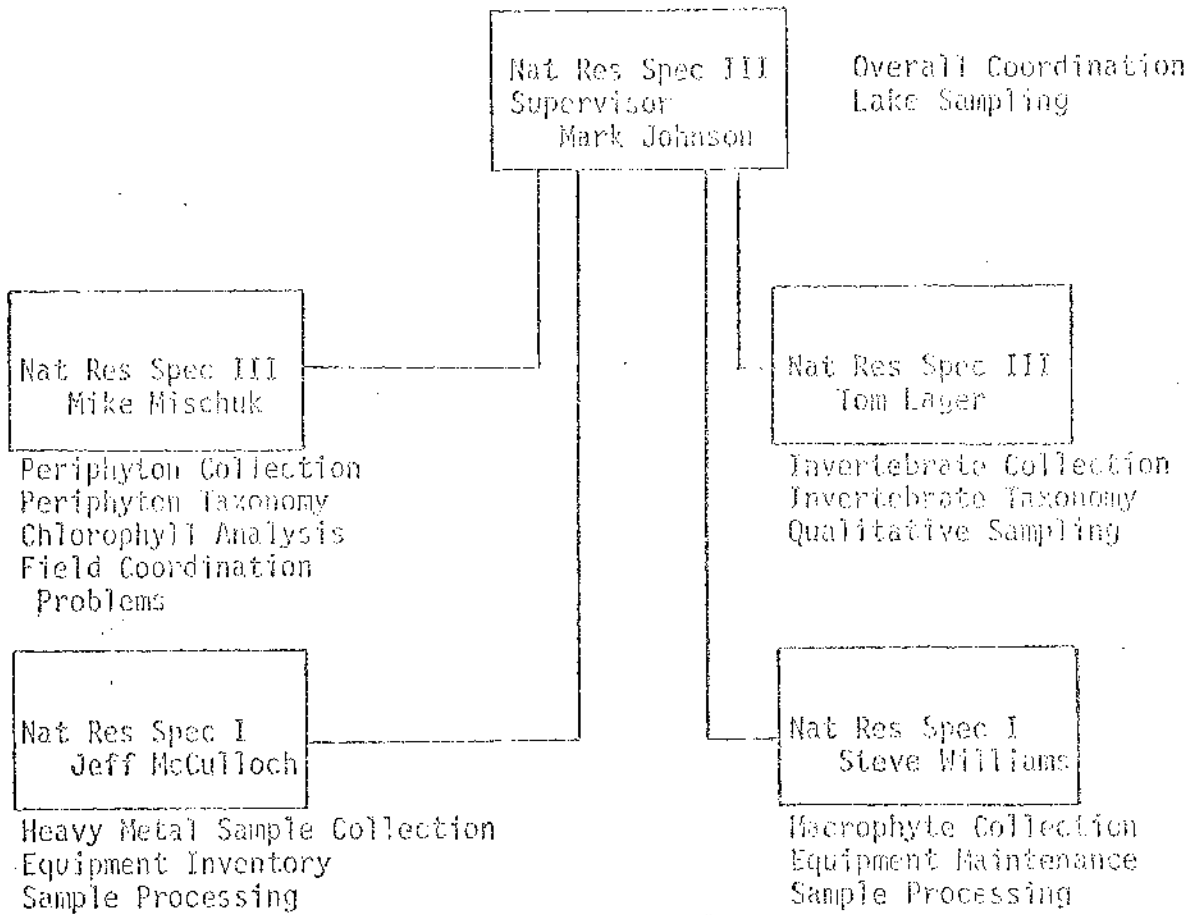
Work Experience

January 1975 - Research Biological Aid, U.S. Environmental Protection
March 1976 Agency, Environmental Research Laboratory-Duluth.
Worked as a technician on the heavy metals and pesticides research teams under the supervision of a research aquatic biologist and a research entomologist. Duties included planning, constructing, and maintaining bioassay systems for fish and aquatic invertebrates, aiding in the conduction of these bioassays to determine toxicant effects, performing routine analysis of water for hardness, alkalinity, acidity, pH, dissolved oxygen, etc, and keeping the records of routine and specific biological and chemical data. Duties also included the operation of a stock culture unit for raising and rearing the laboratory fish Jordanella floridae. In addition, time was spent participating in field collections for both fishes and aquatic invertebrates, collection and review of literature concerning insect drift, design and construction of drift nets, and initial field work with the nets. Experience was gained in the taxonomy of the regional aquatic fauna, and use of abstract journals, scientific journals, literature retrieval systems, books, etc.

Education

Bachelor of Arts with major in biology, University of Minnesota-Duluth, June 1976. Upper division emphasis in fields of entomology, ichthyology, limnology, vertebrate and invertebrate anatomy, animal behavior, and physiology.

Figure SB-1. AQUATIC BIOLOGICAL MONITORING STAFF ASSIGNMENTS



Appendix 5-9. TAXONOMIC REFERENCES.

Zooplankton

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- Brooks, J.L. 1957. The systematics of North American Daphnia. *Mem. Conn. Acad. Arts Sci.* 13:1-180.
- Brooks, J.L. 1959. Cladocera. Pages 587-656 in W.T. Edmondson, editor, *Freshwater Biology*. John Wiley and Sons, New York and London.
- Chengalath, R., C.H. Fernando, and M.G. George. 1971. The Planktonic Rotifera of Ontario with keys to genera and species. *Univ. of Waterloo Biology Series, No. 2*. Waterloo, Ontario.
- Edmondson, W.T. 1959. Rotifers. Pages 420-491 in W.T. Edmondson, editor, *Freshwater Biology*. John Wiley and Sons, New York and London.
- Frey, D.G. 1959. The taxonomic and phylogenetic significance of the head pores of the Chydoridae (Cladocera). *Int. Revue ges Hydrobiol.* 44:27-50.
- Gouldon, C.E. 1968. The systematics and evolution of the Moinidae. *Trans. Amer. Phil. Soc.* 58(6):1-101.
- Megard, R.O. 1967. Three new species of Alona (Cladocera, Chydoridae) from the United States. *Int. Revue ges Hydrobiol.* 52(1):37-50.
- Ruttner-Kolisko, A. 1974. Plankton rotifers. Biology and taxonomy. in H.J. Elster and W. Ohle, editors, *Die Binnengewasser*. Volume XXVI. Suppl. E. Schweizerbart'sche Verlagbuchhandlung, Stuttgart, West Germany.
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- Wilson, N.S. 1959. Free-living Copepoda: Calanoida. Pages 738-794 in W.T. Edmondson, editor, *Freshwater Biology*. John Wiley and Sons, New York and London.
- Yeatman, H.C. 1959. Free-living Copepoda: Cyclopoida. Pages 795-815 in W.T. Edmondson, editor, *Freshwater Biology*. John Wiley and Sons, New York and London.

Phytoplankton

- Bourelly, P. 1966-1972. Les algues d'eau douce. Tome I-III. Roubee and Cie, Paris, France. pp. 569, 438, 505.
- Drouet, F., and W.A. Daily. 1956. Revision of the coccoid Myxophyceae. Butler University Botanical Studies XII, Indianapolis, Indiana.
- Fott, B. 1959. Algenkunde. Gustav Fischer Verlag, Stuttgart, West Germany.
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- Hustedt, F. 1959. Die Kieselalgen. L. Rabenhorst, editor, Kryptogamen-Flora von Deutschland, Österreich, und der Schweiz. Volume VII, Part 2. Akademische Verlagsgesellschaft m. b. h. Leipzig.
- Hustedt, F. 1959-1966. Die Kieselalgen. L. Rabenhorst, editor, Kryptogamen-Flora von Deutschland, Österreich, und der Schweiz. Volume VII, Part 3. Akademische Verlagsgesellschaft m. b. h. Leipzig.
- Patrick, Ruth, and Charles Reimer. 1966. Diatoms of the United States. Volume I. Monographs of the Academy of Natural Sciences of Philadelphia, Number 13.
- Prescott, G.W. 1962. Algae of the Western Great Lakes Area. Wm. C. Brown Co., Dubuque, Iowa.
- Prescott, G.W. 1970. How to Know the Freshwater Algae, 2nd ed. Wm. C. Brown Co., Dubuque, Iowa.
- Randhawa, M.S. 1959. Zygnemaceae. Indian Council of Agricultural Research. New Delhi, India.
- Smith, G.M. 1920. Phytoplankton of the Inland Lakes of Wisconsin. Parts I and II. Wisconsin Geological and Natural History Survey, Bulletin #57. pp. 243, 227.
- Smith, G.M. 1950. The Fresh Water Algae of the United States. McGraw-Hill Book Co., New York.
- Taft, Clarence E., and Celeste W. Taft. 1971. The Algae of Western Lake Erie. Ohio Biological Survey. New Series 4(1):1-189.
- Tiffany, L.H., and M.E. Britton. 1952. The Algae of Illinois. Hafner Publishing Company, New York. (Reprint edition 1971.)

Phytoplankton (contd.)

- Weber, Cornelius I. 1971. A Guide to the Common Diatoms at Water Pollution Surveillance Systems Stations. USEPA-MERC Analytical Quality Control Laboratory, Cincinnati, Ohio.
- West, G.S., and F.E. Fritch. 1927. A Treatise on the British Freshwater Algae. Cambridge University Press.
- Whitford, L.A., and G.J. Schumacher. 1973. A Manual of Freshwater Algae. Aparks Press, Raleigh, North Carolina.

Chironomidae

- Beck, W.M., Jr. 1968. Chironomidae. Keys to Water Quality Indicative Organisms of the Southeast United States. FWPCA, Atlanta, Georgia.
- Darby, R.E. 1962. Midges associated with California rice fields with special reference to their ecology, (Diptera: Chironomidae). Hilgardia 32:1-206.
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- Mason, W.T., Jr. 1968. An Introduction to the Identification of Chironomid Larvae. FWPCA, Cincinnati, Ohio.

Trichoptera

- Elkins, W.A. 1936. The immature stages of some Minnesota Trichoptera. Ann. Entomol. Soc. Amer. 29:656-681.
- Flint, O.S., Jr. 1960. Taxonomy and biology of nearctic limnephelid larvae (Trichoptera), with special reference to species in eastern United States. Entomol. Amer. 40:1-117.
- Flint, O.S., Jr. 1962. Larvae of the caddisfly genus Rhyacophila in eastern North America (Trichoptera: Rhyacophilidae). Proc. U.S. National Museum 113:465-493.
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Hemiptera

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- Hilsenhoff, W.L. 1970. Corixidae (Water Boatmen) of Wisconsin. Wis. Acad. Sci. Arts and Letters. 58:203-235.
- Hilsenhoff, W.L. 1975. Aquatic Insects of Wisconsin. Technical Bull. #89. Wis. Dept. Natur. Resources.
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- Usinger, R.L. 1956. Aquatic Hemiptera. Pages 182-228 in R.L. Usinger, editor, Aquatic Insects of California. University of California Press, Berkeley, California.

Coleoptera

- Brown, H.P. 1970. Aquatic dryopoid beetles (Coleoptera) of the United States. Identification Manual No. 6, Water Pollution Control Research Series 18050 ELD04/72. U.S. Environmental Protection Agency.
- Collier, J.E. 1969. A taxonomic revision of the genus *Oplioservus* (Coleoptera: Elmidae) in the nearctic region. Ph.D. Thesis, University of Minnesota.

Coleoptera (contd.)

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- Hilsenhoff, W.L. 1975. Aquatic Insects of Wisconsin. Technical Bull. #89. Wis. Dept. Natur. Resources.
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Neuroptera

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Ephemeroptera (Mayflies)

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1. INTRODUCTION AND SCOPE

1.1. Introduction

The regional Copper-Nickel Study was designed to provide environmental data in the potential copper-nickel development area. These data can be used first to characterize the region and analyze the potential environmental impact of copper-nickel mining, and secondly, as development proceeds, to determine the actual regional impact.

Because of time limitations, it was not feasible to biologically monitor all lakes for the regional study. Instead, a small number of lakes were selected for sampling based on a combination of factors including the probability of impact from various sources and consideration of how characteristic the lake was for the region in terms of fish populations, lake morphometry, watershed, lake chemistry, and access.

1.2. Scope of Study

Lake sampling included quantitative phytoplankton, zooplankton, and benthic invertebrate sampling, and qualitative benthic invertebrate and macrophyte collection. In lakes, phytoplankton are the principal primary producers. Taxonomic analysis and chlorophyll a measurements of the phytoplankton populations provide measures of a lake's trophic status. Zooplankton and benthic invertebrates link the primary producers to fish. Study of these components provides information on the availability of food for fish and on the "health" of the lake. Heavy metals

levels can be monitored through the study of macrophytes and clams, both heavy metals accumulators.

2. STUDY AREA AND SAMPLING STATIONS

2.1. Study Area

See Operations Manual; Aquatic Biology--Streams for description of study area.

2.2. Sampling Stations

Five lakes were selected for biological sampling (Table L-1). Two stations were located on each lake, except Birch Lake where there were four stations. Stations were located in the approximate center of each lake: one at the inflow end and one at the outflow end. In Birch Lake, stations were located west of Dunka Bay, north of Dunka Bay, between Bob Bay and Stony River, and south of the South Kawishiwi River.

2.2.1. Regional Maps

A regional map showing locations of sampling sites appears in Appendix S-1.

3. METHODOLOGY

3.1. Techniques

3.1.1. Justification of Techniques

Because of time constraints, it was impossible to sample lakes as thoroughly

as necessary to produce reliable quantitative data. As a result, the decision was made to sample in such a way that the greatest variety of species would be collected with the least effort.

To sample phytoplankton, an integrated water sampler was developed based on a sampler constructed by the Minnesota Pollution Control Agency. This sampler was constructed of 1.5-inch diameter PVC pipe. The pipe was in 2-m sections and had the capability of sampling to a depth of 4 m. Each end of the pipe was threaded; so the pipe could be capped. The 4-m length was used to assure that the entire photic zone was sampled. Because various phytoplankton species reside at different levels within the photic zone, we were able to collect one water sample containing virtually all phytoplankton species. The chlorophyll *a* value obtained in this way was also a composite sample.

Zooplankton were sampled by towing a Wisconsin net from the lake bottom to the surface. Again, a single sample collected species found at various depths.

A petite Ponar or Ekman dredge was employed for benthic invertebrate collection. Two basic considerations were made before choosing these dredges. First a light-weight dredge was needed because much of the sampling was done from a canoe, and secondly most of the bottom sediments at the sampling stations were silt. To reduce variability, dredge sampling was done outside the littoral zone.

Qualitative invertebrate sampling similar to stream qualitative sampling

provided information on the benthic fauna of the littoral zone. Qualitative collections also provided information on snail and clam populations which are particularly sensitive to heavy metals.

SCUBA was chosen for macrophyte collection to assure that roots, stems, leaves, and flowers were collected, and to assure that all species along the selected transect were collected. Macrophyte mapping was not attempted because of time constraints, and because macrophyte surveys were conducted during previous Minnesota Department of Natural Resources lake surveys.

3.1.2. Procedures

3.1.2.1. Phytoplankton

Phytoplankton samples were collected by lowering vertically the integrated sampler with both ends open in the water, until the upper end extended two to three inches above the surface. The top was then capped and the sampler raised until the lower end extended two to three inches below the water surface. The lower end was then capped and the sampler removed from the water. Water in the sampler was drained into an 8-liter carboy and mixed. Three 1-liter samples were withdrawn from this mixed-water sample. These samples were kept in a cooler with cool-paks until arrival at the laboratory.

Each 1-liter water sample was filtered through a 0.45 μ m Gelman Type A

glass fiber filter. These filters were then frozen in polypropylene bottles containing 10 ml of acetone until final analysis. Actual chlorophyll determinations were made following the procedures outlined below.

- 1) Masserate the glass fiber filter within the grinding tube for approximately 1 min using a tissue grinder at high rpm.
- 2) After grinding, pour the liquid into a 25 or 50-ml volumetric flask and fill to the line with 90 percent acetone (MgCO_3 sat.).
- 3) Pour this solution into a 50-ml centrifugation tube and place in a centrifuge at 500 gs for 25 min or 1000 gs for 10 min.
- 4) After centrifuging the sample, measure a 4.2-ml aliquot with a spectrophotometer using the Trichromatic Method of Pigment Determination (URESCO/SCOR), and Lorenzen Method of Phacopigment determination (American Public Health Association, 1971).
- 5) Enter results on periphyton and plankton pigment and biomass bench sheets.

In addition to the three 1-liter samples withdrawn for chlorophyll, three 120-ml samples were withdrawn from the integrated water sample at each station for taxonomic analysis. These samples were preserved with Lugol's solution in amber bottles and stored in the dark until analyzed. Taxonomic analysis of the phytoplankton samples was done by Ecology Consultants, Inc. (ECI), Fort Collins, Colorado, following the technique described by Utermoel (1958) and outlined by Vollenweider (1974). The ECI analysis procedures follow.

Taxonomic Analysis of Phytoplankton Samples

1) Sedimentation--Samples are shaken vigorously to uniformly distribute the phytoplankton organisms. A 25 to 100-ml aliquot is withdrawn from each sample (depending upon phytoplankton density), and introduced into a plankton settling cylinder situated upon a combined plate chamber (Wild Heerbrugg, Ltd., 1975). Samples remain undisturbed at room temperature for a minimum of 24 hours. Following sedimentation, cylinders are moved laterally to a drain hole and emptied into a beaker, leaving the sediment in the recessed plate chamber. The supernatant is examined and the occurrence of any buoyant species noted.

2) Identification--Plate chambers containing sedimented material are examined with a Wild M-40 inverted microscope at magnifications of 56X, 140X, 280X, 560X, and 1400X. Standard phyecological references are utilized for taxonomic determinations (see Appendix S-9).

Diatoms, which usually account for a substantial proportion of the phytoplankton populations, are identified from permanent Hyrax slide mounts. Duplicate mounts are prepared from each lake sample during each sampling period to assure that acceptable material is available for analysis. Mounts are made from small aliquots taken from the sedimented material and composited in a 15-ml test tube, and subjected to sulfuric acid-potassium dichromate digestion. Following cooling, samples are centrifuged, an aspirator used to remove the supernatant, and frustules resuspended in distilled water. This is repeated several times until a clear solution with approximately neutral pH remains. One or two drops of the diatom suspension are placed upon clean 25-mm cover slips and heated to dryness, after which they are inverted upon a drop of Hyrax. Diatoms are identified with an Olympus VANOX phase-contrast microscope at magnifications of 600-1500X.

3) Enumeration--Counts are made from sedimented samples on the inverted microscope at 560X utilizing a Whipplegrid with an area of 0.04457 mm². Counts are made on a multiple tally counting machine and recorded on standard ECI algae beach forms. The number of grids examined per sample varies according to sample density; however, grid number remains constant within a specific lake. In accordance with procedures suggested by Margalef (1974), at least 100 individuals of each "important" taxon are counted. Because phytoplankton distribution within the plate chamber is rarely uniform, grids from several areas of the chamber are observed (e.g., center, sides). The counting units utilized were:

Unicells--each cell

Diatoms--each complete frustule (two halves)

Filaments--100- μ m length

Discrete colonies--each 4-8-16-32-64 cell colony
Indiscrete colonies--every eight cells
Dense colonies--every 50 cells

Examples of discrete colonial forms include Pandorina, Volvox, and Oocystis. Indiscrete colonial forms include Achnanthes (Herismopedia), Chroococcus, and Crucigenia. Examples of dense colonies are Microcystis and Aphanolizea.

All identifications, counts, and other pertinent data were recorded on standard algal bench forms. Replicate data were transposed to summary data for presentation in reports.

Phytoplankton densities were calculated according to the following equation:

$$\text{units/ml} = \frac{Ct \times V_o \times A_c \times D_f}{A_g \times N_g \times (V_o - V_p) \times V_s}$$

where:

Ct = count per species

V_o = original sample volume (ml)

V_s = sedimentation volume (ml)

V_p = preservation volume (ml)

D_f = dilution (or concentration factor)

A_g = area of one grid (mm²)

A_c = area of chamber (mm²)

N_g = number of grids counted

3.1.2.2. Zooplankton

Zooplankton were sampled by taking three vertical hauls at each sample site from approximately 0.5 m above the bottom to the surface. Collections

were made with a Standard Wisconsin style plankton net, with an 80- μ m mesh size and 13-cm diameter mouth opening. The length of each haul was recorded. Complete procedures are outlined below.

Collection Procedures

- 1) Slowly lower the Wisconsin net to a depth 0.5 m from the bottom.
- 2) Raise the net in a slow but steady fashion.
- 3) After removing the net from the lake, carefully wash the sides of the net off in the lake without submerging the top of the net.
- 4) Remove the bucket from the net and rinse into a 250-ml polypropylene bottle.
- 5) Add formalin to make a 5-10 percent formalin solution.

Taxonomic Analysis

- 1) The following procedures are used by Ecology Consultants, Inc. for zooplankton analysis. To determine the proper volume for counting, an estimate is made of the zooplankton density for all stations. First, the sample volume is concentrated to 100-ml. Next a 1-ml subsample is removed with a calibrated pipette and placed into a Bogorov counting chamber (Gannon, 1971). This subsample is counted for total zooplankton. Based on this enumeration, a sample volume is calculated to yield sufficient numbers in each individual subsample (ideally, 300 organisms). The sample volume is then readjusted if necessary.
- 2) After the appropriate sample and subsample volumes are determined, the samples are examined. A subsample is removed with a calibrated pipette and placed into the counting chamber. The chamber is examined using a Bausch and Lomb stereozoom binocular microscope (50-70X). Usually some rotifers and dissected copepods are taken from the counting chamber, placed on a slide, and identified using an Olympus Vanox compound microscope (100-400X). All organisms are identified and enumerated in the first subsample. In the second subsample, all taxa are counted except those which are extremely abundant in the first subsample (greater than 30 organisms). If the total zooplankton density is relatively low, additional subsamples are examined for less abundant taxa (those that are less than a total of 30 in the first

two subsamples). If possible, at least 15 percent of the sample is examined. The same volumes and counting procedures are repeated for all samples representing a particular sample date.

The number of organisms per liter is calculated according to the following equation:

$$D = \frac{(Ct) \cdot V_2}{\frac{V_1}{V_3}}$$

where:

D = number of organisms per liter

Ct = total number of organisms in subsamples

V₁ = total subsample volume in ml

V₂ = original sample volume in ml

V₃ = volume of water sampled in liters

All Cladocera and Copepoda are identified to species except for immature Copepoda, which are enumerated as nauplii, cyclopoid copepodites, calanoid copepodites, and harpacticoid copepodites. Rotifera are identified to the lowest possible level (usually genus or species).

3.1.2.3. Benthic Invertebrates

The benthic fauna was quantitatively sampled with a Ponar dredge (petite version, 15.2 x 15.2 cm) or an Ekman dredge (15.2 x 15.2 cm). Samples were sieved through a #40 standard sieve and preserved with formalin. Three replicate samples were collected at each station.

Qualitative invertebrate samples were collected along the shoreline in the vicinity of the deepwater stations. As many of the littoral zone habitats as possible were sampled during the qualitative surveys. In addition to employing aquatic nets and SCUBA for qualitative sampling, rocks and logs were removed from the bottom by hand and organisms

removed. These samples were preserved in formalin for taxonomic analysis. Lake benthic invertebrates were processed similarly to stream invertebrates (see Appendix S-4).

3.1.2.4. Macrophytes

SCUBA was employed to collect aquatic macrophytes along transects from the zone of emergent vegetation to the maximum depth of the littoral zone. Two transects were sampled in each lake (except in Birch Lake with four transects). These transects were located in the vicinity of the qualitative invertebrate stations.

At the laboratory, samples were split with the first portion remaining at the field station for identification, and the remainder shipped for heavy metals analysis. Macrophytes retained at the field station were identified.

3.1.2.5. Heavy Metal Tissue Analysis

Macrophytes and invertebrates (primarily clams and crayfish) collected during the macrophyte survey (see Section 3.1.2.4.) were analyzed for heavy metals. At the laboratory, samples were split, with one portion remaining at the laboratory for identification and the remainder being shipped for heavy metals analysis. See Appendix S-5 for complete details.

3.1.2.6. Other Observations

During each collection, dissolved oxygen, temperature profiles, and Secchi disk readings were recorded.

3.1.2.7. Permanent Archive

A reference collection of benthic invertebrates found in the lakes was incorporated with the stream reference collection. This reference collection contained representative individuals of each taxon collected. Invertebrates were preserved in 70 percent ethanol, except chironomids which were mounted on slides. Macrophytes were dried, pressed, and mounted on herbarium paper. In addition, Hyrax diatom slides were made for inclusion in the archives.

3.1.3. Information Inputs from other Programs

Information and data from the water quality and fisheries studies were necessary for the assimilation of aquatic biology data in the regional characterization. Water quality data are obtained through the Operations and Data Coordination staff. Fisheries data are obtained directly from fish study project group. Data and information from the bioassay program were necessary for impact assessment, and also obtained through the Operations and Data Coordination staff.

3.2. Sampling Frequency

Table L-2 includes the sampling frequency for the various parameters.

3.3. Equipment

See Appendix L-1 for an equipment list.

3.4. Quality Control Provisions

See Operations Manual; Aquatic Biology--Streams for general quality control provisions.

4. RECORD KEEPING AND DATA TRANSMISSION

No specific data notebook was kept; instead, numbered sheets were used and then bound into a notebook upon completion of the monitoring program. A spiral bound notebook was kept for following the movement of samples from lakes, to laboratory, to analysis, and to final report. Pencil was used to make all recordings in the field. Abbreviations used on data sheets are explained in Appendix L-2. Sample data sheets appear in Appendix L-2 and Appendix S-7. Aquatic biology data were transmitted to the Operations and Data Coordination staff on a monthly basis.

5. RESULTS, ANALYSIS, AND REPORTS

See Operations Manual; Aquatic Biology--Streams for a complete numerical analyses list. The only change to be noted is that results of phytoplankton and zooplankton are expressed as number/liter and phytoplankton biomass (chlorophyll a) as mg/l. Quarterly reports were written on the status of the aquatic biology study. In addition, monthly reports of data were provided when available.

6. STUDY STAFF

See Appendix S-8 for study staff resumes.

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1. INTRODUCTION AND SCOPE

1.1. Introduction

Elevated heavy metals levels have been found in a small unnamed creek adjacent to Eric Mining Company's Dunka Pit. These metals' origin is unknown.

The effect of these heavy metals on the Unnamed Creek biota has not been extensively studied. In addition, the effect on Birch Lake biota from heavy metals movement into Bob Bay of Birch Lake has never been studied.

No conclusions were drawn from the results of a 1975 Barr Engineering study (Barr Engineering Co., 1976). A brief qualitative survey conducted by the Regional Copper-Nickel Study, Aquatic Biology staff, indicated some biotic differences between the various stations sampled. However, reasons for these differences could not be determined (Johnson et. al., 1976a). Various factors such as natural habitat variation, channelization, heavy metals, and fluctuating water levels appear to affect Unnamed Creek biota.

Regional Copper-Nickel Study biological monitoring began in May 1976 and included a secondary station at the mouth of Unnamed Creek (Station BB-1) (Johnson et. al., 1976b) and four sites in Birch Lake, which were sampled as part of the lake monitoring program (Johnson et. al., 1976c). Data from this sampling are not yet available.

Because little is known about the biological effects of heavy metals in natural aquatic systems, it was thought that the Regional Copper-Nickel Study might gain valuable data for use in analyzing potential impacts of copper-nickel mining in northeastern Minnesota by intensively sampling Unnamed Creek. Erie Mining Company also indicated a desire to determine the biological effects of heavy metals in Unnamed Creek and in Birch Lake, and thus a joint effort was organized.

1.2. Scope of Study

The study of Unnamed Creek focused on the periphyton and benthic invertebrate components of the ecosystem. Because it was thought that metals would accumulate in the sediments of Birch Lake, emphasis was placed on benthic invertebrates. Phytoplankton were sampled less intensively in Birch Lake. The role of these organisms in the ecosystem and the reasons for sampling them can be found in Operations Manual; Aquatic Biology--Streams and Lakes.

1.3. Time Schedule

Sampling was initiated in July 1976 and was completed in November 1976. A detailed time schedule is included in Table EM-1.

2. STUDY AREA AND SAMPLING STATIONS

2.1. Study Area

The study area was located on the east side of Erie Mining Company's Dunka Pit, east of Babbitt, Minnesota. The area is primarily composed of

bogs. Unnamed Creek originates at a mine dewatering site on Dunka Pit's south end, flows north through the area, and enters Birch Lake through Bob Bay. Dunka River, which originates in bog areas south of Dunka Pit, flows into Birch Lake's Dunka Bay, located northwest of Dunka Pit. Dunka Bay served as a control area for the study of Bob Bay.

2.2. Sampling Stations

Stations were selected for sampling after examining the available chemical and biological data from Unnamed Creek and Birch Lake. Unnamed Creek stations corresponded to stations sampled by Barn Engineering in 1975; except that S-5 was not included and a new station EM-1a, located immediately upstream from the confluence of the main channel and the tributary entering the creek from the west, was added (Table EM-2 and Figure EM-1). Water samples from all stations were analyzed chemically except at Station EM-1a. In Birch Lake, three stations were located in Bob Bay and two stations in Dunka Bay. One additional station was located in the main body of Birch Lake immediately north of Bob Bay (see Table EM-2 and Figure EM-1). Justification for the location of each station is included in Tables EM-2 and EM-3. All stations were in addition to the regular monitoring stations mentioned in Section 1.1. Figures EM-1 and EM-2 include the location of Erie Mining Study sampling stations.

3. METHODOLOGY

3.1. Techniques

3.1.1. Justification of Techniques

In Unnamed Creek, artificial substrates were the primary method of collecting periphyton and benthic invertebrates. Drift nets and qualitative techniques were also employed. Justifications of these techniques are included in Operations Manual; Aquatic Biology--Streams.

Birch Lake phytoplankton were collected with an integrated water sampler, and benthic invertebrates were collected with petite Ponar dredge. This equipment is discussed in Operations Manual; Aquatic Biology--Lakes.

3.1.2. Procedures

3.1.2.1. Periphyton

Three glass slide racks were suspended in Unnamed Creek approximately 30 cm below the water surface, depending on expected water level fluctuations. These slides remained in the stream for three weeks.

In addition, a qualitative survey of the naturally occurring periphyton was carried out. All types of natural substrates were observed and any growth collected. Qualitative samples analysis consists of species identifications.

Procedures as described in Appendix S-3 were followed, except that slide numbers 1 and 3 were taxonomically analyzed (sedimentation counts and pro-

portional diatom counts) and slide numbers 2 and 4 were analyzed for chlorophyll a.

3.1.2.2. Benthic Invertebrates (Unnamed Creek)

Six modified Hester/Dendy samplers were suspended at each stream station for two six-week colonization periods. Samplers were suspended just above the stream bottom.

Three drift collections were made in Unnamed Creek. Drift nets with an upstream opening of 0.025 m², length of 2.4 m, and 440-um mesh size were placed in the stream for 24 hours. The exact starting time was not critical as long as it was during daylight hours, because drift is low and constant during the day with peaks between sunset and sunrise.

Because Hester/Dendy samplers and drift nets are selective in their sampling, qualitative invertebrate samples were collected in order to compile a species list of the entire natural community. Sampling consisted of examining all various stream habitat types such as pools and riffles, various sized rocks, logs and silt for two man-hours. In addition to picking organisms from logs and boulders, the kick-net method was employed wherever feasible. Complete procedures are described in Appendix S-4.

3.1.2.3. Phytoplankton

At each lake station, water was collected with the integrated sampler described in Operations Manual; Aquatic Biology--Lakes. Three replicates were collected for chlorophyll a analysis and phytoplankton taxonomy.

Complete field and analysis procedures are described in Operations Manual; Aquatic Biology--Lakes.

3.1.2.4. Benthic Invertebrates (Lakes)

Six replicate petite Ponar dredge samples were collected at each lake station. Samples were sieved through a standard #40 sieve in the field and the remaining sample preserved in 5-10 percent formalin. Complete procedures are described in Operations Manual; Aquatic Biology--Lakes.

3.1.2.5. Heavy Metals Tissue Analysis

Macrophytes, invertebrates, and periphyton samples were collected in conjunction with regular stream and lake tissue collections in Unnamed Creek, Bob Bay, and Dunka Bay; where adequate material could be found. Collection and processing procedures are included in Operations Manual; Aquatic Biology--Streams and Lakes.

3.1.3. Permanent Archive

A reference collection of benthic invertebrates was placed in a permanent archive. These samples were preserved in 70 percent ethanol. Additionally, Hyrax diatom mounts, chironomid slides, and the remainder of the benthic invertebrates (preserved in 70 percent ethanol) were made for inclusion in the permanent archive.

3.1.4. Analysis of Samples

All field collections and chlorophyll a measurements were the responsi-

Erie Mining, Page 7.

bility of the Regional Copper-Nickel Study, Aquatic Biology staff. Periphyton and benthic invertebrate samples were analyzed by Ecology Consultants, Inc., Ft. Collins, Colorado. This contract was written and financed by Erie Mining Company.

Heavy metals analysis of macrophytes was done at the University of Minnesota, St. Paul, Minnesota, through a Regional Copper-Nickel Study contract. Invertebrates were analyzed for heavy metals by the Department of Natural Resources, Chemistry Laboratory.

3.1.5. Information Inputs From Other Programs

Data from the leaching study were necessary for the complete analysis of the biological data. This information was obtained from the Operations and Data Coordination staff.

3.2. Sampling Frequency

Table EM-4 shows the sampling frequency for the various parameters.

3.3. Equipment

The operations manuals for Aquatic Biology--Streams and Lakes includes equipment lists for sampling procedures employed in the Erie Mining Study.

3.4. Quality Control Provisions

3.4.1. General

Quality control provisions described in Operations Manual; Aquatic

Biology--Streams have been employed in the Eric Mining Study. In addition, the following procedures were instituted to ensure sample integrity:

- 1) all operations of the Unnamed Creek Study were directed by the aquatic biology project head to assure accuracy at each stage of the study;
- 2) Erie Mining Company personnel accompanied aquatic biology staff members on all sampling trips and could accompany the Copper-Nickel staff to the Kawishiwi Field Station to observe sample processing;
- 3) to ensure sample integrity and to eliminate bias, all samples were given a serial number before analysis and shipping;
- 4) strict chain of custody procedures were followed in the handling and transport of all samples collected on the Eric Mining Study.

3.4.2. Calibration, Maintenance, and Testing Equipment

See Operations Manual; Aquatic Biology--Streams.

4. RECORD KEEPING AND DATA TRANSMISSION

Field data were recorded on data sheets in a loose-leaf notebook. All data were recorded in pencil. Sample data sheets are included in Operations Manual; Aquatic Biology--Streams and Lakes. Xerox copies of all field and laboratory data sheets were submitted to Regional Copper-Nickel Study Operations and Data Coordination staff and Phil Brick (Erie Mining Company) when available.

5. RESULTS, ANALYSES, AND REPORTS

5.1. Numerical Analyses

See Operations Manual; Aquatic Biology--Streams for a listing of numerical analyses that were employed in the Eric Mining Study.

5.2. Progress and Final Reports

Monthly reports of the sample data were submitted by Ecology Consultants, Inc. to Eric Mining Company and to the Regional Copper-Nickel Study with all analyses completed within 45 days from receipt of the final samples shipment. A final report covering the results of the 1976 study was written by the aquatic biology project head and completed on September 1, 1977.

6. STUDY STAFF

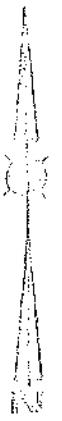
See Appendix S-8 for resumes of study staff.

LOCATION OF BIRCH LAKE

FIGURE PM-1

SAMPLING STATIONS

FOR ERGON MINING STUDY



Appendix S 9. 1977 Modifications to Aquatic Biology Sampling Program.

The 1977 aquatic biology study has been designed to provide additional data on those watersheds where the greatest impact is expected from Cu-Ni development. Also, the study has been expanded into several other areas of interest:

- 1) Qualitative and drift sampling is being done in areas previously not sampled in order to develop a system of classifying streams in the area. Much of this sampling was done in lower order streams (ie. 1st and 2nd). Data from this survey will be related to the more intensive data collection at other stations. Third and fourth order streams are represented by regular monitoring stations.
- 2) Intensive drift, kick net, and periphyton sampling in the Snake River/Little Isabella River Watersheds and at two 4th order stations (regular monitoring stations). In this area, it is hoped to relate stream order and terrestrial vegetation to the invertebrate functional groups and the degree of autotrophy.
- 3) Leaf decomposition study in the Snake River/Little Isabella River Watershed and at a southern and a northern 4th order station.

SAMPLING STATIONS

As in 1976, aquatic biology monitoring stations have been designated primary, secondary, and tertiary. The following is a list of the stations to be sampled in 1977:

Primary: K-1, K-8, SR-2¹, D-1, E-1, P-1, SL-1
Secondary: K-2, P-5, P-2, SR-3, BI-1, SL-2¹
Tertiary: KC-1¹, F-11, SR-4, D-2, P-3, P-4, I-1, K-6, E-2, SR-1¹

¹station designation different in 1976

The following stations have been dropped: BK-1, K-3, K-4, SL-3.

Additional sampling stations will be assigned "SCS" (stream classification system) numbers. Primary SCS stations are located in the Snake River/Little Isabella River Watershed, while secondary SCS stations are spread throughout the entire area. In some cases, SCS stations are also considered as regular monitoring stations. In the following listing of SCS stations, stations which serve as both monitoring and SCS stations are indicated.

Primary SCS station:	SP-1	Mouth Sphagnum Creek	1st order
	LI-3	Little Isabella River headwaters	1st order
	LI-1	Little Isabella River at Forest Rd. 173	2nd order
	LI-2	Little Isabella River at L.J. Campground	2nd order
	SC-1	Snake River below confluence w/Snake Creek	3rd order
	SE-1	Snake Creek at Forest Rd. 173	3rd order
	SL-1	St. Louis River	4th order
	SR-2	Stony River	4th order
Secondary SCS stations:	N-1	Nip Creek	1st order
	KC-1	Keeley Creek tributary	"
	D-3	Dunka headwater	"
	F-2	Filson Creek headwater	"
	NR-2	Nira Creek headwater	"
	SH-1	Shiver Creek	"
	T-1	Toimi Creek	"
	KC-1	Mouth Keeley Creek	2nd order
	CY-1	Coyote Creek	"
	SG-1	Spring Creek	"
	NW-1	North Branch Whiteface River	"
	BC-1	Bear Creek	"
	SE-2	Snake River	"
	NR-1	Nira Creek	"
	DC-1	Denley Creek below Nira Confluence	3rd order

METHODS

I. Periphyton

- A. Periphytometer W samplers are employed for suspending slides at stream stations. These samplers hold eight slides vertically at a constant depth. A diagram is attached.

- B. Two samplers are placed at primary monitoring stations; six random slides are selected for biomass (ash-free dry weight-Weber, 1973) and chlorophyll a determinations; four random slides are selected for (monochromatic-Weber, 1973) permanent diatom mounts. These diatom mounts are prepared by using the potassium permanganate method (Handey, 1976).

- C. One sampler is placed at secondary monitoring stations; three random slides are selected for biomass and chlorophyll a; two random slides are selected for diatom analysis.

- D. At primary stream classification stations one sampler was installed; three random slides will be selected for biomass and chlorophyll a analyses; two slides will be used for diatom analysis.

- E. A 3-week colonization period is again employed.

- F. Qualitative periphyton samples will be collected as in 1976.

II. Benthic Invertebrates

- A. Modified Hester-Dendy samplers will be placed on the stream bottom to eliminate any of the problems caused by low flows. Four samplers will be placed at primary

stations; three samplers at secondary stations. No samplers are placed at stream classification stations. Six-week colonization periods are employed.

B. Three drift nets are placed at all stations during sampling periods for 24 hours. Exceptions to this are small stream classification stations where one or two nets sample essentially the entire stream. New drift nets have been shortened to approximately 1.5m. Current velocity is measured at the time of net installation and retrieval to provide a better estimate volume samples. Organic material collected in the drift nets will be dried at 105°C for 24 hours after the animals have been removed from the sample.

C. Pool and riffle sections are sampled separately during the two-hour qualitative. Time spent in each stream type is recorded. At stream classification stations, the sampling time was reduced if no new invertebrates were found during 15 minutes of sampling.

D. Three 60-second kick-net samples are collected from each primary SCS site at the same time of drift sampling. These samples are collected by standing upstream from an aquatic insect net and disturbing the substrate for 60 seconds. Invertebrates are picked in the field.

III. Comparative Studies

A. Periphyton-Data on the comparability of data collected from the 1976 slide rack and the 1977 Periphytometer II will be collected at K-5. Sixteen slides have been suspended from each type of sampler; eight chlorophyll and eight diatom analyses will be made from each type of periphyton sampler. Taxonomic differences and variability within and between sampling methods will be determined.

B. Benthic Invertebrates - A comparison of differences in samples collected by Hester-Bendy samplers suspended in a stream and those placed on the bottom is also being conducted at K-5. Four samplers have been placed on the bottom and four have been suspended. Samples collected will be analyzed taxonomically. Differences in taxa collected and variability within and between sampling methods will be determined.

C. Drift - Differences in drift during daylight hours and in the dark will be examined at K-2 during a new-moon period. This should provide data on the bias resulting from 24-hour drift sampling.

IV. Leaf Decomposition

A. Ten gram leaf packs of aspen leaves and red pine needles were constructed from 3mm nylon mesh material.

B. Leaf packs were attached on the front side of log placed in a stream. It was attempted to anchor the leaf packs in as natural a manner as possible.

C. Aspen leaf packs were placed at all primary stream classification stations (one station was divided into a pool and riffle sections) while red pine leaf packs were placed on one station of each stream order including the pool/riffle station.

D. Leaf packs are collected at two-week intervals for an eight-week period.

E. After collection, each sample is washed to remove the associated invertebrate fauna for taxonomic analysis. After the invertebrates have been removed from the leaf pack, the remaining material is dried at 105°C for 24 hours to determine weight loss.

F. At the time of each collection, a natural leaf pack is collected to determine the similarity of the fauna associated with natural and artificial leaf packs.

SAMPLING SCHEDULE

Artificial substrates were installed three weeks earlier than in 1975 due to the low stream flows and high water temperatures. A complete chart of sampling frequencies is attached.

SAMPLE ANALYSIS PRIORITIES

Table 2 presents sample analysis priorities. As in 1976, more samples are scheduled to be collected than can be analyzed within the project deadlines. These samples will be available if weather conditions curtails sampling, if a large number of samples are lost or if analysis of samples indicates that further analysis of samples could add significant data on a particular stream.

Table 2. Sample analysis priorities

SAMPLE TYPE	NUMBER OF SAMPLES COLLECTED								How Analyzed
	Monitoring Stations		SOS Stations		SOS Stations		SOS Stations		
	Primary	Secondary	Tertiary	Primary	Secondary	Primary	Secondary		
Chlorophyll	252	125	---	54	---	---	---	453 (CI-MI)	
Biomass	252	62	---	54	---	---	---	269 (SERCO)	
Diatom (Quant.)	168	42	---	36	---	---	---	246 (Cu-MI)	
Diatom (Qual.)	14	14	22	12	30	---	---	92 (Cu-MI)	
Invert (H/D)	84	42	---	---	---	---	---	84 (I.D.) primary 42 (Store) secondary	
Invert (Drift)	126	63	---	72	90	---	---	250 (ZCT) 100 (Cu-MI)	
Invert (Qual.)	21	21	22	16	50	---	---	52 (I.D.) 14 (Store)	
Leaf Packs	---	---	---	156	---	---	---	156 (Cu-MI)	

Table 1. 1977 sampling schedule

	APRIL	MAY	JUNE	JULY	AUGUST	SEPTEMBER
PERIPHYTON						
Quantitative						
Primary		-----				
Secondary		-----				
Primary SCS			-----			
Qualitative						
Primary						
Secondary						
Tertiary						
Primary SCS						
Secondary SCS						
INVERTEBRATES						
Master/Dandy						
Primary		-----				
Secondary		-----		-----		
DRIFT						
Primary						
Secondary						
Primary SCS						
Secondary SCS						
QUALITATIVE						
Primary						
Secondary						
Tertiary						
Primary SCS						
Secondary SCS						
LEAF PACKS						
Primary SCS		-----				

TAXONOMY

Nomenclature for diatoms follows that A.J. Hustedt (1930A) and Patrick and Reimer (1966, 1975) except in the cases of the genus Emilia where Hustedt (1930B) was used and the genus Licisira where the discussion and illustrations of Lund (1962) were used.

Where breakage and corrosion have affected almost all the frustules in a sample, broken frustules were counted. Only fragments with diagnostic features were tallied. The following criteria were used:

Anomooneis, Neidium, Stauroneis and Cyrosigma-

central areas with surrounding striae were used to define the genus

Frustulia-terminal modules with surrounding striae were used to define the genus

Eunotia-terminal ends were used to differentiate the genus. At least half a value was necessary to distinguish between species

Surirella-terminal fragments were used to distinguish the genus but entire values were necessary to distinguish species

Stephanodiscus astraea-at least a half a frustule including the central area was needed to qualify for counting (fragments of all sizes can be identified to species). This taxon may include types identified by other workers as S. niagarae. And follow Lund's (1962) opinion that the latter species is only present in large northern lakes and rare elsewhere on the continent. Bragan (personal communication) refers specimens to S. niagarae whenever a band is present outside the spines. By this criterion almost all frustules in our material would be S. niagarae.

Cyclotella comta was tallied where half or more of a value was present.

This taxon may include forms designated as C. bodanica by other workers

although I failed to find "Flammidea" punctate (Hustedt, 1930A) in this material. Specimens of Cyclotella gonta in the 1-2 centimeter sample from Clearwater Lake were very variable in morphology.

I follow Bradbury and Brugan (personal communication) in designating small stellate Cyclotella as Cyclotella glomerata, although these forms could possibly be referred to C. stelligera since the filamentous growth habit of the former species is broken up in the sediment and striae counts of the two forms are too close for me to distinguish them. Individuals exceeding 10 μ in diameter were as C. stelligera.

Tabellaria fenestrata and Tabellaria flocculosa fragments were tailed only when they included a striated central area. The presence of variable numbers of intercalary bands between the two frustules of this genus requires that striae be present to be sure that an actual frustule is represented.

Melosira granulata V. angustissima was differentiated according to the description and illustration of Hustedt (1930A). Brugan (personal communication) finds that this form is exceedingly rare in northeastern Minnesota lakes. Its presence in high percentages in White Iron lakes would seem questionable, but the specimens cannot be referred to the most likely alternative species, Melosira islandica, because the striae run diagonally. The nominate variety of Melosira granulata is easily distinguished and has been found by Brugan (personal communication) in surface sediments of Fall Lake and Bradbury (1975) in sediments of Shagawa Lake. Taxonomic differentiation of Melosira ambigua and Melosira italica appears to be in a state of confusion. I have attempted to follow the criteria of Lund (1962) that Melosira ambigua has a U shaped sulcus appearing like a small white eye, while Melosira

italica has a V shaped sulcus. Bragan (personal communication) using larger spine size as a criterion, fails to find Melosira italica very commonly in northeastern Minnesota. Whenever frustules of Melosira were obscured or the sulcus was difficult to classify, I have lumped Melosira species into a Melosira spp., undifferentiated" class.

This type undoubtedly contains representatives of several species. Elongate forms of Melosira in Clearwater lake have been reserved to M.italica although they approach the least elongate variants of M. granulata. V.angustissima found in Birch lake.

Melosira distans is distinguished by its parallel striations and valves whose breadth is greater than their diameter. This height/diameter ratio often results in their being seen in their distinctive valve view. Nitzschia species are differentiated from each other by shape, striae count and spacing of keel punctae the genus is easily distinguished. Amphiprora ornata was only present in broken form and was only distinguished from fragments that included the twisted portion of the valve.

Small Naviculas and Achnanthes (less than about 10 μ m) are difficult to distinguish. Whenever a pseudoraphe valve was present an individual was assigned to the genus Achnanthes. Unless identification to species was fairly certain or an individual could not possibly be referred to Achnanthes it was tallied as an unidentified pennate, rather than being referred to the genus Navicula.

Frustules tallied as Diploneis smithii may include the form described by Patrick and Reimer (1966) as D. finaica. Most of these valves were broken and I found it difficult to differentiate which of the two taxa was more nearly correct.

I followed the convention of Bradbury (1975, personal communication) in differentiating

Stephanodiscus minutus from S. haartzschii on the basis of the raised central area of the former species. There is a possibility that these forms are all variants of the same taxon. Unidentifiable corroded centric forms less than 10 μ in diameter may be either of these two Stephanodiscus species, or they may be Cyclotella glomerata.

Melosira roseana was distinguished using Hustedt (1930A). This form has not been recognized by Brugan (personal communication) in Minnesota lakes. Fragments can be differentiated by their striae and curvature. Only two intact valves were noted, one in Gabbro Lake and one in White Iron Lake. Because of its unusual nature an illustration of this form is included (figure 1)

Pollen types were differentiated by the use of reference slides in the collection of the Limnological Research Center and illustrations in McAndrews, Berti, and Norris (1973).

Table 2. cont'd

TAXA	Site: Date: Taxonomist:		SI-D-B 6-8-76		SI-J-B 6-8-76	
			deCottloch	Rischuk	deCottloch	Rischuk
<u>G. angustatum</u>				20	3	2
<u>G. brobissonii</u>				1		
<u>G. gracile</u>			3			
<u>G. parvulum</u>			13		2	5
<u>G. sp.</u>			14	11	2	1
<u>Melosira distans</u>						3
<u>M. granulata</u>			1		2	4
<u>navicula exigua</u> var. <u>capitata</u>					1	
<u>N. minima</u>						3
<u>N. pupula</u>					1	2
<u>N. radiosa</u>					4	
<u>N. radiosa</u> var. <u>tenella</u>						3
<u>N. seminulum</u>						4
<u>N. sp.</u>						3
<u>Reidium</u> sp.						1
<u>Mitschia acicularis</u>						2
<u>N. bacata</u>						2
<u>N. gracilis</u>					3	
<u>N. kutzingiana</u>						4
<u>N. pulea</u>					9	3
<u>N. recta</u>						1
<u>N. sp.</u>					4	9
<u>Stephanodiscus</u> sp.						1
<u>Synedra acus</u>					3	
<u>S. radians</u>			3			
<u>S. rumpens</u>					1	6
<u>S. ulna</u>						1
<u>S. sp.</u>			2	5		5
<u>Tabellaria fenestrata</u>						1
<u>T. flocculosa</u>					7	11
Total Taxa			12	10	24	45

Table 3. Species proportions from comparison of the two projects and consultant taxonomists

Site: E-1-B

Date: 9/21/76

Project Taxonomist: Hirsch
 Consultant Taxonomist: Palmer

TAXA	PROJECT	CONSULTANT
<u>Adiantum affinis</u>		2
<u>A. linearis</u>		3
<u>A. linearis var. pusilla</u>	3	
<u>A. lanceolata</u>	2	
<u>A. microcephala</u>	45	
<u>A. simonsiana</u>	7	45
<u>A. stewartii</u>		1
<u>Asplenium platyneuron</u>	17	9
<u>Asplenium ovale var. pedunculatum</u>		2
<u>Asplenium vitreum</u>	2	3
<u>Calyptra bacillata</u>		3
<u>C. hyalina</u>		1
<u>Cartogramma cruciata</u>	4	
<u>Cocconeis placentula</u>	8	1
<u>C. placentula var. euglypta</u>		1
<u>C. placentula var. lineata</u>		5
<u>Cyclotella glomerata</u>	1	
<u>C. meneghiniana</u>	2	5
<u>C. sp.</u>	2	
<u>Cymbella minuta</u>	3	
<u>C. minuta silesiaca</u>		2
<u>Diploneis oculata</u>		1
<u>Euzozia curvata</u>	3	1
<u>E. monodon</u>		1
<u>E. pectinalis var. minor</u>		1
<u>E. praerupta</u>	1	
<u>E. sp.</u>	2	

Page 3 cont'd

Title: L-3-B cont'd.

Date: 9/21/76

<u>FUNGA</u>	<u>SUBJECT</u>	<u>CONSULTANT</u>
<u>E. leucella</u>		1
<u>Fragilaria brevistriata</u>		1
<u>F. constricta</u>	2	
<u>F. constricta</u> var. <u>penula</u>		3
<u>F. constricta</u> var. <u>vaalier</u>	1	
<u>F. pinnata</u>		2
<u>Frustulia rhomboides</u>	3	
<u>F. rhomboides</u> var. <u>crassinervia</u>		1
<u>F. serinholdii</u>		1
<u>Comadronema intricatum</u>		3
<u>G. olivaceum</u>	3	
<u>G. constrictum</u>	1	
<u>G. parvulum</u>	6	4
<u>G. sp.</u>	3	
<u>G. truncatum</u>		1
<u>Ictosira granulata</u>	2	
<u>I. varians</u>	4	
<u>Iavicula capitata</u>	1	1
<u>I. cryptocephala</u>	16	12
<u>I. arvensis</u>		1
<u>I. bicephala</u>		2
<u>I. bryophila</u>		3
<u>I. contenta</u> var. <u>biceps</u>		1
<u>I. ligna</u>		1
<u>I. elginensis</u> var. <u>neglecta</u>		1
<u>I. exigua</u>	2	

Table 3 cont'd.

Site: LFB cont'd.

Date: 9/23/76

<u>TAXA</u>	<u>PROJECT</u>	<u>CONSULTANT</u>
<u>H. exigua</u> var. <u>capitata</u>		1
<u>H. laevissima</u>		1
<u>H. minima</u>		3
<u>H. notha</u>		11
<u>H. peratunkae</u>		2
<u>H. pupula</u>	2	1
<u>H. pupula</u> var. <u>capitata</u>		1
<u>H. pupula</u> var. <u>mutata</u>		1
<u>H. radiosa</u>	9	5
<u>H. rhychocephala</u>	1	2
<u>H. rhychocephala</u> var. <u>germainii</u>		1
<u>H. schroederi</u> var. <u>escambia</u>		2
<u>H. secreta</u> var. <u>apiculata</u>		1
<u>H. similis</u>		1
<u>H. sp.</u>	9	
<u>H. subannulata</u>		2
<u>H. tenelloides</u>		5
<u>H. viridula</u> var. <u>rosicelata</u>	1	
<u>H. wallacei</u>		2
<u>Hirschia acannodonta</u>		1
<u>H. acicularis</u>	1	3
<u>H. acuta</u>	5	
<u>H. acicularis</u> var. <u>africana</u>		1
<u>H. bicata</u>		9
<u>H. clausii</u>		1
<u>H. confinis</u>		1

Table 3 cont'd.

Site: I-3-a cont'd.

Date: 9/21/76

TAXA	PROJECT	COUNT
<u>H. sp. (cf. <i>dissipata-obsidialis?</i>)</u>		7
<u>H. epiphytica</u>		1
<u>H. fasciculata</u>	2	
<u>H. filiformis</u>	16	
<u>H. frustulum var. <i>perminuta</i></u>		1
<u>H. frustulum var. <i>subsalina</i></u>		9
<u>H. gracilis</u>	7	5
<u>H. kutzingiana</u>		36
<u>H. linearis var. <i>tenax</i></u>		6
<u>H. palea</u>	20	1
<u>H. romana</u>	7	
<u>H. recta</u>		2
<u>H. sigmoidea</u>	1	
<u>H. sp.</u>	4	
<u>H. spiculum</u>		1
<u>H. sp. a</u>		8
<u>Pinnularia sp.</u>	1	
<u>P. termitina</u>		1
<u>Surirella delicatissima</u>		1
<u>S. linearis</u>		1
<u>S. ovata</u>	1	
<u>Synedra acus</u>	5	
<u>S. nana</u>		4
<u>S. pulchella var. <i>lacerata</i></u>		1
<u>S. rupeus</u>	5	7
<u>S. tenera</u>		2

Folle 3 contd.

Site: L-1-B

Date: 9/21/76

TAXA	PROJECT	CONSULTANT
Intellaria fenestrata	2	
E. sp.	7	

DIATOMS

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- 1930 (B) Die Kieselalgen in Böhmen, I. Kryptogamen-Flora von Deutschland, Österreich und der Schweiz, Band VII. Akademische Verlagsgesellschaft, Cest and Portig, K.G. Leipzig.
- Lund, J.M.G., 1962. Phytoplankton from some lakes in northern Saskatchewan and from Great Slave Lake. Canadian Journal of Botany, V. 40, pp 1489-1511.
- McAndrews J.H., Berti A.A., and Norris G., 1973. Key to the Quaternary Pollen and Spores of the Great Lakes Region. Life Sci. Misc. Publ., K. Ont. Museum. Toronto.
- Patrick R. and Reimer C.W., 1955. The diatoms of the United States, Vol. 1: Academy of Natural Sciences of Philadelphia Monographs, No 13, p 1-643, 1-64. 1975. The diatoms of the United States, Vol 2. Academy of Natural Sciences of Philadelphia Monographs, No 13, pp 1-213, pls. 1-28.

Appendix 5.11. Revision of Zooplankton Sampling Analytic Methods.

This appendix describes changes in procedures outlined in the Operations Manual section 3.1.2.2. These changes apply to 1977 zooplankton samples only.

At all stations, three vertical net hauls were taken and the samples integrated in one sample bottle.

The taxonomic analysis of samples taken in 1977 was conducted by project staff. Samples were concentrated in the laboratory to between 20ml and 100ml dependent on a visual estimate of the concentration of animals in the sample. A 1ml subsample was removed with a calibrated pipet after random stirring of the sample. All Protozoa, Rotifera, and copepod nauplii in the 1ml subsample were counted in a Sedgewick Rafter cell using a Bausch and Lomb compound microscope (100-200X). Additional 1ml subsamples were analyzed until 40 or more individuals of the common species had been counted. A 5ml subsample was removed with a Hensen-Stempel pipet and rinsed into a gridded petri dish. All Cladocera and Copepoda in this subsample were counted under a Bausch and Lomb stereozoom dissecting microscope. Additional subsamples were analyzed until 40 or more of the common taxa had been enumerated.

In 1977 samples, Copepoda were identified to suborder (i.e. Calanoida and Cyclopoida) only. Planktonic and protozoans were identified and enumerated in 1977 samples only.

Appendix 5-12. Quality Control Program for the Aquatic Biology Study.

The Quality Control Program of the Regional Copper-Nickel Study was initiated to assure the accuracy of the Aquatic Biology Program and to maintain consistency between project taxonomists and consulting taxonomists in identification of periphyton, invertebrate, and fish taxonomy (Table 1). Procedures were developed for each taxa depending on the analysis methods employed and the degree of taxonomic certainty.

Periphyton

Emphasis in the periphyton program was on diatoms. Although diatoms are the easiest algal group to identify to the species level, taxonomic difficulties occur. These difficulties are the result of the following problems: (1) insufficient taxonomic keys for various diatom genera; (2) difficulty in developing a diatom reference collection from permanent slides; (3) insufficient resolution of diatom ultrastructure; (4) differences in frustule size because of growth sequence; (5) quality of sample preparations; (6) condition and orientation of specimen being identifies; (7) unfamiliarity with some species characteristics.

Because of these difficulties a three phase program was developed. First, random samples were selected from the first 1976 samples analyzed. These samples were analyzed and recounted by the original taxonomist or exchanged with the other staff diatom taxonomist (Table 2). Second, two consulting taxonomists (Table 1) were contracted to reanalyze project slides. It was not possible for consultants to examine specific individuals, since the microscopes employed by project taxonomists did not have etching capabilities to designate these individuals; therefore consultants repeated the species proportional count on each slide. Project and consultants species lists and relative abundances were compared to determine the taxonomists on a single slide (Table 5). Three replicate counts were made on this slide to determine distribution patterns on an individual slide.

The quality control program indicated several problems. Species indentifications within the genera Cymbella, Pinnularia, Melosira, Navicula, Nitzschia and Synedra were inconsistent between project and consultant taxonomists (Table 2, 3, and 4). As a result all species indentifications in these

genera were dropped. *Actinocyclus microcephala* and *A. minutissima* were grouped in 1976 data because the identifications were confused (Table 2, 3, and 4). Fewer taxa were distinguished by project taxonomists than by consultants; additional taxa identified were rare (< 4 frustules) in most cases (Table 3, 4, and 5). Dominant species were found by all taxonomists (Table 5). Differences between all taxonomists project and consultant, were minimal at the generic level (Table 6). Heterogeneous diatom distributions on the slides influenced species lists and relative abundances recorded during slide examination, as indicated in the three counts conducted by project taxonomists (Table 5). Dominant taxa were consistently encountered in every count, although the species lists and relative abundance varied.

Two species names were changed because the diatom nomenclature was revised. *Cocconeis placentula* was changed to *C. placentula* var. *lineata* and *Diatoma elongatum* was changed to *D. teuge* var. *elongatum*.

All permanent periphyton slide mounts were placed in the aquatic biology reference collection for future comparison. The location of this collection is documented in another report.

INVERTEBRATES

Identification to the species level was influenced by the sex, maturity, and condition of the specimen, extent of taxonomic descriptions in the literature, and the need to rear immature specimens to adult stages for positive larval identification.

Taxonomic consultants were contracted to examine invertebrate specimens and verify or correct identifications. Consultant selection was based on authorship of taxonomic reference material used by the project, familiarity with the regional taxa, and recommendations by taxonomists having expertise in specific entomological areas (Table 1). As each additional taxon was identified it was sent to the appropriate consultant for examination and verification. Verified specimens were placed in a reference collection and as record of Study Area taxa for future comparisons.

Four invertebrate samples from 1976, were examined at early stages of the taxonomic work and re-examined after completion of all the 1976 sample analysis to ascertain identification errors. Each taxonomist had previously identified two of the four samples. This check was completed by having each taxonomist re-examine one of their previously identified samples and one of the other taxonomist's samples. This method allowed evaluation of errors by each taxonomist and inconsistencies between taxonomists. The results of the original analysis and the quality control checks were then compared (Table 2). Five types of errors became evident: (1) Identification to a lower taxonomic level (e.g. genus to species) in the quality control check due to greater familiarity with the local taxa, taxonomic keys, and through communication with consultants; (2) Different levels of familiarity with taxonomic characteristics between taxonomists; (3) Misidentifications; (4) Identification to the generic level rather than species level in the quality control check for taxa having poorly defined characteristics, based on consultant recommendations; and, (5) Generic identifications rather than species where specimens were damaged in the initial analysis. Omissions of taxa in the quality control lists were largely caused by removing specimens identified to species for placement in the reference collection during the initial analysis.

The errors were discussed and the following methods developed to circumvent future problems: (1) sending difficult specimens to consultant taxonomists for identification; and, (2) modifying the level of identification to reduce errors detected by the consultants. Baetis having poorly defined characteristics were grouped as recommended by McCafferty (personal communication, 197) for species belonging to the Baetis flavistrigis group spp. Because of errors and the time required to identify Coleoptera, especially small specimens, this order was identified to species and two Hydrophilidae (Hydrochus and Helophorus) were identified to genus. Difficult Stenonema species were all sent to Lewis for verification.

The impact of these errors was minimized by correcting misidentifications as determined by consultants and placing questionable identifications at a taxonomic level where we were confident. Most errors occurred at the species level and will have little impact on the analysis, which will be conducted at a functional group level based on generic and family level identifications.

Ecology Consultants Inc. (ECI) Fort Collins, Colorado was contracted to analyze some quantitative and qualitative samples. The quality of ECI taxonomy was evaluated by project taxonomists; questions of correct identifications and nomenclature were resolved by visiting ECI taxonomists and sending new specimens to our consultants for verification.

FISHES

Fish species not positively identified in the field or laboratory were sent to Dr. J. C. Underhill (University of Minnesota) for identification. These taxa were primarily small minnow (Cyprinidae) species. Verified specimens were placed in the aquatic biology reference collection (Table 8).

Table I. List of taxonomic consultants, area of expertise, and reason for selection; project taxonomists and area of expertise.

<u>Consultant Taxonomist</u>	<u>Area of Expertise</u>	<u>Reason for Selection</u>
Loxte, R. Dept. of Biology Bowling Green University Bowling Green, Ohio 43403	Diatoms	Author of "Environmental Requirements and Biological Tolerance of Freshwater Diatoms"
Reifen, G. Philadelphia Academy of Natural Sciences Nineteenth and the Parkway Philadelphia, PA 19103	Diatoms	Coeditor of diatom taxonomic keys for United States
Beck, Jr., W.M. School of Science and Technology Entomology & Structural Pest Control University, P.O. Box 111 Florida Agricultural & Mechanical U. Tallahassee, Florida 32307	Chironomidae	Author of chironomidae taxonomic keys
Cook, E.F. Dept. Entomology, Fisheries & Wildlife University of Minnesota St. Paul, MN 55108	Diptera, Odonata Megoptera & Neuroptera	Author of Diptera keys; familiar with regional taxa
Gundersen, R. Dept. of Biological Science St. Cloud State University St. Cloud, MN 56301	Coleoptera & Hymenoptera	Author of Coleoptera keys; familiar with regional taxa
Hilsenhoff, L. Dept. of Entomology University of Wisconsin Madison, Wisconsin 53706	Plecoptera	Author of Plecoptera keys; familiar with regional taxa

Lewis, P.A.
Environmental Monitoring
and Support Lab.
U.S. Environmental Protection Agency
Cincinnati, Ohio 45268

McCafferty, W.P.
Purdue University
Dept. of Entomology
West Lafayette, Indiana 47907

Unzicker, J.D.
Illinois Natural History Survey
Natural Resources Building
Urbana, Illinois 61801

Dr. J.C. Underhill
Dept. of Zoology
University of Minnesota
Minneapolis, MN 55455

Project Taxonomist

Johnson, H.D.

Lager, T.H.

Williams, S.H.

Mischuk, H.

McCullough, J.L.

Stenonema & Stenocrus

Author of Stenonema and
Stenocrus Key

Ephemeroptera

Author of Ephemeroptera
Keys; recent issue of
Lewis & Denver University of
Biology, Charlottesville
(Ephemeroptera Subcommittee
and Authors).

Trichoptera

Illinois Natural History Survey
Survey Techniques and Collections
Data Services Division
Section on the Diptera,
Entomology, Royal Ontario
Museum, 600 University Avenue
Toronto, Ontario, Canada
M5S 1A6

Fishes

Author of Fishes, American Midland
Naturalist, Vol. 96, No. 1, 1976

Area of Expertise

Invertebrates

Invertebrates

Invertebrates

Diatoms

Diatom and Invertebrates

Table 2. Comparison of diatom species proportional counts between project taxonomists.

TAXA	Site, Date:	SL-1-B 6-8-76		SL-1-B 6-8-76	
	Taxonomist:	McCulloch	Mischuk	McCulloch	Mischuk
<u>Achnanthes linearis</u>				54	2
<u>A. linearis</u> var. <u>curta</u>					2
<u>A. linearis</u> var. <u>pumila</u>			2		13
<u>A. lanceolata</u>		1			
<u>A. minutissima</u>		246	200	121	121
<u>A. stewartii</u>				4	
<u>Anomoneis serians</u>					1
<u>A. vitrea</u>		4	3	3	4
<u>Cyclotella</u> sp.				4	1
<u>Cymbella minuta</u>			1		
<u>C. sp.</u>		2		2	2
<u>Denticula elegans</u>			1		
<u>Diatoma tenue</u> var. <u>elongatum</u>			1		
<u>Eunotia curvata</u>				1	1
<u>E. elegans</u>					2
<u>E. exigua</u>					2
<u>E. flexuosa</u>					
<u>E. incisa</u>				1	1
<u>E. parallela</u>					1
<u>E. sp.</u>		2		18	17
<u>Fragilaria brevistriata</u>					1
<u>F. construens</u>					4
<u>F. construens</u> var. <u>pumila</u>					1
<u>F. construens</u> var. <u>venter</u>				3	1
<u>F. crotoncuais</u>					2
<u>F. leptostauron</u>					1
<u>F. pinnata</u>					1
<u>F. vaucheria</u>				25	
<u>F. sp.</u>					7
<u>Gomphonema acuminatum</u>		2			

Site: P U B

Date 6/8/76

TAXA	PROJECT	COUNT/DATE
<u>Aclanthus affinis</u>		2
<u>A. exigua</u> var. <u>constricta</u>	1	
<u>A. exigua</u> var. <u>heterovalva</u>		2
<u>A. lanceolata</u> var. <u>dubia</u>	1	
<u>A. linearis</u>	4	
<u>A. microcephala</u>	228	
<u>A. minutissima</u>		208
<u>Amphicoenis vitrea</u>	3	2
<u>Caloneis amphistrana</u>	1	1
<u>Cocconeis plecentula</u> var. <u>lineata</u>		1
<u>Cyathella microcephala</u>		1
<u>C. microcephala</u> var. <u>crassa</u>		1
<u>C. minuta</u>		1
<u>C. minuta</u> var. <u>silesiaca</u>		1
<u>C. sp.</u>	7	1
<u>Denticula elegans</u>		2
<u>Denticula sp.</u>	4	
<u>Diatoma tenue</u> var. <u>elongatum</u>		2
<u>Fragilaria construens</u>	1	1
<u>F. construens</u> var. <u>pumila</u>		2
<u>F. pinnata</u>	1	1
<u>F. sp.</u>	6	
<u>Goeponema parvulum</u>	2	1
<u>G. sp.</u>	6	
<u>Ilavicola capitata</u> var. <u>hungarica</u>	1	
<u>I. cryptocephala</u>	5	1

Table 3 cont'd.

Site: P-4-B cont'd.

Date: 6/3/76

<u>TAXA</u>	<u>FRUITS</u>	<u>COLONY TYPE</u>
<u>H. radiosa</u>	1	
<u>H. radiosa tenuia</u>		1
<u>H. selinaeana</u> var. <u>intermedia</u>		3
<u>H. secreta</u> var. <u>apiculata</u>		1
<u>H. sp.</u>	1	
<u>Hitzschia acicularis</u>	1	
<u>H. acicularis</u> var. <u>africana</u>		1
<u>H. bicata</u>		1
<u>H. dissipata</u>		1
<u>H. kützingeriana</u>		2
<u>H. sp. a</u>		3
<u>Pinnularia</u> sp.	1	
<u>Rhoicosphenia curvata</u>		1
<u>Synedra nana</u>		3
<u>S. rubens</u>		5
<u>Tabellaria fenestrata</u>		1

Table 3 cont'd.

Site: F-1-B

Date: 9/29/76

Project Director: Bechtold

Consultant Taxonomist: Reimer

Taxa	Project	Consultant
<u>Achnanthes</u> <u>deha</u>	0	8
<u>A.</u> <u>linearis</u>	103	17
<u>A.</u> <u>microcephala</u>	242	0
<u>A.</u> <u>minutissima</u>	0	249
<u>A.</u> <u>stewartii</u>	7	0
<u>Anomoeoneis</u> <u>serians</u> var. <u>brechysira</u>	0	4
<u>A.</u> <u>vitrea</u>	6	3
<u>Cyclotella</u> sp.	8	0
<u>Cymbella</u> <u>lunata</u>	0	4
<u>C.</u> <u>miruta</u>	0	2
<u>Cymbella</u> <u>naviculiformis</u>	0	2
<u>C.</u> <u>Cymbella</u> sp.	4	0
<u>Eunotia</u> <u>curvata</u>	2	0
<u>E.</u> <u>flexuosa</u>	2	2
<u>E.</u> <u>naegeli</u>	0	3
<u>E.</u> <u>exigua</u>	0	8
<u>E.</u> <u>incisa</u>	0	3
<u>E.</u> <u>pectinalis</u> var. <u>minor</u>	0	1
<u>E.</u> <u>tenella</u>	0	8
<u>E.</u> sp.	36	0
<u>Fragilaria</u> <u>construens</u> var. <u>humila</u>	0	23
<u>F.</u> <u>construens</u> var. 1 (c.f. sub- salina)	0	8
<u>F.</u> <u>construens</u>	6	0
<u>F.</u> <u>pinnata</u>	0	2
<u>F.</u> <u>vaucheriae</u>	50	0

Table 3 cont'd.

Site: I-1 B

Date: 9/29/76

Project Taxonomist: B. S. P. Koch
 Consultant: G. S. G. Jones

Taxon	Project	Consultant
<u>E. construens</u> var. <u>venifer</u>	6	0
<u>Gomphonema angustatum</u>	6	0
<u>G. angustatum</u> var. <u>sarcophagus</u>	0	2
<u>G. grunowii</u>	0	3
<u>G. parvulum</u>	4	8
<u>G. sp.</u>	4	0
<u>Helosira distans</u> var. <u>alpina</u>	0	4
<u>H. granulata</u>	4	0
<u>H. granulata</u> var. <u>agrestissima</u>	0	15
<u>Helosira sp.</u>	0	5
<u>Navicula arvensis</u>	0	4
<u>N. cryptocephala</u>	0	6
<u>N. exigua</u>	2	0
<u>N. gysingensis</u>	0	2
<u>N. minima</u>	0	3
<u>N. pupula</u>	2	0
<u>N. radiosa</u>	8	0
<u>N. taniula</u>	0	2
<u>N. sp.</u>	0	4
<u>Nitzschia acicularis</u>	0	2
<u>N. acicularis</u> var. <u>africana</u>	0	6
<u>N. bacata</u>	0	13
<u>N. frustulum</u> var. <u>perminuta</u>	0	4
<u>N. gracilis</u>	6	4
<u>N. kulzinciana</u>	0	14

Table 3 contd.

Site: F-F-R contd.

<u>taxa</u>	<u>Project</u>	<u>Consultant</u>
<u>N. palea</u>	18	2
<u>N. sp.</u>	8	0
<u>Stauroneis anceps var. gracilis</u>	0	2
<u>Surirella angusta</u>	0	2
<u>S. delicatissima</u>	0	5
<u>Synedra acus</u>	6	0
<u>S. minutula</u>	0	18
<u>S. nana</u>	0	4
<u>S. rumpens</u>	2	10
<u>S. tenera</u>	0	10
<u>Tabellaria fenestrata</u>	0	1
<u>T. focculosa</u>	14	9
	<u>TOTAL</u>	<u>45</u>

Table 3 contd.

Project Taxonomist: Lechler

Site: P. I. A.

Consultant Taxonomist: Bremer

Date: 9/21/76

Taxa	Project	Consultant
<u>Adiantum cingulifolium</u>	0	7
<u>A. lanceolatum</u>	2	2
<u>A. microcephalum</u>	104	0
<u>A. minutissimum</u>	7	121
<u>Amphipleura pellucida</u>	4	6
<u>Anomoeonides vitrea</u>	2	5
<u>Caloneis bacillum</u>	0	2
<u>Cocconeis placenticola</u>	17	0
<u>Plicatula</u> var. <u>limbata</u>	0	14
<u>Cymbella affinis</u>	123	0
<u>Microcypella</u> var. <u>crassa</u>	0	3
<u>C. minuta</u>	10	16
<u>C. prostrata</u> var. <u>everswaldii</u>	0	4
<u>C. tunida</u>	0	2
<u>C. sp.</u>	2	80
<u>Denticula</u> sp.	10	0
<u>elegans</u>	0	2
<u>Diatoma elongatum</u>	6	0
<u>tenue</u> var. <u>elongatum</u>	0	4
<u>Fragilaria construens</u>	2	8
<u>F. pinnata</u>	0	4
<u>Geophonema intricatum</u>	0	2
<u>G. parvulum</u>	18	8
<u>G. truncatum</u>	0	2
<u>Navicula absoluta</u>	0	2
<u>Crytocephala</u>	6	0

Table 3 contd.

Site: P-I A contd.

<u>Taxa</u>	<u>Project</u>	<u>Conspicuity</u>
<u>Digna</u>	0	1
<u>Gracilis</u>	2	0
<u>Pupula</u>	0	2
<u>Radiosa</u>	2	0
<u>Radiosa</u> var. <u>tentilla</u>	0	15
<u>Rhynoccephala</u>	4	0
<u>Rhynoccephala</u> var. <u>germainii</u>	0	7
<u>Salinarum</u> var. <u>intermedia</u>	0	4
<u>Secreta</u> var. <u>apicalata</u>	0	10
<u>Tripuactata</u>	0	2
sp.	22	
<u>Nitzschia acicularis</u>	10	0
<u>Acicularis</u> var. <u>africana</u>	0	2
<u>Aaphibia</u>	0	2
<u>Bacata</u>	0	10
<u>Capitalata</u>	8	0
<u>Confinis</u>	0	2
Sp. (c.f. <u>dissipata</u>)	0	3
<u>Frustulum</u>	0	2
<u>Frustulum</u> var. <u>subcalina</u>	0	2
<u>Gracilis</u>	0	4
<u>Kutzingiana</u>	0	37
<u>Palca</u>	54	13
Sp. (c.f. <u>paleacea</u>)	0	17
<u>Recta</u>	0	2
<u>Tropica</u>	0	3

Table 3 contd.

Site: P-I A contd.

Taxa	Project	Quantity
Sp. a.	6	44
Sp. b.	0	11
Sp. c.	0	3
Spp.	20	0
<u>Stauroneis anceps</u> c.f. <u>gracilis</u>	0	1
<u>Synedra nana</u>	0	3
<u>Ulna</u>	12	0
<u>Ruspens</u>	2	0
<u>Parasilica</u>	1	0
Sp.	50	0
<u>Cyclotella</u> sp.	2	0
<u>Gloerata</u>	<u>1</u>	<u>0</u>
TOTAL	29	45

Table 4. Species group (total count) comparison between project and consultant taxonomists.

Site: P-3-A

Project Taxonomist: Hite Ditchuk

Date: 9/21/16

Consultant Taxonomist: Lowe

TAXA	PROJECT	CONSULTANT
<u>Achnanthes exigua</u>		1
<u>A. lanceolata</u>	1	
<u>A. microcephala</u>	52	
<u>A. minutissima</u>		69
<u>Amphipleura pellucida</u>	2	
<u>Amphora ovalis</u> var. <u>affinis</u>		1
<u>Anomoeoneis villosa</u>	1	1
<u>Cocconeis placentula</u>	9	7
<u>C. placentula</u> var. <u>linearis</u>		1
<u>Cyclotella glauvata</u>	1	
<u>C. sp.</u>	2	
<u>Cymbella affinis</u>	64	
<u>C. microcephala</u>		3
<u>C. minuta</u>	5	12
<u>C. prostrata</u>		1
<u>C. sp.</u>	1	
<u>C. turgidula</u>		44
<u>Diatoma tenuis</u> var. <u>elongatum</u>	3	2
<u>Denticula</u> sp.	5	
<u>Eunotia septentrionalis</u>		1
<u>Fragilaria brevistriata</u> var. <u>inflata</u>		1
<u>F. construens</u>	1	
<u>F. construens</u> var. <u>venter</u>		2

Table 4 cont'd.

Site: F-3 A cont'd.

Date: 9/11/76

<u>TAXA</u>	<u>PERCENT</u>	<u>COUNT/TAXA</u>
<u>Campoplex angustatus</u>		10
<u>C. parvulus</u>	9	
<u>Belosira cubicus</u>		1
<u>Brachymeria capitata</u> var. <u>brachymeria</u>		3
<u>B. cryptoccephala</u>	3	
<u>B. gregaria</u>		1
<u>B. gracilis</u>	1	
<u>B. pupula</u>		2
<u>B. radiosa</u>	1	
<u>B. rhynchoccephala</u>	2	
<u>B. seminifera</u>		1
<u>B. sp.</u>	11	
<u>B. tripunctata</u>		2
<u>B. viridula</u> var. <u>rostellata</u>		2
<u>Brachymeria</u> sp. 1		4
<u>B. sp. 2</u>		1
<u>Bittschia aculeata</u>	5	4
<u>B. aculeata</u>		1
<u>B. bacata</u>		2
<u>B. capitellata</u>	4	
<u>B. conomic</u>		2
<u>B. dissipata</u>		1
<u>B. crustulum</u>		3
<u>B. gracilis</u>		2
<u>B. kutzingiana</u>		30

Table 1 contd.

Site: P. J. A. contd.

Date: 9/23/76

<u>WSP</u>	<u>PLANT</u>	<u>COLLECTOR</u>
<u>N. indica</u>	27	25
<u>N. paleacea</u>		9
<u>H. vossana</u>		1
<u>N. subliniaris</u>		1
<u>H. sp.</u>	10	
<u>N. tropica</u>		1
<u>Synedra parasitica</u>	1	
<u>S. radians</u>		2
<u>S. ruficeps</u>	1	
<u>S. uina</u>	6	1
<u>S. sp.</u>	25	
<u>Tabellaria sp.</u>	3	

Table 4 (cont'd.)

Site: P-1-A

Date: 5/16/77

Project: LAGOON/SLR - MICHIGAN

Consultant: LAGOON/SLR - FORD

TAXA	PROJECT	CONSULTANT
<u>Achnanthes minutissima</u>	179	189
<u>A. linearis</u>		1
<u>Anomoeoneis vitrea</u>	2	2
<u>Cocconeis pediculus</u>		1
<u>Cymbella lunata</u>	1	
<u>C. minuta</u>	10	6
<u>Denticula elegans</u>	3	
<u>Diatoma anceps</u>	2	
<u>Diatoma tenue</u> var. <u>elongatum</u>	9	5
<u>Fragilaria brevistriata</u>		1
<u>F. capucina</u>		1
<u>F. construens</u> var. <u>pumila</u>	1	
<u>F. construens</u> var. <u>venter</u>		2
<u>F. crotonensis</u>	4	
<u>F. vaucheriae</u>	1	4
<u>Gomphonema angustatum</u>	9	
<u>G. clevei</u>	2	
<u>G. dichotomum</u>		4
<u>G. parvulum</u>	3	17
<u>G. sp.</u>	2	
<u>Navicula capitata</u>	1	
<u>N. cryaloccephala</u>	1	7
<u>N. menisculus</u>		2
<u>N. radiosa</u> var. <u>tenella</u>	2	
<u>N. secreta</u> var. <u>apiculata</u>	7	

Table 2 (cont)

Site: P-V Acumld.

Date: 5/15/77

TAXA	PROJECT	CONSULTANT
<u>N. sp.</u>	1	
<u>N. trigonata</u>	1	
<u>Nitzschia amphibia</u>		1
<u>N. lacata</u>	1	
<u>N. capillata</u>		1
<u>N. discopota</u>	2	
<u>N. trusaleum</u>	1	
<u>N. hutzingeri</u>	1	
<u>N. sp.</u>	1	
<u>N. brevis</u>		1
<u>Synedra acus</u>		1
<u>S. amphicephala</u>	1	
<u>S. delicatissima</u>		3
<u>S. parasitica</u>	1	
<u>S. pulchella</u>		2
<u>S. pulchella</u> var. <u>lacerata</u>	1	
<u>S. rupeus</u>		1
<u>S. sp.</u>	6	
<u>S. tenera</u>		2
<u>S. ulna</u>	4	3

Table 4 (cont.)

Site: U-1A

Date: 5/17/77

Project: 1-10-77; 115-107
Conduct: 1-10-77; 1-10-77

<u>TAXA</u>	<u>PROTON</u>	<u>COMBUSTION</u>
<u>Actinanthus minutissima</u>	3	3
<u>Cyathella sp.</u>	2	
<u>Diatoma anceps</u>	2	
<u>D. laeve var. elongatum</u>	36	50
<u>Fragilaria construens var. venter</u>		1
<u>F. sp.</u>	1	
<u>F. vaucheriae</u>	14	16
<u>Gomphonema angustatum</u>	18	49
<u>G. parvulum</u>	1	2
<u>G. sp.</u>	7	
<u>Havicula cryptocephala</u>		1
<u>H. viridula var. avinacea</u>		1
<u>Hitzschia kutzingiana</u>	1	
<u>il. sp.</u>	1	
<u>il. tropica</u>		1
<u>il. sp. 1</u>		1
<u>Sorirella brigdalei</u>		1
<u>Synedra amphicephala</u>		1
<u>S. delicatissima</u>	4	1
<u>S. minuscula</u>	17	2
<u>S. nana</u>		1
<u>S. acus</u>	1	
<u>S. pulchella</u>		3
<u>S. pulchella var. lacunata</u>	5	
<u>S. radians</u>		5

Site 1 - contd.

Site 1 - contd.

Date / /

<u>TAXA</u>	<u>NUMBER</u>	<u>PERCENT</u>
<u>S. rugens</u>	4	12
<u>S. varia</u>		2
<u>S. longica</u>	3	6
<u>S. alba</u>	132	93

Table 1 cont'd.

Site: D-1-A cont'd.

Date: 5/17/77

<u>TAXA</u>	<u>PROJECT</u>	<u>COMPARISON</u>
<u>M. sp.</u>	1	
<u>Navicula lanceolata</u>		1
<u>N. notha</u>	1	
<u>N. seminulum</u>		1
<u>N. sp.</u>	1	
<u>Nitzschia frustulum</u>	1	
<u>N. gracilis</u>		1
<u>N. palea</u>		1
<u>Synedra acus</u>		2
<u>S. delicatissima</u>	12	1
<u>S. minuscula</u>	5	1
<u>Synedra nana</u>		3
<u>S. radians</u>		6
<u>S. ruspens</u>		2
<u>S. sp.</u>	3	
<u>S. tenera</u>		4
<u>S. ulna</u>	12	4
<u>Tabellaria fenestrata</u>	5	1
<u>T. flocculosa</u>	21	3

Coll. 4 - 1960

Site: B-3-A

Date: 5/11/77

Project: Terrestrial Insects

Continent: Colombia

<u>TAXA</u>	<u>PROBES</u>	<u>COUNTS</u>
<u>Achnanthes flexella</u>	5	3
<u>A. minutissima</u>	136	162
<u>Anemoneopsis vitrea</u>	10	8
<u>Cyclorella sp.</u>	1	
<u>Cydella angustata</u>		1
<u>C. cesatii</u>	20	8
<u>C. cistula</u>		1
<u>C. minuta</u>	1	
<u>C. sp.</u>	1	
<u>C. sp. 1</u>		1
<u>C. sp. 2</u>		1
<u>Denticula elegans</u>	4	
<u>Dialema tenue var. elongatum</u>	29	24
<u>Eunotia curvata</u>		1
<u>Fragilaria capucina</u>		4
<u>F. construens</u>	1	
<u>F. construens var. pusilla</u>	2	1
<u>F. construens var. venter</u>		1
<u>F. vaucheriae</u>		1
<u>Gomphonema angustatum</u>	6	
<u>G. intricatum</u>		4
<u>G. sp.</u>	2	
<u>G. truncatum</u>		1
<u>G. sp. 1</u>		1
<u>Melosira ambigua</u>		1
<u>M. granulata</u>	1	

Table 5. Comparison of diatom species proportional counts between consultants and between consultants and project taxonomists for sites 9-1-F-A, 21 September 1976.

Taxa	Project Taxonomist		Consultant Taxonomist	
	Count #1	Count #2	Count #1	Count #2
<u>Amphioxys diarex</u>		1		1
<u>A. radiata</u>		2		2
<u>A. lanceolata</u>			1	
<u>A. lanceolata</u> var. <u>dubia</u>	2		1	
<u>A. viridula</u>		1		
<u>A. lindahis</u> var. <u>curta</u>		1		
<u>A. microlepis</u>	71	59	62	59
<u>A. microlepis pallidula</u>		1	2	
<u>Amphioxys ovalis</u>		1		1
<u>Amphioxys virens</u>		1	3	2
<u>Caloneis acutilium</u>		1	1	
<u>Caloneis acuminata</u>				14
<u>C. diademata</u> var. <u>virata</u>	5	5	7	2
<u>C. diademata</u>	2	4		5
<u>C. diademata</u> var. <u>brassii</u>			1	
<u>C. diademata</u>			3	
<u>C. diademata</u> var. <u>autocostata</u>				3
<u>C. diademata</u>			1	0
<u>C. sp.</u>				
<u>Amphioxys diademata</u> var. <u>brassii</u>			10	
<u>Amphioxys diademata</u> var. <u>brassii</u>			2	

Taxa Project Taxonomist Consultant Mycorrhizal

	Count #1	Count #2	Count #3	Refers	Level
<u>Denticula elegans</u>				1	
<u>Diplazis finlayi</u>	1				1
<u>Eurotia septentrionalis</u>		1			
<u>E. sp.</u>					
<u>Ergatis brevispina</u> var. <u>inflata</u>				4	1
<u>E. Donstruens</u>					
<u>E. constrictus</u> var. <u>glabra</u>		1			
<u>E. constrictus</u> var. <u>helveticus</u>	2				3
<u>E. Leptostreum</u> var. <u>glabra</u>	1				
<u>E. pilosella</u>				2	
<u>E. sp.</u>	3		2	1	
<u>Ergonome incrustans</u>	10	1			20
<u>E. intricatum</u>				1	
<u>E. Ranzolium</u>	2	1	2	1	
<u>E. truncatum</u>				1	
<u>E. sp.</u>	6	3			
<u>Helosira orbicula</u>					1
<u>H. granulata</u>				1	
<u>Helicula absoluta</u>					
<u>H. levensis</u>	1	1	4		
<u>H. declinata</u> var. <u>humboldtii</u>					1
<u>H. oryctolobosella</u>					
<u>H. sp.</u>				1	
<u>H. Swinhoei</u>					3
<u>H. munda</u>					
<u>H. Becki</u>	2				
<u>H. sp.</u>					
<u>H. sp.</u>				1	3

Table 5 cont'

Taxa	Project Taxonomist			Consultant Taxonomist	
	Count #1	Count #2	Count #3	Reimer	Love
<u>N. rufosa</u> var. <u>tenella</u>		3	1	5	
<u>N. rhynchospora</u>	4	2	8	4	
<u>N. rhynchospora</u> var. <u>gambelii</u>				1	
<u>N. salinarum</u> var. <u>intermedia</u>			1	2	
<u>N. setacea</u> var. <u>apiculata</u>			1	5	
<u>N. schiauleri</u>					1
<u>N. trifurcata</u>				1	3
<u>N. viridula</u> var. <u>rostellata</u>					2
<u>N. sp.</u>	1		1		10
<u>Nitassonia acicularis</u>	4	3	5		5
<u>N. acicularis</u> var. <u>africana</u>				1	
<u>N. arpatzia</u>	3	5	2	1	2
<u>N. pacata</u>	1	3	3	10	4
<u>N. communis</u>					4
<u>N. confinis</u>				1	
<u>N. missinaga</u>	2	2	7	2	2
<u>N. frustulum</u>	1	1		1	5
<u>N. frustulum</u> var. <u>subulifera</u>				1	
<u>N. gracilis</u>				2	4
<u>N. Kuepzingiana</u>	14	34	30	19	30
<u>N. palae</u>	4	2		7	50
<u>N. paleacea</u>	17	9		9	13
<u>N. neera</u>				1	
<u>N. rotunda</u>					2
<u>N. sublinearis</u>					2

Table 5 cont'd

Taxa	Project Taxonomist			Consultant Taxonomist	
	Count #1	Count #2	Count #3	Reimar	Love
<u>S. tropica</u>				2	2
<u>S. sp.</u>	23	24	10	30	
<u>Stauroneis anceps</u>				1	
<u>Synechis minuscula</u>	1	1	1	2	
<u>S. rana</u>				3	
<u>S. radians</u>				1	
<u>S. ulva</u>		1	1		
<u>S. sp.</u>	0				
<u>Tarellaria flocculosa</u>	1				
Total Taxa	29	33	27	75	36

Table IV. Comparison of diatom genera proportional counts at the upper level and lower compliance and below compliance sites and project locations for site 1 on P.A. 29 September 1976.

Taxa	Project	Taxonomic	
		Level	Region
Achnanthes	73	70	53
Amphipleura	P*		
Ampicora	P	P	
Araneoneis	P	1	3
Colanais	P		1
Cocconeis	5	8	7
Cymbella	70	59	53
Diatoma	1	2	2
Diploneis	P		
Enotia	P	P	
Fraxillaria	3	2	6
Graphionema	9	10	6
Kelosira	P	1	
Navicula	11	7	22
Nitzschia	73	81	83
Stauroneis	P		P
Synedra	4		2
Tabellaria	P		
Total Taxa	18	12	12

*P - less than one frustule present in sample.

Table 1. Comparison of insecticide identifications between original and quality control analysts.

Site: M-30-A
Date: 4/11/76

Original analyst: J. L. Taylor
Quality control analyst: S. Williams

Taxa	Original	Quality Control
Diptera	1	
Simuliidae	27	20
Trichoptera	4	
Psychomyiidae		1
Psychomyia flavida	1	
Hydropsychidae	20	170
Hydropsyche sp.	25	35
H. cuning	7	
Chenilobryche sp.	121	21
Phryganeidae	1	
Phryganea cf. cinerea	1	
Polycentropodidae		
Baureclipsis sp.	2	2
Limnephilidae		1
Ephemeroptera		
Meleagrinidae		
Meleagris sp.	32	39
Stenonema vicarium	8	
Baetidae		116
Baetis sp.	143	51
B. hageni	12	
B. infersularis	8	
B. levifans	3	
Leptophlebiidae	53	53
Ephemerellidae		
Ephemerella sp.	14	13
Tricorythidae		
Tricorythodes sp.	22	23
Caenidae		
Caenis sp.	2	1
Odonata		
Anisoptera		1
Corduliidae		
Epi <theca< th=""></theca<>	1	
Zygoptera		1
Calopterygidae		
Calopteryx sp.	4	3
Megaloptera	1	
Corydalidae		1

Table 7 cont.
Site: T3H A cont.

Taxa	Original	Quantity Coll.
Collembola		1
Dytiscidae	2	1
Hydrophilidae	3	3
Hydrophilus sp.	2A	
Hydrophilus sp.	1A	
Hydrophilidae		
Hydrophilus sp.		2
Flutidae		
Anacropelma glabratus	2A	2A
Glyptotendipes sp.	2	2
G. facitoides	2A	
Stenotendipes sp.	2	4
S. crenata	1A	
Hemiptera		
Cixiidae		
Cixius sp.	2	2
Cixius sp.	3	2
Kerriatolus sp.	17	11
Veliidae		
Phragmatelia sp.	20	20
Lepidoptera	1	2
Collembola	3	1
Acaric	3	
Amphipoda		
Talitridae		
Hyalolella azteca	1	1
Pelecypoda		
Sphaeriidae	1	1
Gastropoda		
Physidae		
Physa sp.	1	1
Hirudinea	1	1
Decapoda	2	2
Oligochaeta	1	
Terrestrial Adults	269	272

Table 1 (cont.)

Site: SR-3 (1968)

Taxa	Original	Quality Control
Ephemeroptera		
Ephemerella sp.	1	2
E. attemorata	1	
E. deficiens	2	
E. scitula	1	
Tricoptera		
Tricoptodes sp.	1	1
Cocci		
Cocci sp.	9	6
Ephemeroptera		
Ephemerella sp.		5
E. strians	6	
Odonata		
Gomphidae	1	3
Dromogomphus soligosus	1	
Hemilus bicostylus	2	1
Gomphus sp.		2
O. vancouverensis	5	
Coleoptera		
Gyrinidae		
Gyrinus sp.	2	
Elmidae		
Macronychus glabratus	9	1
Stenelmis sp.	3	4
Stenelmis crenata	6	
Amphipoda		
Talitridae		
Hyalella azteca	4	2
Hirudinea	2	2
Pelecypoda		
Sphaeriidae	25	26
Gastropoda	2	3
Physidae		
Physa sp.	2	1
Hexapoda		
Veliidae		
Rhagovelia sp.	31	23
Belostomatidae		
Leithocerus sp.	4	3
Cerridae		
Limnogonus sp.	2	2

CAPI-7 contd.

Site: 9-140
Date: 1/24/76

Origin: 1. Louisiana: 1. 1973
Quality Control: 1. 1973

Taxa	Original	Quality Control
Diptera		
Rhagothripidae		
<i>Atherix variabilis</i>	3	3
Simuliidae	5	5
Trichoptera		
Hydropsychidae	7	5
<i>Hydropsyche</i> sp.	21	9
<i>H. cunna</i>		11
Hydropsilidae		
<i>Hydropsila</i> sp.	1	1
Brachycentridae		
<i>Hemiptera</i> sp.	8	5
Lepidostomatidae		
<i>Lepidostoma</i> sp.	1	2
Leptoceridae		
<i>Cnephia</i> sp.	2	1
<i>C. raculata</i>		1
<i>Oreocera</i> sp.		1
<i>O. chrysocens</i>	2	
Plecoptera		
2		1
Heurysidae		
<i>Asphincura</i> Linda		1
Perlidae		1
<i>Acronyia</i> sp.	11	
<i>A. lycorias</i>	3	3
Ephemeroptera		
1		1
Heptageniidae		
<i>Stenonema interpuclatum</i>	1	
<i>S. cordatum</i>	1	
<i>S. mimeltonka</i>		1
<i>Heptagenia hebe</i>	3	1
Baetidae	23	
<i>Baetis</i> sp.	1	18
<i>B. intercalaris</i>		5
<i>Closon</i> sp.		2
<i>Pseudocloson</i> sp.	1	
<i>P. carolina</i>	1	
<i>P. cingulatum</i>	2	1
Ephemerellidae		
<i>Ephemerella attenuata</i>	2	
<i>E. bicolor</i>	1	
<i>E. invaria</i>	1	
<i>E. serrata</i>	18	20
<i>E. sordida</i>	5	

Table 7 cont'd.

Site: 01-00 cont'd.

Taxa	Original	Quality Corrected
Coenidae		
Coenid. sp.	4	4
Ephemeridae		
Ephemera sp.	1	1
Coleoptera		
Elmidae	1	2
Opilicorynus sp.		
Hagelius sp.	4	
Stenelmis sp.	2	2
S. granata		2
Oligochaeta	1	1
Pelecypoda		
Saxeridae	2	2
Terrestrial Insects	1	3

total = 7 conid.

total = 121 conid.

taxa	Original	Quaternary Collection
Phlebotominae	4	
Perilidae		2
<i>Acanomyia</i> sp.		1
<i>A. tygonias</i>	3	1
<i>Peromyzina</i> sp.		2
<i>P. media</i>	10	7
Peritoididae		
<i>Isoperla drusila</i>	24	1
<i>I. frizoni</i>	4	18
Chloroperlinae		
<i>Hastagella</i> sp.	2	
<i>H. brevis</i>		1
Ephemeroptera		
Siphonuridae		
<i>Isonychia</i> sp.	1	1
Heptageniidae		
<i>Ephonia</i> sp.	5	2
<i>Siphonura</i> sp.		7
<i>S. interconcolorata</i>	6	
<i>Siphonura</i> sp.		30
<i>S. lucum vicillipolus</i>	2	2
<i>S. tubum</i>	8	
<i>S. rubrum</i>	4	2
<i>S. terminatum</i>	28	
Baetidae	21	32
<i>Baetis</i> sp.	25	
<i>B. frondalis</i>	1	10
<i>Pseudocloeon</i> sp.	5	8
<i>P. carolina</i>	15	
<i>P. dubium</i>	1	
<i>P. parvulum</i>	6	
Leptophlebiidae		29
<i>Paraleptophlebia mollis</i>	14	
Ephemerellidae		
<i>Ephemerella</i> sp.	2	4
<i>E. bicolor</i>	5	
Caenidae		
<i>Caenis</i> sp.	1	1
Odonata		
Aeschnidae		
<i>Boyeria</i> sp.	3	3

Table 7 cont'd.

Site: ME-1-01 cont'd.

Taxa	Original	Quality Corrected
Coleoptera		
Cyrillidae		
<u>Cyrtus</u> sp.	2	2
Flatidae		
<u>Macronychus glabratus</u>		2
<u>Opliosorus</u> sp.	5	
<u>O. pallidus</u>	1	
<u>Stenelmis</u> sp.	12	12
Hemiptera		
Gerridae		
<u>Gerris</u> sp.	6	3
Megaloptera		
Corydalidae		
<u>Rigoria</u> sp.	3	
Pelecypoda		
Aphaeridae	6	4
Gastropoda		
Physidae		
<u>Physa</u> sp.	2	2
Heratoda		2
Oligochaeta	2	
Hirudinea		3
Turbellaria	1	
Terrestrial Insects	2	

Table 7 cont'd.

Site: CR-3-01
Date: 1/7/16Original Formulas: 7. Lager
Quality Control Laboratory: F. Lager

Taxa	Original	Quality Control
Diptera		
Simuliidae		4
<i>Fusculina</i> sp.	3	
<i>Simulium</i> sp.	7	
Tipulidae		
<i>Tipula</i> sp.	1	
Trichoptera		
Polycentropodidae		
<i>Neaerolipsis</i> sp.	41	37
Philopotamidae		
<i>Chilarra</i> sp.		1
<i>C. socia</i>	1	
<i>C. obscura</i>	6	6
Hydropsychidae		
<i>Hydropsyche</i> sp.	103	155
<i>H. betteni</i>	3	
<i>H. cf. cunius</i>		17
<i>Cheumatopsyche</i> sp.	129	135
Plecoptera		
Perlidae		
<i>Acroneuria</i> sp.	4	2
<i>A. lycorius</i>	3	1
<i>Paragnetina media</i>	9	8
<i>Perlostia</i>	11	7
Ephemeroptera		
Heptageniidae		
<i>Stenonema</i> sp.	1	
<i>S. exiguum</i>	4	
<i>S. quinquispinum</i>	1	1
<i>S. rubra</i>	2	1
<i>S. terminatum</i>	1	
Baetidae		
<i>Baetis</i> sp.	2	6
<i>B. levitans</i>	5	
<i>Pseudocloeon carolina</i>	1	2
Leptophlebiidae		
<i>Paraleptophlebia mollis</i>	4	
<i>P. cf. praepedita</i>	1	3
<i>Choroterpes basalis</i>	1	

Table 8. Fish species in the aquatic biology collection collection

Tota

Catostomidae

Catostomus commersoni

Centrarchidae

Micropetrus salmoides

Cottidae

Cottus bairdi

Cyprinidae

Chrosomus nigricans

C. ves

Koleidionus crysoleucas

R. volucellus

Pimephales promelas

Rhinichthys cataractae

Semotilus atromaculatus

S. marginata

Gasterosteidae

Culaga inconstans

Ictaluridae

Noturus gyrinus

Umbridae

Umbra limi

Appendix D-12. List of Sampling Sites, Locations and Collection Year

Site	Township Range, Section	Stream Order	Stream Name	Site Classification	Years Sampled
BB-1	T.61, R.12, S.35	1	Hannaed Creek	S	1976
BC-1	T.61, R.15, S.35	2	Bear Creek	SCS ²	1977
BI-1	T.62, R.12, S.23	3	Deer Island River	S	1976, 1977
CA-1	T.59, R.10, S.12	2	Coyote Creek	SCS ²	1977
D-1	T.60, R.12, S.9	3	Dunka River	P	1976, 1977
D-2	T.60, R.12, S.27	3	Dunka River	T	1976, 1977
D-3	T.59, R.12, S.16	1	Dunka River	SCS ²	1977
DC-1	T.61, R.11, S.28	3	Denley Creek	SCS ²	1977
E-1	T.60, R.15, S.25	3	Embarrass River	P	1976, 1977
E-2	T.60, R.14, S.15	3	Embarrass River	T	1976, 1977
F-1	T.62, R.11, S.24	2	Filson Creek	S, SCS ²	1976, 1977
F-2	T.62, R.11, S.25	1	Filson Creek	SCS ²	1977
I-1	T.61, R.9, S.6	5	Isabella River	T	1976, 1977
K-1	T.63, R.11, S.3	5	Kawishiwi River	P	1976, 1977
K-2	T.63, R.12, S.26	4	Shagawa River	S	1976, 1977
K-3	T.63, R.11, S.20	5	Kawishiwi River	T	1976
K-4	T.63, R.11, S.32	5	Kawishiwi River	T	1976
K-5	T.62, R.11, S.31	5	Kawishiwi River	S	1976, 1977
K-6	T.63, R.10, S.24	4	Kawishiwi River	T	1976, 1977
K-7	T.62, R.11, S.23	5	Kawishiwi River	T	1976, 1977
K-8	T.62, R.10, S.6	5	Kawishiwi River	P	1976, 1977
KC-1	T.61, R.11, S.17	2	Keeley Creek	S, SCS ²	1976, 1977
KC-2	T.61, R.11, S.10	1	Keeley Creek	SCS ²	1977
LI-1	T.61, R.9, S.29	3	Little Isabella River	SCS ¹	1977
LI-2	T.59, R.8, S.5	2	Little Isabella River	SCS ¹	1977
LI-3	T.59, R.8, S.9	1	Little Isabella River	SCS ¹	1977
R-1	T.60, R.11, S.34	1	Rip Creek	SCS ²	1977
RR-1	T.61, R.10, S.31	2	Rina Creek	SCS ²	1977
NR-1	T.56, R.14, S.23	3	North Branch Whiteface River	SCS ²	1977
P-1	T.58, R.15, S.13	4	Partridge River	P	1976, 1977
P-2	T.58, R.14, S.9	4	Partridge River	S	1976, 1977