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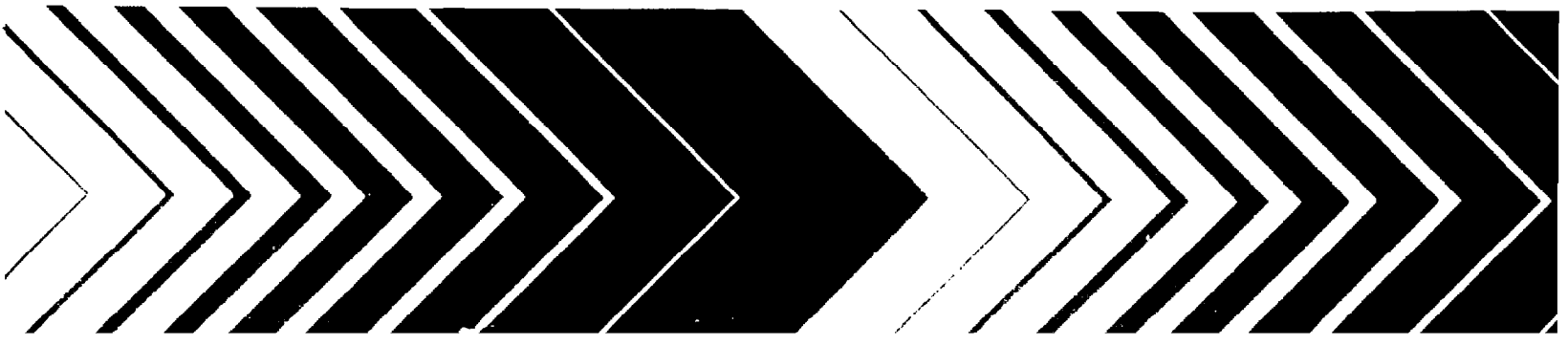
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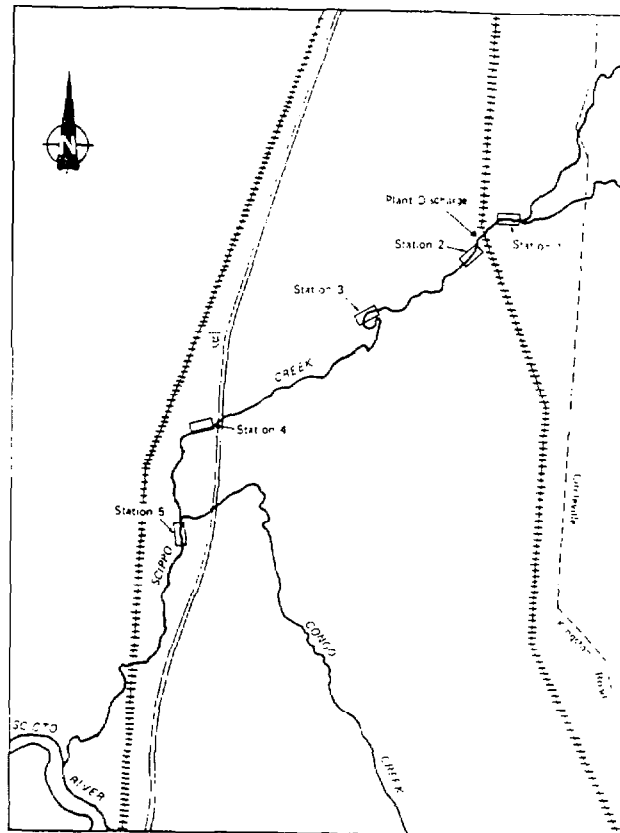
Research and Development



# Validity of Effluent and Ambient Toxicity Tests for Predicting Biological Impact, Scippo Creek, Circleville, Ohio



VALIDITY OF EFFLUENT  
AND AMBIENT TOXICITY TESTS  
FOR PREDICTING BIOLOGICAL IMPACT,  
SCIPPO CREEK, CIRCLEVILLE, OHIO



Edited by

Donald I. Mount, Ph.D. (a)  
and  
Teresa J. Norberg-King (a)

(a) U.S. Environmental Protection Agency. Environmental Research Laboratory--Duluth, 6201 Congdon Blvd., Duluth, Minnesota 55804.

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LIST OF INVESTIGATORS

LABORATORY TOXICITY TESTS

Donald I. Mount(a) and Teresa J. Norberg-King(a)

TIME-OF-TRAVEL STUDY AND FLOW MEASUREMENTS

Jonathan C. Yost(b)

PERIPHYTIC COMMUNITY

Ronald J. Bockelman(b)

BENTHIC MACROINVERTEBRATE COMMUNITY

Michael T. Barbour(b)

FISH COMMUNITY

David A. Mayhew(b) and David P. Lemarie(b)

COMPARISON OF LABORATORY TOXICITY DATA AND  
RECEIVING WATER BIOLOGICAL IMPACT

Teresa J. Norberg-King(a) and Nelson A. Thomas(a)

PRINCIPAL INVESTIGATOR: Donald I. Mount, Ph.D.(a)

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(a) U.S. Environmental Protection Agency. Environmental Research Laboratory--Duluth, 6201 Congdon Blvd., Duluth, Minnesota 55804.

(b) EA Engineering, Science, and Technology, Inc. (formerly called Ecological Analysts, Inc.), Hunt Valley/Loveton Center, 15 Loveton Circle, Sparks, Maryland 21152.

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## FOREWORD

The Complex Effluent Toxicity Testing Program was initiated to support the developing trend toward water quality-based toxicity control in the National Pollutant Discharge Elimination System (NPDES) permit program. It is designed to investigate, under actual discharge situations, the appropriateness and utility of "whole effluent toxicity" testing in the identification, analysis, and control of adverse water quality impact caused by the discharge of toxic effluents.

The four objectives of the Complex Effluent Testing Program are:

1. To investigate the validity of effluent toxicity tests in predicting adverse impact on receiving waters caused by the discharge of toxic effluents.
2. To determine appropriate testing procedures which will support regulatory agencies as they begin to establish water quality-based toxicity control programs.
3. To provide practical case examples of how such testing procedures can be applied to a toxic effluent discharge situation involving a single discharge to a receiving water.
4. To field test short-term chronic toxicity tests including the test organisms, Ceriodaphnia reticulata and Pimephales promelas.

Until recently, NPDES permitting has focused on achieving technology-based control levels for toxic and conventional pollutants in which regulatory authorities set permit limits on the basis of national guidelines. Control levels reflected the best treatment technology available, considering technical and economic achievability. Such limits did not, nor were they designed to, protect water quality on a site-specific basis.

The NPDES permits program, in existence for over 10 years, has achieved the goal of implementing technology-based controls. With these controls largely in place, future controls for toxic pollutants will, of necessity, be based on site-specific water quality considerations.

Setting water quality-based controls for toxicity can be accomplished in two ways. The first is the pollutant-specific approach which involves setting limits for single chemicals, based on laboratory-derived no-effect levels. The second is the "whole effluent" approach which involves setting limits using effluent toxicity as a control parameter. There are advantages and disadvantages to both approaches.

The "whole effluent" approach eliminates the need to specify a limit for each of thousands of substances that may be found in an effluent. It also includes all interactions between constituents as well as biological availability. Such limits determined on fresh effluent may not reflect toxicity after aging in the stream and fate processes change effluent composition. This problem is less important since permit limits are normally applied at the edge of the mixing zone where aging has not yet occurred.

To date, eight sites involving municipal and industrial dischargers have been investigated. They are, in order of investigation:

1. Scippo Creek, Circleville, Ohio
2. Ottawa River, Lima, Ohio
3. Five Mile Creek, Birmingham, Alabama
4. Skeleton Creek, Enid, Oklahoma
5. Naugatuck River, Waterbury, Connecticut
6. Back River, Baltimore Harbor, Maryland
7. Ohio River, Wheeling, West Virginia
8. Kanawha River, Charleston, West Virginia

This report presents the site study on Scippo Creek, Circleville, Ohio, which was conducted in August 1982. The stream is small and receives discharge from one industry.

This project is a research effort only and has not involved either NPDES permit issuance or enforcement activities.

Rick Brandes  
Permits Division

Nelson Thomas  
ERL/Duluth

PROJECT OFFICERS  
Complex Effluent Toxicity  
Testing Program

## EXECUTIVE SUMMARY

EPA recently issued a water quality-based policy which provides for control of the discharge of toxic substances through the use of numerical criteria and effluent toxicity limits in NPDES permits. This policy is the first broad scale effort to use effluent toxicity limits in the NPDES permit program and a scientific basis for this approach is needed.

This report describes the first site study on Scippo Creek at Circleville, Ohio, which receives only one discharge from a chemical resins plant using batch operations. Scippo Creek is a small sunfish/bass stream flowing through an agricultural area in central Ohio. Previous biological studies by the State of Ohio had shown measurable adverse impact below the outfall and a grab sample of effluent tested before the study indicated high toxicity. Effluent dilution toxicity tests were run with two test species both onsite and at a remote laboratory. In addition, toxicity tests were conducted onsite on ambient samples from four river stations. Biological studies were conducted at those stations and included benthic macroinvertebrates, fish, and periphyton.

The results of this study revealed no biological impact in the stream except for a small area of changed species composition at the outfall which is presumed to be caused by a physical change in the substrate from settled precipitate which clogged the sediment interstitial spaces. No toxicity to C. reticulata, fathead minnows, or resident species was measured in the 100 percent effluent.

The processed waste is held in a detention tank after treatment. Several times each week the tank is pumped and treated waste is discharged. The initial grab sample of effluent was apparently taken when process waste was being discharged, but the composite sampling process used in this study reduced peak concentrations. Importantly, the composite sample toxicity results best predicted the lack of community impact. New treatment equipment had been installed after a previous biological survey which was conducted by Battelle Laboratories (1971). Operation of this

new equipment may have improved waste treatment and presumably that is why little or no effect was found in the stream. Correctly predicting no impact to a receiving stream is a requirement of tests used for regulatory purposes.

## QUALITY ASSURANCE

Coordination of the various studies was completed by the principal investigator preceding and during the onsite work. A reconnaissance trip was made to the site before the study and necessary details regarding transfer of samples, specific sampling sites, dates of collections, and measurements to be made on each sample were delineated. The evening before the study began, a meeting was held onsite to clarify again specific responsibilities and make last minute adjustments in schedules and measurements. The mobile laboratory was established as the center for resolving problems and adjusting of work schedules as delays or weather affected the completion of the study plans. The principal investigator was responsible for all Quality Assurance-related decisions onsite.

All instruments were calibrated by the methods specified by the manufacturers. For sampling and toxicity testing, the protocols described in the referenced published reports were followed. Where identical measurements were made in the field and laboratory, both instruments were cross-calibrated for consistency.



## 1. INTRODUCTION

This study was the first site investigated in the Complex Effluent Testing Program. The site was chosen because the stream was small and effluent-dominated by one discharge. Equipment and methods had been untried for onsite testing and the mobile laboratory had just been assembled. Many logistical and procedural details had to be developed before more complex sites could be attempted. Special emphasis was placed on improving test procedures and simplifying equipment needs, as well as meeting the major objective which was to use toxicity tests to predict expected biological impact in the stream.

This report is organized into sections corresponding to the project tasks. Following an overview of the study design and a summary of the description of the site, the chapters are arranged into toxicity testing, hydrology, and ecological surveys. An integration of the laboratory and field studies is presented in Chapter 8. All methods and support data are included in the appendixes for reference.

## 2. STUDY DESIGN AND SITE DESCRIPTION

The effluent evaluated was from a plastics resin plant in central Ohio that discharged to a small stream in a flat, rich agricultural area. There were no other known discharges to the stream. The influent was taken from a well and most of the discharge was cooling water. The process water was treated in rotating biological contactors and held in tanks capable of holding the waste volume generated in 30 days. Several times each week, the treated waste was pumped into the cooling water discharge and then discharged into Scippo Creek. The temperature of the discharge was considerably cooler than the stream at the time of the study in July 1982. There was a substantial amount of precipitate from the well water observed below the outfall.

Study components included 7-day Ceriodaphnia reticulata toxicity tests on samples from each of four river stations and various concentrations of the effluent; 7-day larval growth tests on fathead minnows in various concentrations of the effluent; tests of indigenous species; ambient toxicity caging studies; time-of-travel analysis for the effluent; and quantitative assessment of the benthic macroinvertebrate, periphytic, and fish communities. The study was conducted 9-16 August 1982.

The study area on Scippo Creek was located above the confluence with the Scioto River. Scippo Creek (Figure 2-1) is shallow (less than 0.6 m in depth) and 10-20 m in width at the study area. Pool areas predominate with periodic riffle sections along its length. The study area incorporated 6.7 river kilometers (RK) of stream and five sampling locations. Habitats sampled were riffles and pools for benthic macroinvertebrates and a combination of both for fish. Periphyton samples were taken from run areas or pools where available. The station locations as depicted in Figure 2-1 are:

Station 1--0.28 km upstream of the effluent outfall. The sampling station was located in a straight stretch, approximately 20 m in length, downstream of a bend in the creek. The station was shaded approximately 80 percent by deciduous canopy. Stream width was approximately 15 m. The riffle substrate consisted of pebble-cobble, with

varying amounts of sand deposited among the rocks. The substrate of the pools was primarily sand, with small amounts of mud. The pools were relatively free of debris but did contain some leaf packs.

- . Station 2--0.1 km downstream of the outfall. The station was located approximately 10 m downstream of a slight bend in the creek. Shading was provided by a deciduous canopy which covers about 40 percent of the station. Stream width was about 10 m. The substrate of the riffle consisted of pebble and gravel, with some cobble overlying bedrock. Pools were deepest at this station and contained some debris (e.g., logs, branches) and leaf packs.
- . Station 3--1.3 km downstream of the outfall. The sampling station had little canopy cover (less than 25 percent) and was approximately 15 m in width. Substrate of the riffle area was primarily pebble and gravel, with pockets of sand. The pools had sand-mud bottoms and contained a fallen tree.
- . Station 4--3.7 km downstream of the outfall and immediately downstream of the U.S. Rte. 23 bridge. Canopy cover at Station 4 was near 100 percent. The creek width was approximately 20 m. The riffle consisted of cobble and pebble substrate with some sand. The substrate of the pools was sand with little debris.
- . Station 5--5.3 km downstream of the outfall and immediately downstream of the confluence with Congo Creek. The canopy cover at Station 5 was about 90 percent. The riffle substrate was cobble and pebble overlying bedrock. Some sand pockets were also present. The width of the creek at this station was approximately 20 m. The pools were sand substrate and free of debris.

See Table C-1 for a pool vs. riffle habitat description at the sampling locations.

Temperature, dissolved oxygen, specific conductance, and pH were monitored during biological collections and the first half of the fish caging study. The instruments used for water quality measurements were a Hydro-lab Model 1041, a YSI Model 57 Dissolved Oxygen Meter, and a YSI Model 33 Salinity-Conductivity-Temperature Meter. Dissolved oxygen ranged from 7.9 to 13.6 mg/liter, with many readings above 100 percent saturation. The pH ranged from 7.5 to 8.5.

The temperature effect from the discharge was variable, sometimes causing virtually no change in receiving water temperature, and at other times decreasing the temperature at Station 2 by 8 C. Although diel temperature patterns were not studied, the water had returned to normal temperature range at Station 4.

At Station 1, conductivities from 550 to 590  $\mu\text{mhos}$  were recorded over 10-13 August. However, the discharge caused rapid, large variations in conductivity downstream. One such event occurred on 12 August at Station 2, when conductivity increased from 806 to 1,229  $\mu\text{mhos}$  in 10 minutes, and to 1,535  $\mu\text{mhos}$  30 minutes later. Approximately 8 hours later, a reading of 630  $\mu\text{mhos}$  was recorded, and 2,390  $\mu\text{mhos}$  was measured the following day. At Station 3, the conductivity ranged from 720 to 1,170  $\mu\text{mhos}$  and was fairly stable at Stations 4 and 5 with ranges of 640-700 and 640-680  $\mu\text{mhos}$ , respectively.

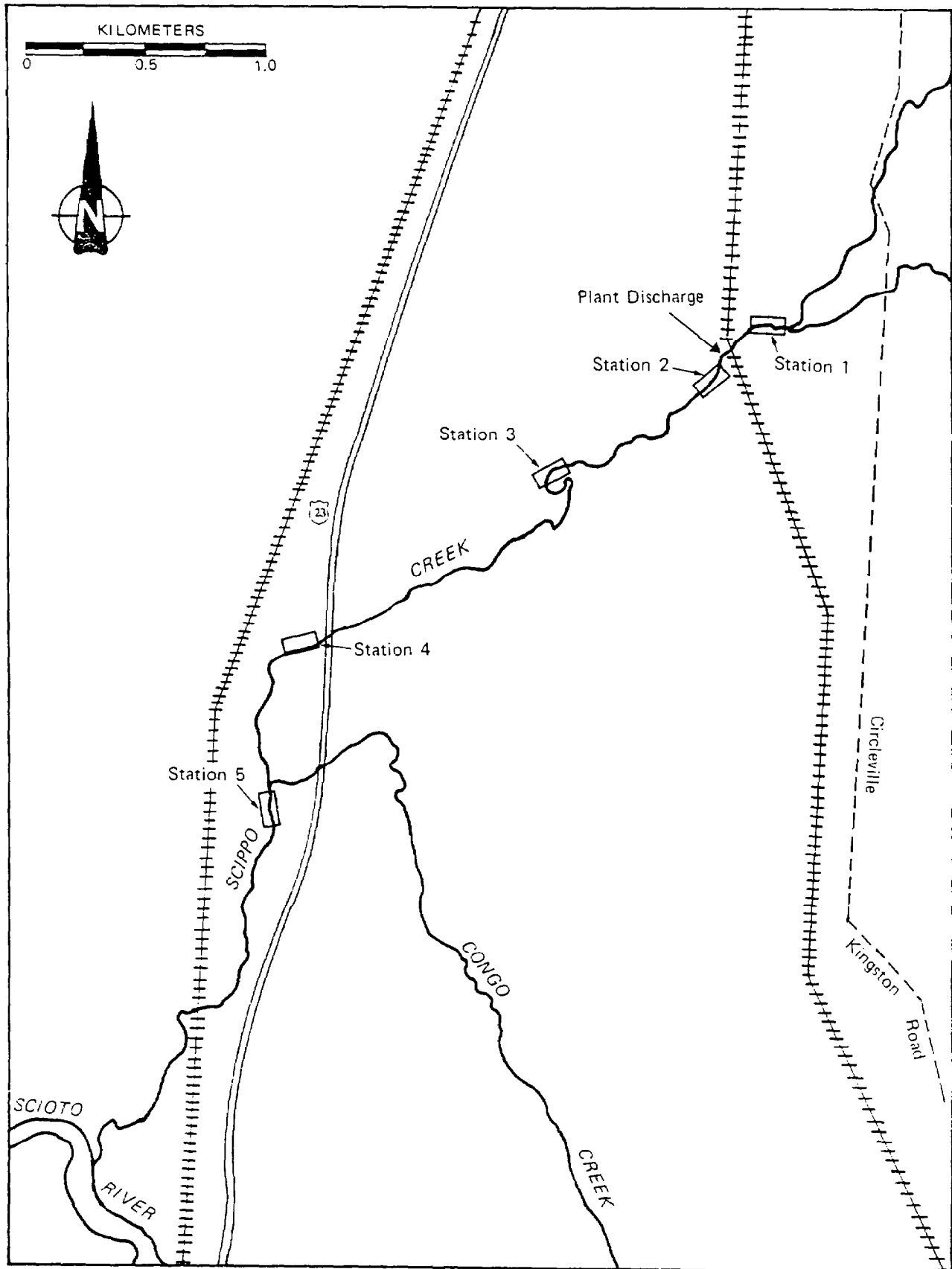


Figure 2-1. Map of study site on Scippo Creek, Circleville, Ohio.

### 3. LABORATORY TOXICITY TESTS

Laboratory toxicity tests using Ceriodaphnia reticulata, fathead minnows, and resident species were conducted to determine the maximum effluent concentrations that would not have chronic toxicity, and to measure the ambient toxicity before and after the effluent is discharged in order to estimate the persistence of the toxicity (Stations 1-4). Several subsidiary objectives were also pursued. Samples of effluent were shipped to Duluth to determine if shipping and delayed testing would produce different results from those of onsite testing. Additional tests at Duluth were done in Lake Superior water to see what effect a different dilution water might have on the results. Descriptions of the toxicity test methods are presented in Appendix A.

Animals from eight different families found in the stream were tested onsite to see if the resident organisms were more or less sensitive than the laboratory animals. If there were differences, the acceptable effluent concentration (AEC) for the resident species could be estimated by dividing the acute/chronic ratio into the LC50 values of the resident species.

Another toxicity test procedure was used with bluntnose minnows (Pimephales notatus). The minnows were caged and set at Stations 1-4. Due to infection, difficulties in capture and handling, and effects of lower water temperatures in the effluent, the test results are regarded as invalid and have not been presented.

#### 3.1 CHEMICAL AND PHYSICAL TEST CONDITIONS

In the onsite tests, the dissolved oxygen (DO) concentration in fathead minnow and resident species tests ranged from 4.6 to 7.7 mg/liter, as measured at the end of each 24-hour period. Initially, DO was very near saturation. In the Ceriodaphnia reticulata tests, DO ranged from 6.6 to 8.0 mg/liter. The pH in all tests was from 7.5 to 8.2. Temperature for the fathead minnows and resident species was from 18.5 to 25 C, and was 25±1 C C) for C. reticulata.

In the shipped-effluent tests conducted in Duluth, Minnesota, DO was 4.8-7.9 and 4.8-7.2 mg/liter for the lake and receiving water tests, respectively. The pH was 7.7 and 8.4 in the lake and receiving water, respectively. Water temperature was maintained at between 24 and 26 C.

Receiving water ranged from 300 to 310 mg/liter hardness (as CaCO<sub>3</sub>) before effluent was added, and up to 400 mg/liter in high effluent concentrations. Corresponding values for alkalinity were 250-260 and 324 mg/liter, respectively.

### 3.2 RESULTS OF ONSITE TOXICITY TESTING

Table 3-1 contains the data from the larval growth test with fathead minnows (Pimephales promelas) exposed to various effluent concentrations diluted with receiving water and tested onsite. The weights are actual values for each replicate and the treatment mean is a weighted average of the replicate means. There was no significant difference in survival or weights at any effluent concentration. Fathead minnow weights were slightly higher at the 25 and 100 percent effluent exposure, perhaps attributable to the additional food in the effluent. The statistical analyses for the weight and survival data are described in Appendix A.

Data from the onsite tests with C. reticulata, using the effluent dilution test and the ambient toxicity test, are shown in Table 3-2. The results in both tests, and especially in those test solutions with no or low effluent concentrations, are heavily influenced by a fungal growth in the test containers that entrapped the animals and prohibited swimming. Although the entrapped animals lived for several days and produced some young, their development was impaired and the test results are not useful in evaluating direct toxicity. When the animals were transferred each day, they were dislodged from the growth by directing a jet of water from the eye dropper and considerable force was needed to free them. They soon became entrapped again because the fungal growth would develop in a few hours although the beakers were thoroughly brushed during washing, and rinsed before reuse. In the ambient test, the fungus problem was worse at Station 1 above the outfall and diminished downstream which

suggests that the fungal growth was not caused by the effluent. Young production from surviving females was not significantly different among stations (Table 3-2). As a result of the fungus, survival was not concentration-dependent and, therefore, any effluent-caused mortality cannot be ascertained.

Table 3-3 contains the resident species data. One of the fish species tested, Pimephales notatus, died of a fungal infection within the first 24 hours. Survival between exposure concentrations was similar for the remaining seven species, and generally varied from 40 to 100 percent for all seven genera. Lowest survival was observed in the middle concentrations. Mortalities could not be attributed to effluent toxicity, only to handling.

### 3.3 RESULTS OF LABORATORY TESTING--DULUTH

The survival and growth data for larval fathead minnow growth tests, conducted at the Environmental Research Laboratory in Duluth, Minnesota, with receiving water and Lake Superior water, are given in Table 3-4. There were no significant differences observed for either the growth or survival data for the receiving water dilution test. Survival was generally lower in the Lake Superior dilution water than in the receiving water test. However, there were no significant differences for survival or growth in any effluent concentrations with Lake Superior water as the diluent.

The data for C. reticulata reproduction and survival in various concentrations of effluent and two diluent waters are presented in Table 3-5. In the receiving water test, none of the exposure groups were significantly lower than the control for either reproduction or survival. In the Lake Superior water test, survival was also not significantly lower between concentrations. All exposure groups at concentrations of 5 percent effluent, and above, had significantly higher ( $P \leq 0.05$ ) young production which may be a result of additional food.



The fungal growth that entrapped the test animals in onsite tests did not occur in the tests done at Duluth, Minnesota, in either dilution water. The reason for this is unknown. The results of these shipped-effluent tests are considered valid for evaluating toxicity of the effluent because control survival was acceptable.

#### 3.4 DISCUSSION AND CONCLUSION

The results of the tests using fathead minnows and C. reticulata indicate no adverse chronic effect even at 100 percent effluent. The resident species tests gave no evidence of acute toxicity nor did the shipped-sample tests with the standard species. Based on these data, no effect of the discharge on Scippo Creek would be expected, even close to the point of discharge. Visual inspection of the discharge area revealed yellow-orange deposits of precipitate which might cause a physical effect, especially on the benthic organisms. For those species able to utilize the increased microorganism population associated with the effluent, a beneficial effect might be expected.

The resident species tests were unsatisfactory because of handling mortality. If such species are to be tested, a suitable acclimation period must be provided. In addition, for those species that live in flowing water, a water current should be provided in the test chamber. However, despite these considerations, it can still be concluded that the 100 percent effluent was not toxic to resident species.

TABLE 3-1 MEAN DRY WEIGHTS AND SURVIVAL FOR FATHEAD MINNOW LARVAE  
 ONSITE EFFLUENT DILUTION TEST IN RECEIVING WATER

<u>Replicate</u>	<u>Larval Weight (mg)</u>					
	<u>Percent Effluent (v/v)</u>					
	<u>100</u>	<u>25</u>	<u>10</u>	<u>5</u>	<u>1</u>	<u>Control</u>
A	0.17	0.16	0.18	0.25	0.29	0.30
B	0.22	0.22	0.16	0.22	0.20	0.21
C	0.37	0.37	0.22	0.22	0.18	0.12
D	0.20	0.20	0.19	0.21	0.18	0.11
Weighted mean(a)	0.238	0.240	0.187	0.225	0.213	0.192
SE(b)	0.034	0.034	0.033	0.033	0.033	0.035

<u>Replicate</u>	<u>Percent Survival</u>					
	<u>Percent Effluent (v/v)</u>					
	<u>100</u>	<u>25</u>	<u>10</u>	<u>5</u>	<u>1</u>	<u>Control</u>
A	100	70	90	100	100	100
B	90	100	100	90	100	90
C	90	90	90	100	100	60
D	90	100	100	100	90	100
Mean	93	90	95	98	98	88

(a) Mean for the group of four replicates, calculated as a weighted mean.

(b) Standard error of the weighted means.

TABLE 3-2 SURVIVAL AND YOUNG PRODUCTION FOR Ceriodaphnia reticulata  
 IN THE ONSITE EFFLUENT DILUTION TEST IN RECEIVING WATER  
 AND FOR AMBIENT TOXICITY TESTS(a)

Receiving Water Test

<u>Percent Effluent (v/v)</u>	<u>Percent Survival</u>	<u>Mean Number of Broods</u>	<u>Mean Number of Young Per Female</u>	<u>95 Percent Confidence Intervals</u>
Control	30	3.0	14.3	9.8-19.0
1	60	2.3	10.8	6.5-14.9
5	70	3.0	13.0	7.2-18.6
10	70	3.0	15.1	11.7-18.7
25	80(b)	3.0	14.4	10.8-18.0
100	40	2.8	13.3	5.2-21.2

Ambient Stream Test

<u>Station</u>	<u>Percent Survival</u>	<u>Mean Number of Broods</u>	<u>Mean Number of Young Per Female</u>	<u>95 Percent Confidence Intervals</u>
1	10	3.0	13.0(c)	--(d)
2	60	3.0	14.8	12.6-17.0
3	50	2.2	12.8	7.6-18.0
4	60	3.2	17.5	14.3-20.6

- (a) The results were affected by fungal growth in the test containers which entrapped the Ceriodaphnia reticulata. Organism development was impaired and control mortality was high so these results are not useful in evaluating direct toxicity.
- (b) Survival was significantly higher than control ( $P \leq 0.05$ ).
- (c) Mean number of young per single surviving adult.
- (d) Confidence intervals were not calculable due to the small sample size of surviving females. See Appendix A for description of statistical analysis.

TABLE 3-3 96-HOUR PERCENT SURVIVAL OF RESIDENT SPECIES EXPOSED TO  
EFFLUENT CONCENTRATIONS

<u>Test Organisms</u> (a)	<u>Percent Effluent (v/v)</u>							
	<u>100</u>		<u>50</u>		<u>25</u>		<u>Control</u>	
	<u>A</u>	<u>B</u>	<u>A</u>	<u>B</u>	<u>A</u>	<u>B</u>	<u>A</u>	<u>B</u>
<u>Etheostoma</u> sp.	100	100	100	100	100	100	100	100
<u>Orconectes</u> sp.	100	100	100	100	100	100	100	100
<u>Hydropsyche</u> sp.	60	60	80	80	80	80	100	80
Heptageniidae	40	80	60	80	40	60	40	80
Philopotamidae	40	60	40	20	20	40	80	40
Ancylidae	100	100	80	100	100	100	100	100
Psephenidae	100	80	100	60	80	100	100	80

(a) One of the species tested, Pimephales notatus, died of a fungal infection within 24 hours of test initiation.

Note: A and B represent replicate test results.

TABLE 3-4 MEAN DRY WEIGHTS AND SURVIVAL FOR FATHEAD MINNOW LARVAE  
EFFLUENT DILUTION TESTS IN TWO DILUTION WATER TYPES AND  
SHIPPED EFFLUENTS

Receiving Water <sup>(a)</sup>	Larval Weight (mg)					
	Percent Effluent (v/v)					
	100	25	10	5	1	Control
A	0.58	0.42	0.47	0.48	0.42	0.42
B	0.40	0.50	0.48	0.29	0.46	0.50
C	0.47	0.52	0.49	0.38	0.39	0.54
D	0.52	0.48	0.35	0.39	0.41	0.51
Weighted mean(b)	0.495	0.482	0.458	0.393	0.421	0.466
SE(c)	0.026	0.029	0.030	0.031	0.029	0.029

Receiving Water <sup>(a)</sup>	Percent Survival					
	Percent Effluent (v/v)					
	100	25	10	5	1	Control
A	90	90	100	100	100	100
B	80	100	100	70	100	100
C	90	100	100	90	90	90
D	90	100	60	90	100	100
Mean	88	98	90	88	98	98

Lake Superior Water	Larval Weight (mg)					
	Percent Effluent (v/v)					
	100	25	10	5	1	Control
A	(d)	0.48	0.28	0.40	--	0.30
B	(d)	0.44	0.45	0.44	0.33	0.45
C	(d)	0.49	0.39	0.43	0.32	0.42
D	(d)	0.44	0.37	0.46	0.25	0.36
Weighted mean(b)	0.495	0.461	0.378	0.434	0.301	0.381
SE(c)	0.026	0.025	0.025	0.027	0.026	0.031

Lake Superior Water	Percent Survival					
	Percent Effluent (v/v)					
	100	25	10	5	1	Control
A	90	90	70	80	50	60
B	80	100	90	100	80	60
C	90	90	90	90	90	60
D	90	100	90	100	80	60
Mean	88	95	85	93	75	60

(a) From Scippo Creek.

(b) Mean for the group of four replicates, calculated as a weighted mean.

(c) Standard error of the weighted means.

(d) The 100 percent effluent test was conducted once. The data are provided under the receiving water test data.

TABLE 3-5 SURVIVAL AND YOUNG PRODUCTION FOR Ceriodaphnia reticulata  
 EFFLUENT DILUTION TESTS IN TWO DILUTION WATER TYPES AND  
 SHIPPED EFFLUENTS

<u>Percent Effluent</u>	<u>Percent Survival</u>	<u>Mean Number of Broods</u>	<u>Mean Number of Young Per Female</u>	<u>95 Percent Confidence Intervals</u>
<u>Lake Superior Water</u>				
Control	90	2.8	14.4	10.9-17.8
1	100	2.9	18.2	15.8-20.6
5	100	2.9	19.6(a)	17.9-21.3
10	80	3.0	22.3(a)	19.4-25.3
25	60	3.0	21.2(a)	21.4-24.5
<u>Scippo Creek Water</u>				
Control	90	3.0	20.6	19.5-21.6
1	100	3.0	20.0	18.0-22.0
5	100	2.9	20.5	18.1-22.9
10	100	3.1	21.0	18.2-23.8
25	90	3.0	21.8	19.9-23.6
100	100	3.0	21.2	20.2-22.2

(a) Mean is significantly greater than the control mean ( $P \leq 0.05$ ).

#### 4. TIME-OF-TRAVEL STUDY AND FLOW MEASUREMENTS

The objective of the hydrology study in Scippo Creek was to ascertain time-of-travel for the effluent, from the discharge to the end of the study area. Two complementary tasks were performed: flow measurements at the biological stations (10 and 13 August 1983), followed by the release of dye and subsequent monitoring of its passage downstream (10 August 1983). The sampling and analytical methods of the hydrological data are presented in Appendix B.

The average cross-sectional velocity from a flow measurement is physically different from a dye study velocity measurement. The flow measurement represents the average velocity through a specific cross-section and is dependent on the cross-sectional area. In contrast, the dye study velocity represents an actual time-of-travel between two points and is more representative of average conditions over a reach.

The results of the dye monitoring at Stations 2 and 3 are shown in Figure 4-1. Following release of the Rhodamine WT dye (1330 hours) in the effluent, the leading edge of the dye reached Station 2 at 1426 hours and the peak of the dye distribution (a concentration of 207 ppb), occurred at 1432 hours. At Station 3, located 1.2 km farther downstream, the leading edge was observed at 1645 hours. The peak dye concentration (37.5 ppb) arrived at 1735 hours. The dye samples collected at Station 4 (1845-2245 hours) showed no dye above background level. The observed time interval for the peak dye concentration to pass from Station 2 to Station 3 yields an average velocity for this section of Scippo Creek of 11 cm/sec.

Table 4-1 presents the flows and average cross-sectional velocity measured at the biological sampling stations. On 10 August, a flow of 0.033 m<sup>3</sup>/sec was measured upstream of the discharge. The average of the three downstream flows was 0.107 m<sup>3</sup>/sec. The flow difference measured between Stations 1 and 2 of 0.100 m<sup>3</sup>/sec (2.3 mgd) is consistent with the nominal reported discharge flow of 2.5 mgd (0.109 m<sup>3</sup>/sec). The average velocity calculated from the dye study of 11 cm/sec is more similar to

the measured velocities at Stations 4 and 5 than at Stations 2 and 3. The higher velocities measured at Stations 2 and 3 (31.1 and 22.9 cm/sec, respectively) appear to be associated with narrower river widths. They are not representative of that portion of the river. Using the velocity of 11 cm/sec resulting from the time-of-travel study, the peak dye distribution would have been expected at Station 4 at 2345 hours. Since sampling stopped at 2245 hours, the leading edge of the dye at Station 4 was probably not sampled.

The time-of-travel study velocity of 11 cm/sec is equivalent to an exposure time of 2.5 hours for each kilometer of movement downstream of the average water parcel from the point of discharge. Water parcels in the leading edge of the distribution would have experienced an exposure time of less than average, whereas parcels in the tail of the distribution would have had longer exposure times. Between Stations 2 and 3, the leading edge of the dye distribution traveled at 14.43 cm/sec, which is equivalent to 1.9 hours of exposure time for each kilometer downstream. Thus, it would be expected that at a 1-km station, the average exposure time is 2.5 hours, with the majority of water parcels having an exposure between 1.9 and 3 hours. At a 2-km station, the average exposure time is 5 hours, with the majority of water parcels having an exposure between 3.8 and 6 hours.

The longitudinal dispersion coefficient for a flow channel (units of area divided by time) is a measure of the rate of the spatial expansion of a group of water parcels with respect to its center of mass. The center of mass moves downstream at the average stream velocity, whereas individual parcels disperse due to turbulence, velocity gradients, and associated phenomena in natural streams. Using Equations B-2 and B-3, the longitudinal dispersion coefficient for Scippo Creek is 17.7 m<sup>2</sup>/min.



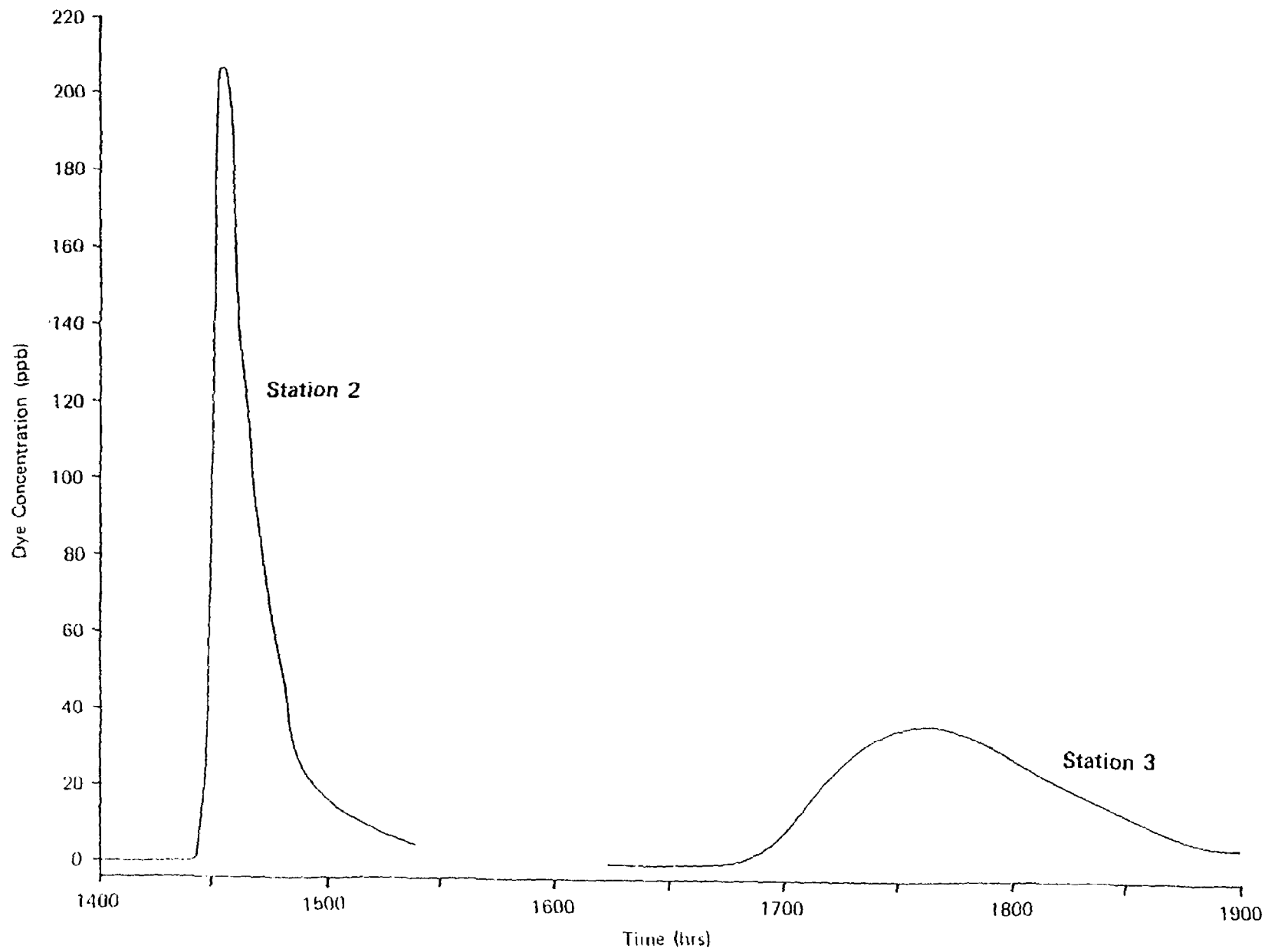


Figure 4-1. Time-of-travel study on Scippo Creek from Station 2 to Station 3 (injection time = 1330).

TABLE 4-1 MEASURED FLOWS ON SCIPPO CREEK

<u>Station</u>	<u>Date</u>	<u>Flow(a)</u> <u>(m<sup>3</sup>/sec)</u>	<u>Average</u> <u>Velocity</u> <u>(cm/sec)</u>
1	10 AUG 1982	0.033	5.5
2	10 AUG 1982	0.133	31.1
3	10 AUG 1982	0.102	22.9
4	10 AUG 1982	0.086	7.3
4	13 AUG 1982	0.080	11.3
5	13 AUG 1982	0.120	9.1

(a) Obtained from measured velocities and the cross-sectional area of the creek at each station.

## 5. PERIPHYTIC COMMUNITY

The study investigated the periphytic community by measuring chlorophyll a and biomass. The relatively short reproduction time and rapid growth of periphytic algae result in quick response to changes in water quality. A change in the periphytic community may be either a reduction of an important habitat or food source for other organisms or the enhancement of nuisance species of algae (that neither support lower trophic levels nor are aesthetically pleasing).

### 5.1 CHLOROPHYLL a AND BIOMASS MEASUREMENTS

The samples for chlorophyll a and biomass analyses were collected on 12 August 1982 from Stations 1 through 4. The samples contained large amounts of sediment and flocculant material, except at Station 1. Due to excessive silt, replicates 2A and 4C had to be discarded.

Chlorophyll a values ranged from 16.4 to 330.0 mg/m<sup>2</sup>. Both of these extreme values were from Station 3 (Table 5-1). This substantial range in values may be caused by changes in natural stream conditions, habitat availability, or sampling conditions. Mean chlorophyll a values ranged from 44.7 to 131.7 mg/m<sup>2</sup> at the four stations. The upstream station (Station 1) and the farthest downstream station sampled for periphyton (Station 4) had similar mean values for chlorophyll a: 38.1 and 39.2 mg/m<sup>2</sup>, respectively. Mean chlorophyll a values at Station 2 averaged 129.7 mg/m<sup>2</sup>. Station 3 averaged 131.7 mg/m<sup>2</sup>. Results of Analysis of Variance (ANOVA) demonstrated that there was no difference among stations when all data were considered, versus a significant difference ( $P \leq 0.05$ ) among stations when Station 3 chlorophyll a values were omitted.

Periphyton biomass was lowest at Station 1 and highest at Station 2. Station 1 had a mean biomass, measured as ash-free dry weight (AFDW), of 19.9 g/m<sup>2</sup>. Station 2 had a mean of 70 g/m<sup>2</sup> AFDW. Mean periphyton biomass at Station 3 decreased by a factor of 1.7 from Station 2, and averaged 40.9 g/m<sup>2</sup> AFDW. Periphyton biomass was lower at Station 4, where the average was 28.2 g/m<sup>2</sup> AFDW. Station 3 had the largest range

between replicates (4.6-107.0 g/m<sup>2</sup>), with the highest and lowest AFDW. Chlorophyll a and AFDWs are measures of algal biomass. Since analyses for these parameters were from the same samples, similar results between replicates would be expected. Results of ANOVA indicated that there was no significant difference in AFDW between stations when all data were considered. However, when Station 3 data were omitted, very significant differences ( $P \leq 0.01$ ) between remaining stations were found.

An autotrophic index (AI) was calculated following that of Weber (1973). The index was based on the ratio of AFDW to chlorophyll a. Results of the autotrophic index were not consistent with the biomass data. The AI values (Table 5-1) show that a relatively large number of either heterotrophic (nonalgal) taxa or nonliving organic matter was present at all stations. These values were highest at Station 4 and lowest at Station 3. These results indicated that the biomass data did not provide a complete estimate of the periphyton community.

## 5.2 EVALUATION OF THE PERIPHYTIC COMMUNITY

Effects on the periphytic community due to the effluent cannot be determined from the data obtained in this study. The increase in chlorophyll a at Stations 2 and 3, below the discharge, suggests enrichment although within-station (replicate) variation was high, especially at Station 3. A similar trend of increasing biomass was noted but was probably due to a combination of periphytic and non-algal constituents. However, no identifications were made to ascertain the composition of the periphytic community.

TABLE 5-1 CHLOROPHYLL a AND BIOMASS MEASUREMENTS OF THE  
 PERIPHYTIC COMMUNITY, SCIPPO CREEK, AUGUST 1982

<u>Station/Replicates</u>		<u>Chlorophyll a</u> <u>(mg/m<sup>2</sup>)</u>	<u>Biomass(a)</u> <u>(g/m<sup>2</sup>)</u>	<u>Autotrophic</u> <u>Index<sup>(b)</sup></u>
1	A	40.6	24.6	
	B	77.0	29.5	
	C	26.7	12.4	
	D	34.6	13.0	
	Mean	44.7	19.9	522
2	A	--(c)	--(c)	
	B	185.0	97.1	
	C	61.2	42.7	
	D	143.0	70.6	
	Mean	129.7	70.1	540
3	A	152.0	41.2	
	B	330.0	107.0	
	C	16.4	4.6	
	D	28.5	10.6	
	Mean	131.7	40.9	311
4	A	37.0	26.4	
	B	28.5	19.1	
	C	--(c)	--(c)	
	D	52.1	39.2	
	Mean	39.2	28.2	719

(a) Ash-free dry weight.

(b) Weber 1973.

(c) Sample rejected because of excessive sediment load.

## 6. BENTHIC MACROINVERTEBRATE COMMUNITY

This survey investigated the benthic community in Scippo Creek. Samples were collected at five stations. Because of the relatively low degree of mobility, the benthic community is considered to be a good indicator of response to adverse conditions at specific locations. The degree of community stability within the study areas can be measured by comparing composition and dominance. An alteration in community structure, standing crop, or species composition of the benthos, beyond the limits of normal fluctuation within the receiving waterbody, would be regarded as an adverse effect. Increased abundance of nuisance insect larvae or other benthic species also would be regarded as adverse effects.

A description of the sampling and analytical methods is presented in Appendix C. Supportive data are summarized in Appendix D.

### 6.1 COMMUNITY COMPOSITION

The benthic community of riffle habitats in Scippo Creek comprised 104 taxa of which only 20 contributed  $\geq 1$  percent to the community population (Table 6-1). Of the 104 taxa collected during August, only two macroinvertebrates, Chironomus spp. and Cricotopus tremulus (both midges), constituted greater than 10 percent of the benthic fauna. Six insect taxa composed greater than 50 percent of the fauna, suggesting that, although the benthic community is diverse in variety of taxa, the structure of the community is dominated by relatively few insect species. Of the 20 taxa and life stages composing one percent or more of the benthos, 12 are in the Chironomidae family. This midge-dominated community is present at all stations.

### 6.2 SPATIAL COMPARISON OF KEY TAXA

Community diversity data based on number of taxa and abundance of individuals within taxa show that diversity was lowest at Station 2 and similar at the other stations (Table 6-2). Conversely, evenness, which compares relative distribution of individuals within taxa among stations,

was also lowest at Station 2. Redundancy, which reflects relative dominance of taxa, was highest at Station 2. These community differences at Station 2 were the consequence of the lowest number of taxa (43 taxa) and the greatest abundance of specimens (17,761 organisms/m<sup>2</sup>).

Figure 6-1 illustrates this pattern of decreasing diversity at Station 2 and increase at Station 3 to a value similar to that noted at Station 1. The number of taxa also decreases from Station 1 to its lowest point at Station 2, increases at Station 3, decreases again at Station 4. It then increases to a maximum of 70 taxa at Station 5. A  $\chi^2$  test was used to test for differences in the number of taxa encountered at each station compared to the expected composition of the reference station. The results of this test indicated that the lower number of taxa encountered at Station 2 was significantly different ( $P \leq 0.05$ ) from the number of taxa at Station 1 (Table D-6). The number of taxa at Stations 3, 4, and 5 was not significantly different from the control. The total number of organisms at each station follows a pattern of low density at Station 1, an increase to peak abundance at Station 2, followed by a steady decrease at the downstream stations to a density at Station 5 similar to that at Station 1.

The community at Station 2 was dominated by two taxa, each of which composed more than 20 percent of the benthos (Table 6-1), whereas no taxon constituted more than 20 percent of the benthos at other stations. The overwhelming dominance of Chironomus spp. and Cricotopus tremulus at Station 2 was not found at any other station. The dominance of these taxa at Station 2 was responsible for the lower diversity index at that station.

Chironomidae and Oligochaeta were present in peak densities at Station 2. They composed 93 percent of the benthos at that station (Figure 6-2). Both groups steadily decreased in abundance downstream. In contrast, Trichoptera and Ephemeroptera decreased from Station 1 to their lowest densities at Station 2 and then increased at downstream stations (Figure 6-3). The chironomid abundance trend was primarily due to three taxa--Chironomus spp., Cricotopus tremulus, and Polypedilum convictum--all

similarly distributed among stations, although at different abundance levels (Figure 6-4). Only at Station 4 were two of these species-- C. tremulus and P. convictum--not found. Results of a one-way Analysis of Variance (ANOVA) and Tukey's Studentized Range Test performed on these three chironomid taxa indicated that the greater densities at Station 2 were highly significantly different ( $P = 0.0001$ ) from densities at other stations (Table D-3). For P. convictum, the densities at Stations 2 and 3 were not significantly different. No significant differences in abundance were found among Stations 1, 4, and 5 for all three species. The high abundance of midges at Station 3 was caused primarily by genera not present in abundance at other stations. Two of these midges--Cricotopus trifascia and Rheotanytarsus spp.--were not found at any other station. Paratanytarsus spp. was uncommon, except at Station 3 (Table 6-1).

Cheumatopsyche spp. and Hydropsyche spp. are the dominant trichopterans in the study area, reflecting the abundance trend of the group among stations (Figure 6-5). Results of the ANOVA and Tukey's test performed on Hydropsyche spp., Cheumatopsyche spp., and early instar Hydropsychidae indicated that lower densities at Station 2 were very significantly different ( $P = 0.0001$ ,  $0.0001$ , and  $0.009$ , respectively) from those at other stations (Table D-4). However, overlap in the station means (natural log-transformed) indicates that distinct station differences in the early life stage of Hydropsyche larvae were not apparent in August 1982. Baetis spp. is the numerically dominant mayfly in the study area and, with the early instars, accounts for the abundance of the mayfly group (Figure 6-6). Although densities of Baetis spp. were very significantly different ( $P = 0.0031$ ) among stations (Table D-5), the Tukey's range test results exhibited overlap of station means. No significant differences in the distribution among stations were found with early instar Baetidae (Table D-5). Differences in abundance between the caddisflies and mayflies are the reversed abundance peaks at Stations 3 and 4. Both groups decreased in numbers at Station 5.



### 6.3 EVALUATION OF THE BENTHIC COMMUNITY

In a survey of the benthic community of Scippo Creek, conducted in July 1971, effects to the community were found to extend approximately 1.6 km downstream from the outfall (Battelle Laboratories 1971). The present trichopteran- and ephemeropteran-dominated community was absent from riffle habitats according to Battelle. Battelle Laboratories (1971) reported an abrupt recovery of the community at a distance located approximately 3.3 km downstream of the outfall. However, no collections were made between the 1.6- and 3.3-km sites to ascertain more specifically where recovery occurred. They also reported that the benthic communities below the recovery zone were more stable than Station 1 because of the greater diversity values in the recovery zone.

Results of this August 1982 study revealed an improvement in the benthic community, as measured by the increase in numbers of individuals and taxa, at all sites compared to Battelle Laboratories (1971) results. The variety of taxa and community abundance in 1982 increased substantially from 1971 indicating the benthos had a more complex community structure. Although mayflies and caddisflies remain major components of the community, by 1982 midges became numerically dominant and the most diverse group. In addition, oligochaetes, crustaceans other than crayfish, and miscellaneous organisms were collected in 1982.

Riffle areas immediately downstream from the outfall (Station 2, 92.3-m distance) were not devoid of biota as reported by Battelle Laboratories (1971). The greatest abundance in this study was found at Station 2. However, the benthic community at Station 2 had low diversity values compared to other stations and had a predominance of a relatively few midge taxa. Although habitat characteristics were similar between stations, the flow regime differed. In addition, a fungal growth appeared all over the substrate, which also may account for population differences. The hydropsychids at Station 2 responded adversely to either water quality conditions or fungal growth (Figure 6-5).

For the most part, the hydropsychids (caddisflies) are collectors and gatherers, whereas the dominant midges at Station 2 are herbivores, thus eliminating competition for food as a factor regulating abundance. Species of Baetis (mayflies) are herbivores and detrital feeders (Merritt and Cummins 1978) and might be considered competitive for food with the midges found at Station 2. However, the chlorophyll a content of the periphyton was high (Chapter 5), indicating that food availability was not influencing the distribution of Baetis. Grazing pressure from the large numbers of minnows, particularly creek chubs (Chapter 7), at Station 2 was also evaluated as a possible cause in the reduction of key benthic taxa. However, the total benthic population was most abundant at this station, suggesting that predation was not a limiting factor to benthic colonization.

There was a substantial increase in numbers of mayflies, Baetis, at Station 3 and below, similar to the increase in abundance of the trichopteran, Cheumatopsyche and Hydropsyche.

Station 3 was affected by the discharge during Battelle Laboratories' study (1971). In contrast, the highest diversity value for the 1982 survey occurred at this station, as well as the peak density of Baetis. The high diversity value and high abundance of benthic organisms depicts a different community at Station 3 than at Station 4, where there was a decrease in the diversity index and a slightly different species composition of the benthic community. However, results of the  $\chi^2$  analysis indicate that there was no difference in number of taxa. The community farthest downstream (Station 5) had a high diversity value and the largest number of taxa (70), but was not significantly different from Stations 1, 3, and 4 in number of taxa (Table D-6).

A localized effect on the benthic community of Scippo Creek was observed at Station 2, but the conditions reported by Battelle Laboratories (1971) have improved. Some of the observed effects may be due to habitat alteration by fungal growth and deposition of iron precipitates. The history of effluent treatment modifications within the 11 years between studies was not reviewed to ascertain the reason for the improvement.

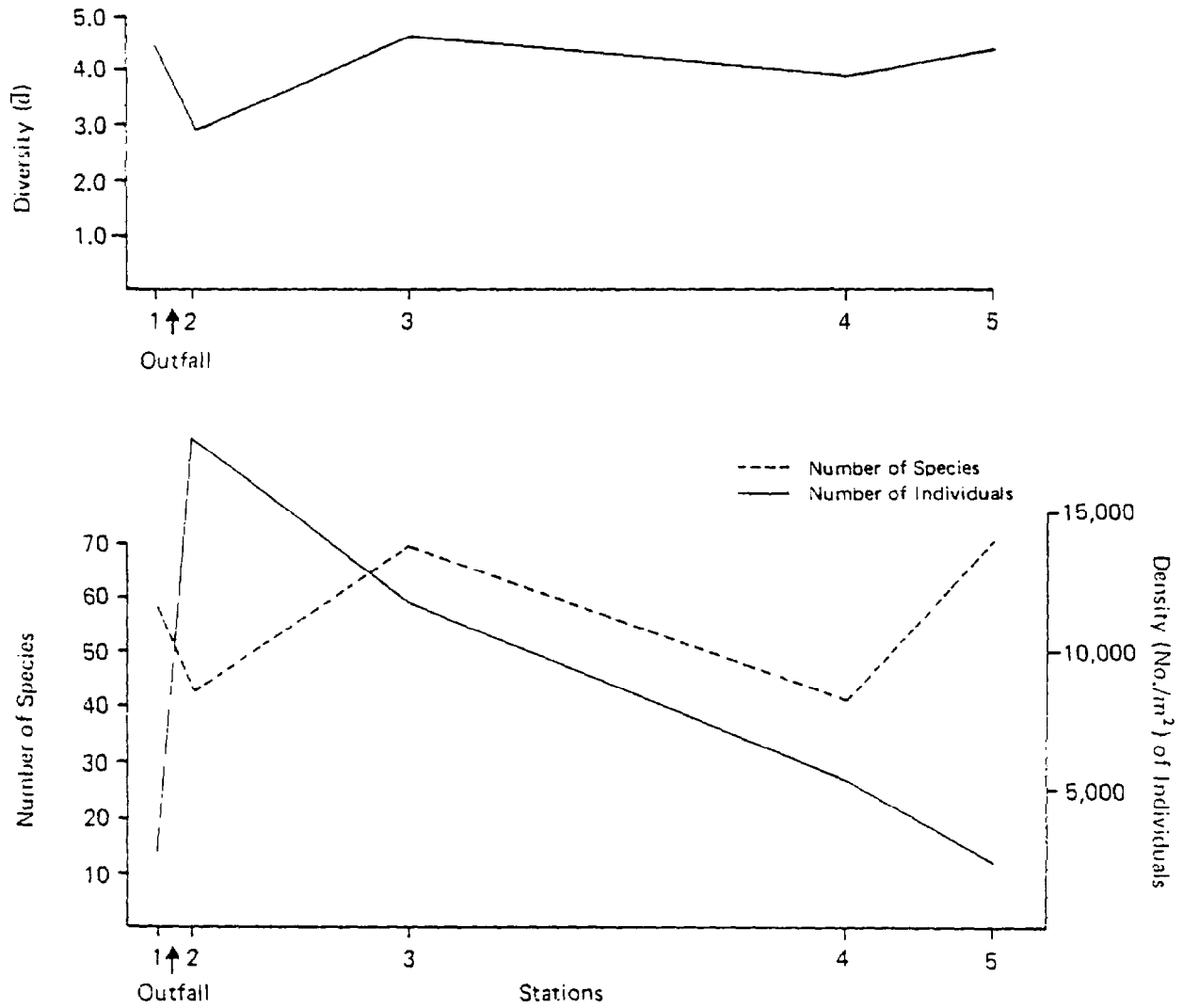


Figure 6-1. Diversity index ( $\bar{d}$ ) and components of the index in Scippo Creek.

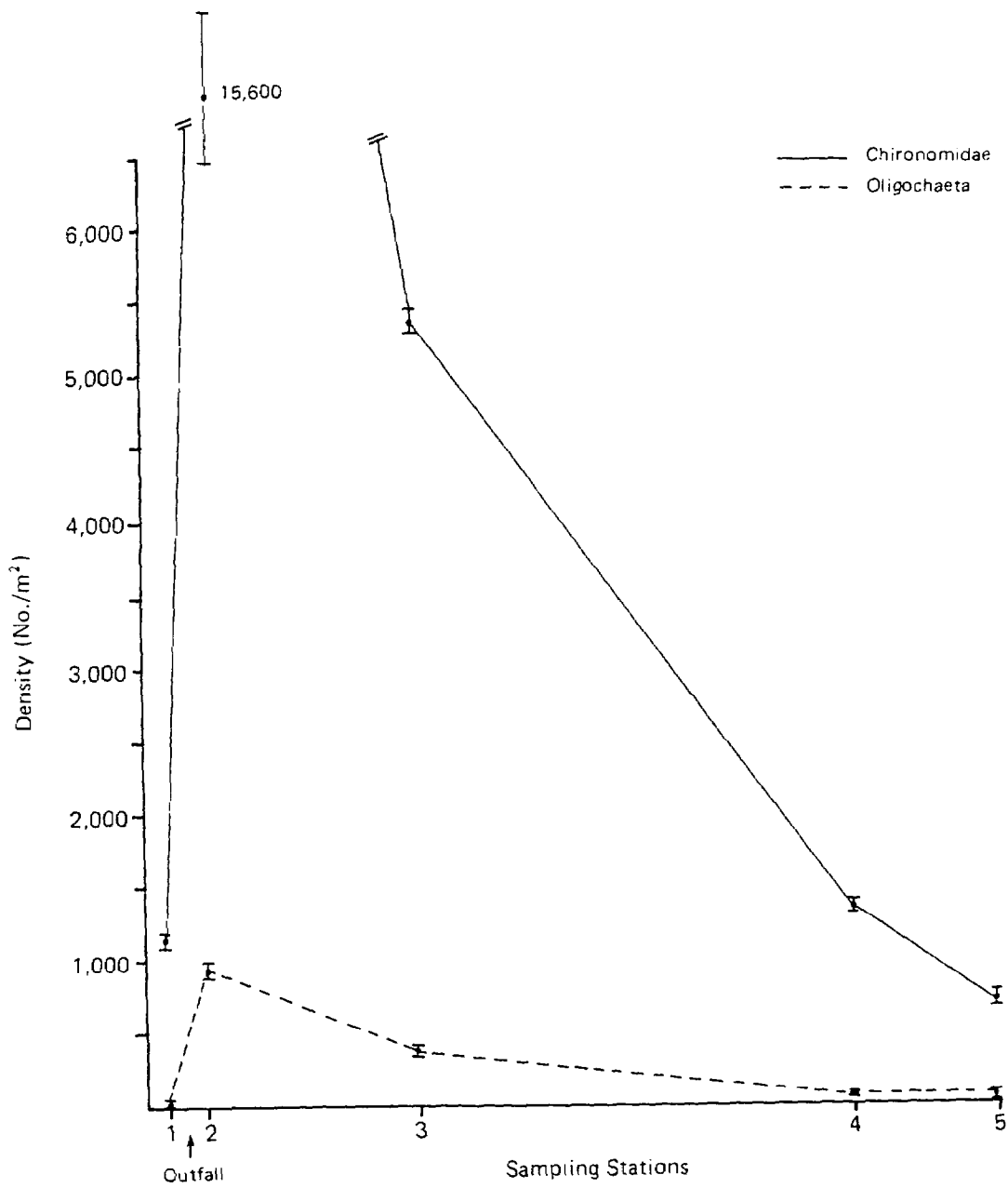


Figure 6-2. Mean density of Chironomidae and Oligochaeta in Scippo Creek. The standard deviation is indicated by brackets.

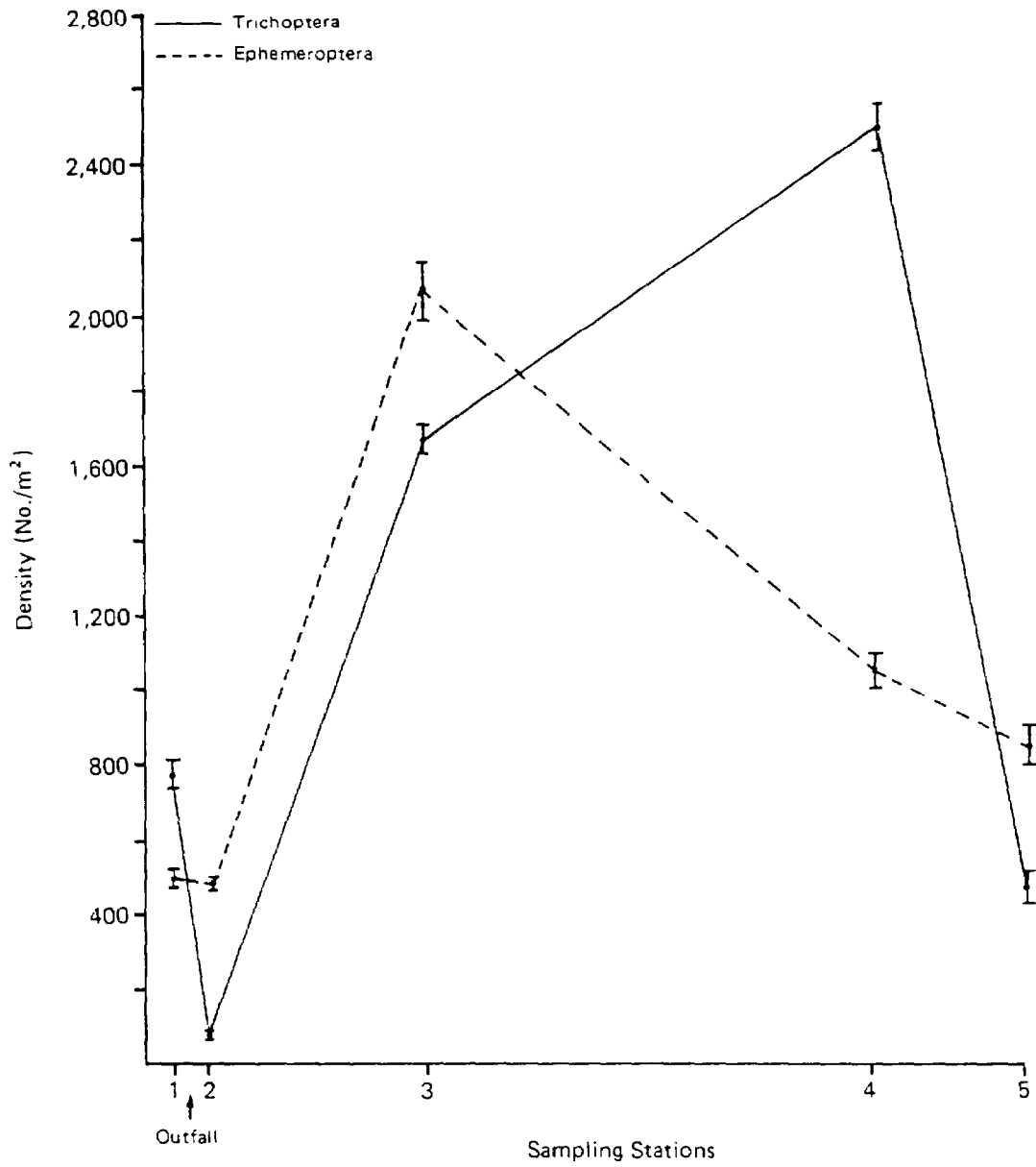


Figure 6-3. Mean density of Trichoptera and Ephemeroptera in Scippo Creek. The standard deviation is indicated by brackets.

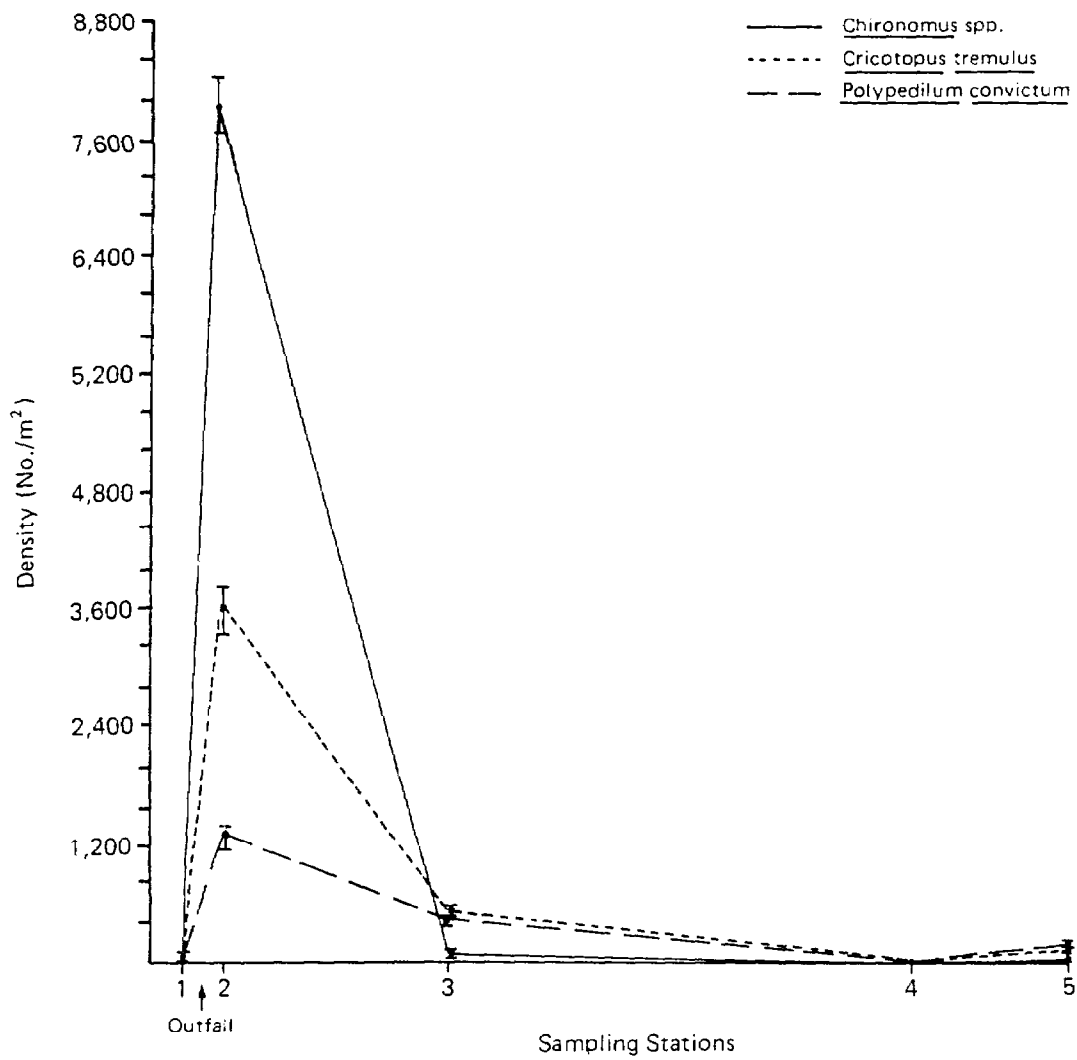


Figure 6-4. Mean density of Chironomids (midges) in Scippo Creek.  
The standard deviation is indicated by brackets.

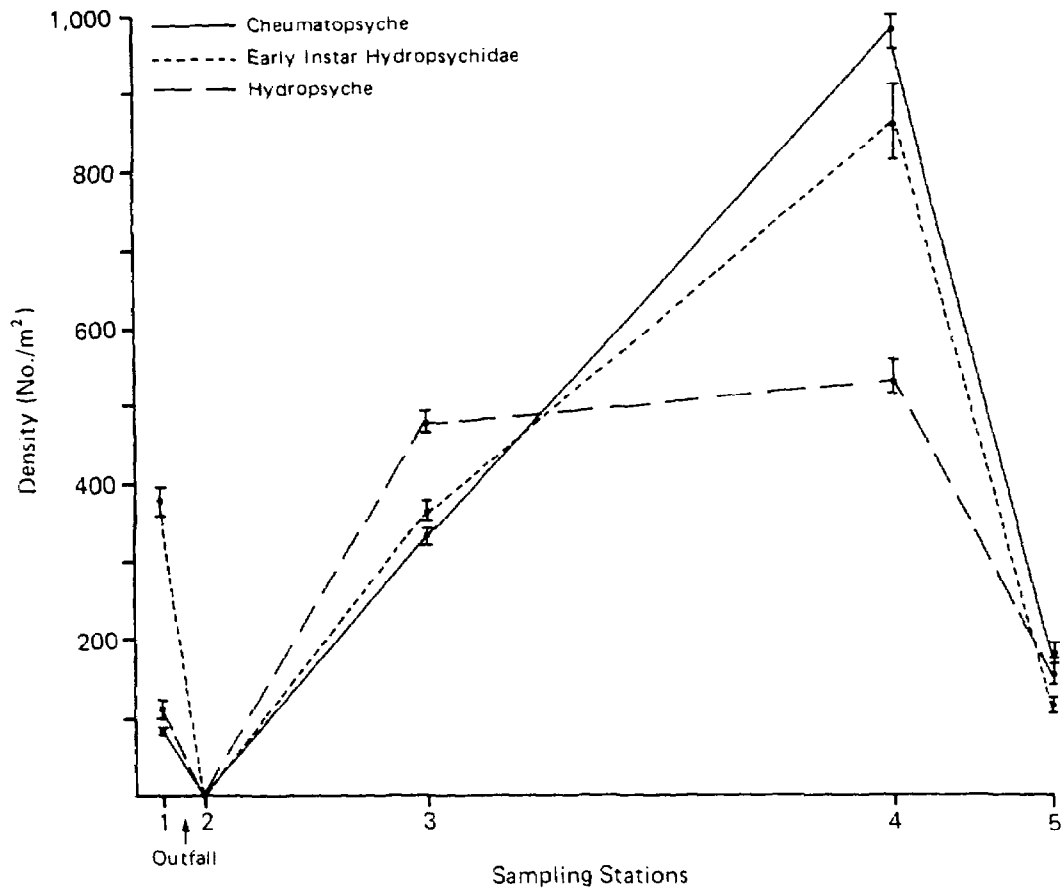
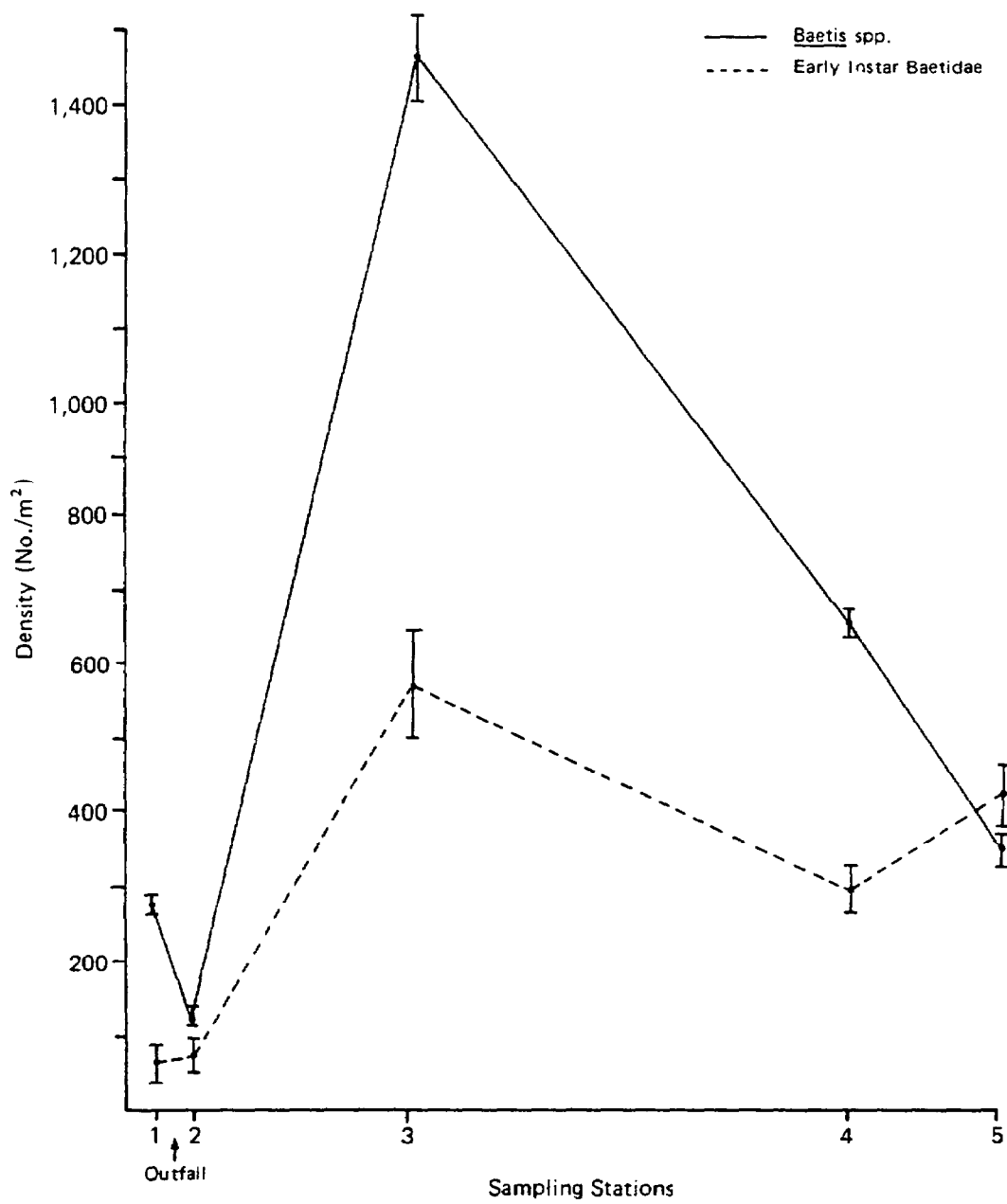


Figure 6-5. Mean density of Trichopterans (caddisflies) in Scippo Creek. The standard deviation is indicated by brackets.



**Figure 6-6. Mean density of Ephemeropterans (mayflies) in Scippo Creek. The standard deviation is indicated by brackets.**



TABLE 6-1 DENSITY AND PERCENT COMPOSITION OF THE MOST ABUNDANT BENTHIC MACROINVERTEBRATE SPECIES AT EACH SAMPLING STATION, SCIPIO CREEK, AUGUST 1982

Species/Life Stage (a)	Station 1		Station 2		Station 3		Station 4		Station 5		Mean	
	Density (no./m <sup>2</sup> )	Percent Composition	Density (no./m <sup>2</sup> )	Percent Composition	Density (no./m <sup>2</sup> )	Percent Composition	Density (no./m <sup>2</sup> )	Percent Composition	Density (no./m <sup>2</sup> )	Percent Composition	Density (no./m <sup>2</sup> )	Percent Composition
<i>Chironomus</i> /L.	4.52	0.17	7,966.50	44.85	18.08	0.15	0.00	0.00	6.78	0.30	1,599.18	19.99
<i>Cricotopus</i> (C.) <i>tremulus</i> /L.	22.60	0.86	3,609.27	20.32	589.86	4.94	0.00	0.00	11.30	0.51	846.60	10.58
<i>Baetis</i> /N.	273.46	10.43	117.57	0.66	1,464.48	12.26	659.92	12.11	354.82	15.89	574.04	7.17
<i>Polypedium</i> (P.) <i>convictum</i> /L.	31.64	1.21	1,301.76	7.33	458.78	3.84	0.00	0.00	20.34	0.91	362.50	4.53
<i>Chironomidae</i> /P.	36.16	1.38	687.04	3.87	806.82	6.76	146.90	2.69	61.02	2.73	347.59	4.34
<i>Hydropsychidae</i> /L.(b)	375.16	14.31	0.00	0.00	352.56	2.95	865.58	15.88	115.26	5.16	341.71	4.27
<i>Cheumatopsyche</i> /L.	74.58	2.84	0.00	0.00	329.96	2.76	974.06	17.87	176.28	7.89	310.98	3.89
<i>Ephemeroptera</i> /N.(c)	65.54	2.50	72.32	0.41	576.30	4.83	293.80	5.39	424.88	19.03	286.57	3.58
<i>Empididae</i> /L.	110.74	4.22	72.32	0.41	980.84	8.21	201.14	3.69	36.16	1.62	280.24	3.50
<i>Rheotanytarsus</i> /L.	13.56	0.52	0.00	0.00	754.84	6.32	565.00	10.36	58.76	2.63	278.43	3.48
<i>Hydropsyche</i> /L.	103.96	3.97	0.00	0.00	474.60	3.97	531.10	9.74	164.98	7.39	254.93	3.19
<i>Simuliidae</i> /L.	0.00	0.00	0.00	0.00	840.72	7.04	11.30	0.21	2.26	0.10	170.86	2.14
<i>Tanytarsus</i> /L.	282.50	10.78	27.12	0.15	474.60	3.97	38.42	0.70	11.30	0.51	166.79	2.08
<i>Thienemannimyia</i> group/L.	192.10	7.33	264.42	1.49	201.14	1.68	92.66	1.70	9.04	0.40	151.87	1.90
<i>Phaenopsectra</i> (P.)/L.	0.00	0.00	655.40	3.69	11.30	0.09	0.00	0.00	0.00	0.00	133.34	1.67
<i>Cricotopus</i> (C.) <i>bicinctus</i> /L.	40.68	1.55	85.88	0.48	449.74	3.77	4.52	0.08	0.00	0.00	116.16	1.45
<i>Microtendipes</i> /L.	74.58	2.84	429.40	2.42	24.86	0.21	0.00	0.00	4.52	0.20	106.67	1.33
<i>Polypedium fallax</i> group/L.	0.00	0.00	368.38	2.07	88.14	0.74	0.00	0.00	0.00	0.00	91.30	1.14
<i>Microtendipes</i> /L.	255.38	9.74	0.00	0.00	117.52	0.98	0.00	0.00	54.24	2.43	85.43	1.07
<i>Caenis</i> /N.	47.46	1.81	293.80	1.65	27.12	0.23	4.52	0.08	29.38	1.32	80.46	1.01
<i>Bothrioneurum vejdovskyanum</i> /L.	0.00	0.00	388.72	2.19	0.00	0.00	2.26	0.04	2.26	0.10	78.65	0.98
<i>Hydroptila</i> /L.	126.56	4.83	18.08	0.10	160.46	1.34	51.98	0.95	4.52	0.20	72.32	0.90
<i>Rheocricotopus</i> /L.	6.78	0.26	15.82	0.09	287.02	2.40	15.82	0.29	11.30	0.51	67.35	0.84
<i>Polypedium</i> (P.) <i>scalgenum</i> /L.	0.00	0.00	151.42	0.85	18.08	0.15	0.00	0.00	153.68	6.88	64.64	0.81
<i>Hydropsychidae</i> /P.	11.30	0.43	9.04	0.05	262.16	2.20	24.86	0.46	6.78	0.30	62.83	0.79
<i>Acarina</i>	15.82	0.60	9.04	0.05	176.28	1.48	79.10	1.45	11.30	0.51	58.31	0.73
<i>Polypedium illinoense</i> /L.	0.00	0.00	0.00	0.00	99.44	0.83	178.54	3.78	4.52	0.20	56.50	0.71
Immature tubificids with capilliform chaetae	2.26	0.09	262.16	1.48	0.00	0.00	6.78	0.12	4.52	0.20	55.14	0.69
Diptera/P.	18.08	0.69	63.28	0.36	162.72	1.36	22.60	0.41	4.52	0.20	54.24	0.68
<i>Nais variabilis</i>	2.26	0.09	18.08	0.10	230.52	1.93	0.00	0.00	2.26	0.10	50.62	0.63
<i>Physella</i>	18.08	0.69	189.84	1.07	15.82	0.13	0.00	0.00	2.26	0.10	45.20	0.56
Tricladida	2.26	0.09	9.04	0.05	108.48	0.91	97.18	1.78	0.00	0.00	43.39	0.54
<i>Cricotopus tritaenia</i> /L.	0.00	0.00	0.00	0.00	198.88	1.67	0.00	0.00	0.00	0.00	39.78	0.50
<i>Rheotanytarsus</i> /P.	0.00	0.00	0.00	0.00	187.58	1.57	0.00	0.00	0.00	0.00	37.52	0.47
<i>Tricorythodes</i> /N.	42.94	1.64	0.00	0.00	2.26	0.02	85.88	1.58	45.20	2.02	35.26	0.44
<i>Paratanytarsus</i> /L.	0.00	0.00	15.82	0.09	133.34	1.12	0.00	0.00	2.26	0.10	30.28	0.38
<i>Gladotanytarsus</i> /L.	51.98	1.98	0.00	0.00	22.60	0.19	6.78	0.12	70.06	3.14	30.28	0.38
<i>Stenelmis</i> /L.	9.04	0.34	90.40	0.51	15.82	0.13	15.82	0.29	13.56	0.61	28.93	0.36
other species	309.62	11.81	569.52	3.21	818.12	6.85	474.60	8.71	352.56	15.79	504.88	6.31
Total	2,621.60		17,761.34		11,941.84		5,451.12		2,232.88		8,001.75	

(a) Life stage notations are: L. = larvae, P. = pupae, N. = nymph. When no lifestage is indicated, organisms were not identified to life stage.

(b) Also referred to as early instar *Hydropsychidae* (Figure 6-5).

(c) Also referred to as early instar *Baetidae* (Figure 6-6).

TABLE 6-2 SHANNON-WIENER DIVERSITY INDICES AND ASSOCIATED EVENNESS AND REDUNDANCY VALUES CALCULATED ON BENTHIC MACROINVERTEBRATE DATA, SCIPPO CREEK(a)

<u>Station</u>	<u>Diversity</u>	<u>Evenness<sup>(b)</sup></u>	<u>Redundancy<sup>(b)</sup></u>	<u>No. of Species</u>	<u>No. of Individuals</u>
1	4.4696	0.7630	0.2397	58	2,622
2	2.9494	0.5435	0.4572	43	17,761
3	4.6697	0.7644	0.2363	69	11,942
4	3.8906	0.7044	0.2971	46	5,451
5	4.3586	0.7111	0.2933	70	2,233

(a) Calculated on a log base 2.

(b) The sum of evenness and redundancy pairs equals one.

## 7. FISH COMMUNITY

The fish community is the highest trophic level potentially affected by discharges to Scippo Creek. This survey investigated the fish community to discern any changes in composition and dominance from previous surveys and to evaluate the response at various stations. A description of the sampling and analytical methods is presented in Appendix C. Species names and common names are provided in Appendix D.

### 7.1 COMMUNITY STRUCTURE

The fish collections yielded 19 species and three taxa of fish that could be identified to only the family or genus level (Table 7-1). Four families were represented in the study area, but a maximum of three occurred at any one station. The stoneroller, creek chub, sand shiner, rainbow darter, and Johnny darter were common species to all five stations. Five additional species were encountered upstream at a collection site for resident species toxicity testing: quillback, pumpkinseed, warmouth, and the black and golden redhorses.

Station 1 yielded 17 species, including seven smallmouth bass, one rock bass, and one small Lepomis sp., the only centrarchids collected (Table 7-1). The catches at Stations 2 through 5 contained mainly cyprinids, with small percentages of darters and suckers. The largest number of specimens was collected at Station 2. The substantial depth and cover in the pool area and greater effectiveness of seining was at least partly responsible for the larger catches. Creek chubs and stonerollers composed over 90 percent of the catch at Station 2. The numbers of specimens and taxa caught at Stations 3, 4, and 5 were all less than those caught at Station 1. The poorest species and family representation occurred at Station 4, where five species of cyprinids and four species of darters were collected.

## 7.2 EVALUATION OF THE FISH COMMUNITY

A fish survey was conducted on Scippo Creek by the State of Ohio Environmental Protection Agency (EPA) in October 1974. The station locations used in that study were similar to Stations 1, 2, and 4 in this study (Figure 2-1). EPA also used 92.3-m sections, but made 40 hauls with a 9.2 x 3.7 m deep seine at each station.

The abundance and number of species in 1982 at Station 1 were similar to those found by the State of Ohio EPA (1974); however, the species composition was somewhat different. No darters were collected in 1974, whereas 25 rainbow and Johnny darters were collected in this study. The catostomids were represented at Station 1 by a small number of fish in 1974, but none was collected in 1982. Also, more centrarchids were collected than in the previous study.

The abundance and diversity of fish found at Station 2 in 1982 far exceeded those collected in 1974. Four species of darter were collected in this study, whereas only one Greenside darter was caught in 1974.

Station 4 had the poorest family and species representation of the five stations studied in 1982, with 235 fish from ten species and two families. In 1974, only 43 fish from eight species and three families were collected. The darters were well represented in both studies, with four species caught in each case.

The number of taxa collected at Stations 2 through 5 were not significantly lower than Station 1, the reference station, as indicated by a  $\chi^2$  test. In contrast, the number of individual fish collected increased 400 percent from Station 1 to Station 2, then decreased to 18-30 percent of the catch at Station 1 for the remaining stations (Table 7-1). These large differences in number of individuals were highly significant, at  $P \leq 0.0001$ . The Centrarchidae family was not found below the outfall; however, the darters were fairly well represented at all five stations. The catostomids, found at the upstream collection site but not at Station 1, were present at three of the downstream stations, although not in

in great numbers. Subtle variation in habitat could account for the differences between stations in composition and abundance within the fish community. In addition, the area sampled for Station 1 was 50 percent larger than for the other stations.

The number of species collected at Scippo Creek varied from 10 to 17, with the highest number collected at Station 1 (Table 7-1). The number of species is so similar among Stations 2 through 5 (results of a  $\chi^2$  test were statistically nonsignificant) that community structure appeared to be unchanged among the downstream stations. The reduction in fish collected downstream of Station 2 does not coincide with expected response to effluent toxicity. Usually toxic effects diminish downstream. This difference in abundance may be attributable to either habitat differences or enrichment of food sources.

TABLE 7-1 ABUNDANCE OF FISH SPECIES, SCIPPO CREEK, AUGUST 1982

Taxa	Station				
	1 (a)	2 (b)	3	4	5
Cyprinidae (small)	23			5	
Cyprinid hybrid	1				
Creek chub	90	1,469	41	50	28
Blacknose dace	2	2	8	1	7
Spotfin shiner	8		2		26
Bluntnose minnow	263	131	53	117	
Stoneroller	161	1,404	21	27	22
Striped shiner	26	2			15
Sand shiner	93	8	5	18	21
Silverjaw minnow	43	56			
Silver shiner	27		22		11
White sucker		16	1		3
Northern hogsucker		1			1
Rock bass JUV	1				
Smallmouth bass YOY	4				
Smallmouth bass JUV	3				
<u>Lepomis</u> sp.	1				
Greenside darter		1	2	1	1
Rainbow darter	21	4	24	5	5
Fantail darter		2	2		
Johnny darter	4	7	2	2	2
Banded darter			1	9	
Total number of taxa(c)	17	13	13	10	12
Total number of individuals(d)	771	3,103	184	235	142

- (a) Totals from 138.5-m sampling section; all other stations were 92.3 m.  
 (b) Aliquot procedures used.  
 (c)  $\chi^2$  test results were: nonsignificant differences among stations. Station 1 was used as the expected value.  
 (d)  $\chi^2$  test results were: highly significant differences among stations ( $P \leq 0.0001$ ). Station 1 was used as the expected value.

Note: JUV = juvenile.  
 YOY = young of the year.

## 8. COMPARISON OF LABORATORY TOXICITY TEST DATA AND RECEIVING WATER BIOLOGICAL IMPACT

One of the objectives of the Complex Effluent Testing Program is to determine which toxicity tests best predict the receiving stream biological impact. Through comparative studies, the reliability of effluent toxicity tests for protecting the aquatic community can be determined. Biological field surveys are useful in assessing pollutant impact, but are of little or no value in determining how much each discharge affects the receiving waterbody. In the development of permit limits, a relationship must be established between the effluent and receiving water impact. Chronic toxicity tests have the potential to measure toxicity in the receiving stream and to predict biological impact. The major problem in establishing this relationship is using laboratory toxicity data from one or two species to predict the community effects for many species.

The development of short, chronic tests has made onsite acquisition of chronic data practical. Toxicity data, expressed as an effect concentration (e.g., the acceptable effluent concentration (AEC)), can provide the quantification needed to set treatment requirements in order to reduce toxic water quality impact. If the AEC is not exceeded in the stream, it can be concluded that there will be no toxic impact from the effluent.

The AEC, as measured in the laboratory on a few species, must compensate for the extrapolation from toxicity data for a few tested species to an AEC for the many species in the community. The sensitivity of any test organism, relative to that of the species in the community, is not known. Therefore, if toxicity is found, there is no method to predict whether many species, or just a few, would be adversely affected at similar concentrations, since the sensitivities of the species in the community also are not known. For example, at a given waste concentration, if the test species has a toxic response and if the species is very sensitive, then only those few species in the community of equal or greater sensitivity would be predicted to be adversely affected. Conversely, if the test

species is tolerant of the effluent, then many more species in the community should be adversely affected at similar concentrations. Thus, the number of species lost due to a toxic effluent cannot be related to the degree of toxicity measured in the toxicity test, unless the position of the tested species within the sensitivity range of the community is known. In this study with only one effluent, the position of the tested species sensitivity would remain the same so long as the communities at each station had the same sensitivity range.

The loss of one or two species from a community is not likely to be considered an adverse effect. Such small changes may be due either to sampling, habitat differences, or the result of the suspected effluent. Further, the toxicity test results only reflect toxicity over the 7-day test period. In contrast, the biological community is a result of adaptation and reaction to many past events that affected the community which include many factors other than the effluent.

The conceptual framework for the data comparison does not rely on test species being a surrogate for any one species or group of species within any community. The fathead minnow data are not intended to predict only the response of the fish community, nor are the C. reticulata data intended to predict only the response of the zooplankton community. However, the conceptual framework does rely on the assumption that the test species' sensitivity is within the range of the sensitivities of species that comprise the biological community.

#### 8.1 PREDICTIONS OF INSTREAM COMMUNITY IMPACTS BASED ON EFFLUENT DILUTION TEST AND AMBIENT TOXICITY TEST RESULTS

In this study, two organisms, C. reticulata and fathead minnows, were used to assess effluent toxicity. Neither test species exhibited acute or chronic toxic responses to the effluent. The AEC for both species was greater than 100 percent effluent concentration. These results predict no adverse effect from the discharge. The biological survey results revealed no conclusive evidence of toxic effect from the single discharge in Scippo Creek. Since species sensitivity is the basis



for the comparison of the toxicity tests and instream community data, it is most desirable to use the total number of species/taxa collected at each station. Other community measures are not regarded as valuable as the number of species/taxa. The community loss index is overly sensitive to habitat effects. Diversity is not useful for cases where the sensitive species of the community are not dominant.

Numbers of organisms and taxa were high below the outfall, but there was a decrease in benthic macroinvertebrate taxa immediately below the outfall at Station 2. However, this decrease was probably due to a habitat loss, caused by the obvious clogging of the interstitial spaces in the substrate which the invertebrates inhabit. If the loss of invertebrate taxa at Station 2 were due to effluent toxicity, one would not expect to see an increase at Station 3, a decrease at Station 4, followed again by an increase at Station 5. A better explanation would be sampling variations or habitat differences.

Fish species also show a marked decrease in number of species at stations downstream of Station 1. This loss may be due to the larger area sampled at Station 1. In addition, the number of fish species is lower at Stations 4 and 5 than at Stations 2 and 3. This pattern is not to be expected if effluent toxicity is the cause.

## 8.2 SUMMARY

The results of the Scippo Creek study demonstrated that the tests are practical to conduct onsite or using shipped samples. The fungal problem was obviously not effluent-caused, but is of concern if such tests are to be routinely used. Any measurement, including simple chemical ones, occasionally fail or show interferences. Toxicity tests are no exception. The fungal problem encountered in the ambient toxicity tests (which was also observed all over the substrate in the benthic macroinvertebrate analysis) was conspicuous and would certainly have caused rejection of test results in routine uses. The important issue is whether this problem occurs frequently. Only continued use will tell.

The effluent toxicity tests predicted no toxic impact on Scippo Creek from the discharge. The field survey found a localized small reduction in the number of taxa approximately 100 m from the outfall at Station 2. This reduction is probably due to a habitat change from the physical clogging of spaces between rocks in the stream bed--not from toxicity.

For regulatory use, the correct prediction of a nontoxic effect is as important as the prediction of a toxic effect. If the localized effect was due to physical alteration of the substrate, corrective action imposed by a regulatory authority would be quite different from the case where the localized effect was due to toxicity. Treatment of the process waste would not aid in the removal of precipitate from the cooling water.

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## A. TOXICITY TEST AND ANALYTICAL METHODS

For the effluent dilution toxicity tests, the dilution water was collected as a grab sample from just upstream (Station 1) of the outfall during late morning of the day it was used. The effluent was collected as a 24-hour composite sample by continuously pumping a small quantity from the discharge flow. Compositing began in late afternoon and the discharge was relatively constant. Therefore, the composite was essentially flow-proportional. Refer to Mount and Norberg (1984) and Norberg and Mount (in press) for a detailed presentation of methods.

Onsite toxicity testing was conducted using Ceriodaphnia reticulata, fathead minnows, and resident species.

Effluent and upstream dilution water samples were air-shipped each day to Duluth for additional laboratory toxicity testing. At ERL-Duluth, the 7-day larval fathead minnow tests and the C. reticulata tests were conducted using shipped receiving water and Lake Superior water as diluents.

In all these tests, new test solutions were made daily from a new 24-hour composite effluent sample and a new grab sample of receiving water. For those tests using Lake Superior dilution water, a new sample was used. The resident species were neither fed nor acclimated before the test was begun. Small rocks collected from Scippo Creek were placed in the benthic invertebrate test chambers as a substrate.

For the fathead minnow and C. reticulata tests, concentrations of 100, 25, 10, 5, and 1 percent effluent were tested. For the resident species tests, only 100, 50, and 25 percent effluent concentrations were tested.

The various concentrations were made by measuring effluent and stream water using graduated cylinders of various sizes, then mixing each concentration in a polyethylene container. All vessels to which effluent or ambient water was in contact were glass, polyethylene, or aluminum. All samples were at or near DO saturation when solutions were made.

Enough test solution was mixed in one batch for the fathead minnow, C. reticulata, and resident species tests.

No chemical measurements for specific chemicals were made. Routine water chemistry, such as DO and pH, was measured in various samples daily. Many of the DO measurements were made just before changing test solutions to determine the minimum values occurring.

Test solutions were changed daily so that in the effluent dilution tests, the fish and C. reticulata were exposed to a new 24-hour composite effluent sample each day which was made up in a new daily grab sample of receiving water. In addition to the effluent dilution tests, four ambient stations were established, one above the outfall and three spaced downstream for measurement of receiving water toxicity. These stations were the same as those used for the biological survey. A daily grab sample was taken at each station and 10 C. reticulata were exposed to each sample for 24 hours, all in separate 30-ml beakers containing 15 ml of water sample.

#### A.1 Ceriodaphnia TESTS

The C. reticulata were from the Duluth culture. They were placed one animal to each of ten 30-ml beakers for each concentration or ambient station sample tested. Fifteen ml of test water were placed in each beaker and a newly born C. reticulata, less than 6 hours old, was used. One drop (0.05 ml) of a food solution containing 250  $\mu$ g yeast was added daily. Each day the adult was moved to a new test solution, a 15-ml volume, with an eye dropper; food was again added. When young were present, they were counted and discarded. Temperatures were maintained at 23-25 C. For the effluent dilution tests, the same concentrations were used as described for the fish. Light was kept very dim to avoid algal growth and to keep conditions comparable to those used for culturing at Duluth. The culture procedures and test method are provided in Mount and Norberg (1984).

## A.2 FATHEAD MINNOW TESTS

For the larval fathead minnow tests, a chamber 30.5 x 15.2 x 10.2-cm deep was made and divided by three glass partitions which resulted in four compartments 12.7 x 7.6 x 10.2-cm deep. The partitions stopped 2.5 cm short of one side of the chamber and a piece of stainless steel screen was glued from one chamber end to the other and across the ends of each compartment. This left a narrow sump 2.5 x 30.5 x 10.2-cm deep along one side of the chamber to which each of the four compartments was connected by its screen end. In this way, the compartments could be filled and drained by adding to or removing water from the sump but retaining the fish in the compartments relatively undisturbed. This design allowed four replicates for each concentration. These are not true replicates in the pure statistical sense because there was a water connection between compartments. However, there was virtually no water movement between compartments as judged by DO measurements. (In some cases there were measurable DO differences between compartments.) When the compartments were filled or drained, some water would mix into other chambers.

Each day 0.1 ml of newly hatched brine shrimp were fed three times to the fish. Fish survival was determined each day. Live brine shrimp were available during the entire daylight period of 16 hours. Light intensity was low.

Each day the compartments were siphoned using a rubber "foot" on a glass tube to remove uneaten brine shrimp. Additional test solution was removed from the sump until about 500 ml remained in the four compartments combined, which equaled about 1 cm of depth or 10-15 percent of the original volume. Then, approximately 2,000 ml of new test solution were added slowly into the sump. The larval fish were able to easily maintain their position against the current. Fish were assigned to compartments one or two at a time in sequential order. They were less than 24 hours post-hatch at the beginning of the test, and were obtained from the Newtown Fish Toxicology Laboratory culture unit.

Because of inadequate temperature control in the mobile lab, the onsite tests with fathead minnows were conducted with temperatures varying from 18 to 25 C. These lower temperatures reduced growth of the minnows from that expected at a constant 25 C. The C. reticulata were kept in a constant temperature cabinet and were not so affected.

At the end of the test, the fish were counted and preserved in 4 percent formalin. Upon return to the Duluth laboratory, they were rinsed in distilled water, oven dried at 98 C for 18 hours, and weighed to the nearest 0.01 mg on an analytical balance. Four lots of 10 fish were preserved at test initiation and later weighed to give an estimate of initial weight. This method is described in more detail in Norberg and Mount (in press).

### A.3 RESIDENT SPECIES TESTS

Resident species were collected from the stream above the outfall and tested in chambers 61.0 x 15.2 x 10.2 cm arranged exactly as the larval fathead minnow test chambers, but each with five compartments 12.7 x 12.2 x 10.2-cm deep. Three liters were used to fill each chamber. Each day, 3 liters were added to chambers after 80 percent of the solution was siphoned out. Five species were tested, one species per compartment, and two such chambers for each concentration provided duplicate test compartments for each species. In addition, two fish and one crayfish species were tested in 30.5-cm diameter battery jars filled with 10 liters of test solution. All but 1 liter was siphoned out each day and 10 liters of new solution were added. Five organisms of each species in each of two replicates were used for the test.

### A.4 FISH CAGING STUDY

The caging study was conducted using commercially available 6-mm (1/4 in.) mesh metal minnow traps whose openings had been plugged with rubber stoppers. The total volume of each cage was approximately 11.5 liters. Three cages were used at each of the four stations and were labeled Prep A, B, and C. Each cage was secured to the bank with a light line.



Fish used in the caging study were collected from locations upstream from the discharge near the Kinston Pike bridge. The bluntnose minnow (Pimephales notatus) was selected for its abundance and relative ease of identification with minimal handling stress. The fish were transported and held in 18.9-liter buckets.

Ten fish were placed in each of three cages. To reduce stress at each handling, care was taken to move the fish quickly but gently in a very fine mesh net. Observations were made daily at approximately the same time and the number of live fish was recorded. Dead fish were removed and discarded.

## A.5 QUANTITATIVE ANALYSES

### A.5.1 Ceriodaphnia reticulata

The statistical analyses of the C. reticulata data were performed using the procedure of Hamilton (1984) as modified by Rogers (personal communication). In this procedure the young production data were analyzed to obtain the mean number of young per female per treatment. Daily means were calculated and these means were summed to derive the 7-day mean young value. By this method, any young produced from females that die during the test are included in the mean daily estimate. Using this procedure, mortalities of the original females affect the estimate minimally, but the mortality of the adult is used along with the young production to determine overall toxicity effects. Confidence intervals are calculated for the mean reproductivity using a standard error estimate calculated by the bootstrap procedure. The bootstrap procedure subsamples the original data set (1,000 times) by means of a computer to obtain a robust estimate of standard error.

A Dunnett's two-tailed t-test is performed with the effluent test data to compare each treatment to the control for significant differences. For the ambient station data, Tukey's Honestly Significant Difference Test is used for the ambient toxicity test data to compare stations.

### A.5.2 Fathead Minnows

The four groups' mean weights are statistically analyzed with the assumption that the four test chamber compartments behave as replicates. The method of analysis assumes the variability in the mean treatment response is proportional to the number of fish per treatment. MINITAB (copyright Pennsylvania State University 1982) was used to estimate a t-statistic for comparing the mean treatment and control data using weighted regressions with weights equal to the number of replicates in the treatments. The t-statistic is then compared to the critical t-statistic for the standard two-tailed Dunnett's test (Steel and Torrie 1960). The survival data are arcsine-transformed prior to the regression analyses to stabilize variances for percent data.

## B. HYDROLOGICAL SAMPLING AND ANALYTICAL METHODS

On 10 August 1982, prior to the dye release, flow measurements were made at Stations 1, 2, and 3 in order to assist in estimating the arrival time of dye at Stations 2, 3, and 4. Additional flow measurements were made on 10 August at Station 4 after the dye release and on 13 August at Stations 4 and 5. The measurements were made with a Teledyne Gurley pygmy flow-meter. At each station, the velocity measurements were made along a transect with the distance between each reading not exceeding 0.3 m and at a depth of 0.6 m of the water column.

At 1330 hours on 10 August 1982, 145.8 g of 20 percent solution of Rhodamine WT dye was released in the effluent prior to its point of discharge into Scippo Creek. At Stations 2 (0.1 km), 3 (1.3 km), and 4 (3.70 km) downstream from the point of discharge, grab samples were collected near midstream at an approximate 0.1-m depth in 200-ml plastic bottles. The sampling interval was initially 15 minutes at each station and decreased as the main dye mass approached. At Station 2, samples were collected from 1345 to 1523 hours. During passage of the main dye mass, samples were collected at 15- and 30-second intervals (1427-1438 hours). At Station 3, samples were collected from 1600 to 1900 hours, with a 2-minute interval used between 1653 and 1815 hours. At Station 4, samples were collected from 1845 to 2245 hours with a 5-minute interval after 2000 hours.

Grab samples were processed in a Turner Designs fluorometer set in the discrete sample mode. The fluorometer had been calibrated prior to the study and calibration was checked each day it was used with standard dye solutions. The fluorometer data were converted to dye concentration, C(ppb), using the relationship:

$$C(\text{ppb}) = SR \exp [0.027(T-20)] \quad (\text{Equation B-1})$$

where

- S = slope from the calibration regression for the appropriate fluorometer scale
- R = fluorometer reading
- T = temperature (C) of the grab sample at the time it was processed

This relationship includes a correction factor for the temperature dependence of fluorescence.

Carter and Okubo (1970) show that the dispersion characteristics of a channel, as measured by the longitudinal dispersion coefficient, may be identified by studying the distribution of dye introduced as an instantaneous point source. The variance ( $\sigma^2$ ) of the longitudinal distribution of the dye concentration, when plotted against time, provides a relationship whose slope is related to the longitudinal dispersion coefficient (K). Mathematically this relationship is

$$K = \frac{1}{2} \frac{d\sigma^2}{dt} \quad (\text{Equation B-2})$$

Carter and Okubo also show a simple method of calculating the variance by fitting a Gaussian distribution to the dye tracer concentration data. The standard deviation, square root of the variance of a Gaussian distribution, is given by

$$= \frac{1}{2\pi} \frac{\text{area under concentration curve}}{\text{peak concentration}} \quad (\text{Equation B-3})$$

The area and peak concentration parameters of the observed dye concentration data at each station may be used with Equation B-3 in order to fit an equivalent Gaussian distribution to the data. The resulting standard deviation of the Gaussian distribution may be used with Equation B-2 to calculate the longitudinal dispersion coefficient.

Multiplying the dispersion coefficient by the travel time (to a point downstream) yields an area value that is proportional to the distance between the leading and trailing edges of the dye distribution multiplied by the mean width of the river. As a result, the dispersion coefficient can be used to characterize the spatial distribution of water particles for a given exposure time.

## C. BIOLOGICAL SAMPLING AND ANALYTICAL METHODS

Water quality measurements consisting of temperature, dissolved oxygen, pH, and conductivity were taken at every station. The instruments used for water quality measurements were a Hydrolab Model 1041, a YSI Model 57 Dissolved Oxygen Meter, and a YSI Model 33 Salinity-Conductivity-Temperature Meter.

### C.1 PERIPHYTON SURVEY

Natural substrates (rocks) were sampled quantitatively using an epilithic algal bar-clamp sampler at each of four stations (Stations 1, 2, 3, and 4). All samples were taken from the lower end of riffle areas and runs located at each station. Four replicate samples were taken at each station for chlorophyll a and biomass measurements. These samples were filtered using 0.45- $\mu$ m filters and stored in ice to await analysis in the laboratory. One sample consisting of a composite of two bar-clamp collections was taken from each station for cursory identification (genus level) and abundance estimates. These samples were preserved in M3 preservative to await analysis. However, identifications were not conducted due to budget constraints.

Biomass measurements of ash-free dry weights (AFDW) and chlorophyll a were analyzed from the filters in the laboratory. A small plug (of equal size) was removed from each filter for chlorophyll a analyses. Chlorophyll a was determined spectrophotometrically after instrument calibration with a chlorophyll a standard (Sigma chemicals) extracted in a 90 percent acetone solution. The plugs of the filters were macerated, and chlorophyll a was extracted with a 90 percent acetone solution. For AFDW, the remaining portions of the filters were dried at 105 C to a constant weight and ashed at 500 C. Distilled water then was added to replace the water of hydration lost from clay and other minerals. Samples were redried at 105 C.

The chlorophyll a and biomass replicate data for each station were analyzed quantitatively by using one-way analysis of variance (ANOVA). In both cases, ANOVAs were conducted on data from all stations and again on data from only Stations 1, 2, and 4. Because of the high variation in the data, Station 3 was omitted from the second analysis.

## C.2 BENTHIC MACROINVERTEBRATE SURVEY

Benthic samples were collected from the pool and riffle habitats at all five stations. Five replicate samples were collected from each of the two habitats at each station. A Hess sampler (881 cm<sup>2</sup>) was used to sample the benthos in the pool habitat. Because of shallow depth (5-10 cm) of the riffle habitat, a Surber sampler (881 cm<sup>2</sup>) was used to collect the benthos from this habitat at each station. The mesh size on the Hess sampler is 363  $\mu$ m, whereas that of the Surber sampler is 500  $\mu$ m. Samples were preserved in 10 percent buffered formalin and returned to the laboratory for analysis. Samples from the pool habitat were not processed, primarily due to budget constraints. Emphasis on the riffle habitat was believed sufficient to detect effects.

The benthic samples contained large amounts of detritus and organisms and were subsampled to expedite organism sorting and identification. Subsampling was done using EA's pneumatic, rotational sample splitter (patent pending). Samples were sorted with the aid of a Wild M-5 dissecting microscope. Organisms were sorted into major taxonomic categories and preserved in 80 percent alcohol to await identification. Organisms were identified to the lowest practical taxon, using appropriate keys and references. Oligochaetes and chironomid larvae were mounted on microslides prior to identification.

A  $\chi^2$  test was used to test differences in the number of benthic taxa among stations. The number of taxa encountered at the upstream station (Station 1) was assumed to be an estimate of the expected number of taxa to be found at all stations of similar habitat.

A one-way ANOVA was used to test for differences in abundance of key taxa among stations. The data were natural log-transformed to ensure a normal distribution and equal variances at all stations. A Tukey's Studentized Range Test was performed where a significant station effect was obtained from the ANOVA. Analyses were conducted using Minitab and SAS PROC GLM.

### C.3 FISH SURVEYS

Fish collections were made at all five stations on Scippo Creek (Figure 2-1). The sections were 92.3 m long, except at Station 1 where a distance of 138.5 m was used. Each section contained pool and riffle habitats, although in varying proportions (Table C-1). The pools were sampled using either a 12 or 13.8 x 3.7 m bag seine with 0.32-cm mesh. A 10.2 x 3.7-m deep straight seine with 0.32-cm mesh was used in the riffles employing the "kick-seine" technique. The number of seine hauls or kick seines varied according to the width and other physical characteristics to ensure complete sampling of the area within the station.

The fish data were quantitatively analyzed using the  $\chi^2$  test on the number of taxa per station and the number of specimens per station. Data for Station 1 were used as the expected values.



TABLE C-1 HABITAT CHARACTERIZATIONS OF THE SAMPLING STATIONS

<u>Station</u>	<u>Percent of Station Area</u>	
	<u>Pool</u>	<u>Riffle</u>
1(a)	60	40
2	80	20
3(b)	75	25
4	70	30
5	55	45

(a) 138.5-m long station.

(b) Pool and riffle separated by 73.8 m of run.

## D. BIOLOGICAL DATA

TABLE D-1 RANKED ABUNDANCE LISTING OF ALL MACROINVERTEBRATES  
COLLECTED, SCIPPO CREEK, AUGUST 1982

<u>Species Name/Life Stage</u>	<u>Number</u>	<u>Percent</u>	<u>Cumulative Percent</u>
CHIRONOMUS/L.	1,599.176	19.985	19.985
C. (CRICOTOPUS) TREMULUS GRP.	846.596	10.580	30.565
BAETIS/N.	574.040	7.174	37.739
POLYPEDILUM (S.S.) CONVICTUM/L.	362.504	4.530	42.270
CHIRONOMIDAE/P.	347.588	4.344	46.614
HYDROPSYCHIDAE/L.	341.712	4.270	50.884
CHEUMATOPSYCHE/L.	310.976	3.886	54.770
EPHEMEROPTERA/N.	286.568	3.581	58.352
EMPIDIDAE/L.	280.240	3.502	61.854
RHEOTANYTARSUS/L.	278.432	3.480	65.334
HYDROPSYCHE/L.	254.928	3.186	68.519
SIMULIIDAE/L.	170.856	2.135	70.655
TANYTARSUS/L.	166.788	2.084	72.739
THIENEMANNIMYIA GRP.	151.872	1.898	74.637
P. (PHAENOPSECTRA)/L.	133.340	1.666	76.303
C. (CRICOTOPUS) BICINCTUS GRP.	116.164	1.452	77.755
DICROTENDIPES/L.	106.672	1.333	79.088
POLYPEDILUM FALLAX GRP./L.	91.304	1.141	80.229
MICROTENDIPES/L.	85.428	1.068	81.297
CAENIS/N.	80.456	1.005	82.302
BOTHRIONEURUM VEJDOVSKYANUM	78.648	0.983	83.285
HYDROPTILA/L.	72.320	0.904	84.189
RHEOCRICOTOPUS/L.	67.348	0.842	85.031
POLYPEDILUM (S.S.) SCALAENUM/L.	64.636	0.808	85.839
HYDROPSYCHIDAE/P.	62.828	0.785	86.624
ACARINA	58.308	0.729	87.352
POLYPEDILIUM ILLINOENSE/L.	56.500	0.706	88.059
IMM TUBIF WITH CAP CHAET	55.144	0.689	88.748
DIPTERA/P.	54.240	0.678	89.426
NAIS VARIABILIS	50.624	0.633	90.058
PHYSELLA	45.200	0.565	90.623
TRICLADIDA	43.392	0.542	91.165
CRICOTOPUS TRIFASCIA/L.	39.776	0.497	91.662
RHEOTANYTARSUS/P.	37.516	0.469	92.131
TRICORYTHODES/N.	35.256	0.441	92.572
PARATANYTARSUS/L.	30.284	0.378	92.950
CLADOTANYTARSUS/L.	30.284	0.378	93.329

Note: N. = Nymph  
L. = Larvae  
P. = Pupae  
U. = Unidentified  
S.S. = sensu strictu

Capitalization of taxa is due to computerized format.

TABLE D-1 (CONT.)

<u>Species Name/Life Stage</u>	<u>Number</u>	<u>Percent</u>	<u>Cumulative Percent</u>
STENELMIS/L.	28.928	0.362	93.690
MICROTENDIPES PEDELLUS/L.	26.668	0.333	94.024
RHYACODRILUS	23.504	0.294	94.317
ENCHYTRAEIDAE	23.052	0.288	94.605
IMM TUBIF W/O CAP CHAET	22.600	0.282	94.888
HYDROPTILIDAE/P.	22.600	0.282	95.170
CERATOPOGONIDAE/L.	20.792	0.260	95.430
EMPIDIDAE/P.	20.792	0.260	95.690
ELMIDAE/L.	18.080	0.226	95.916
GASTROPODA	16.724	0.209	96.125
PROCLADIUS/L.	14.012	0.175	96.300
HYDRA	13.108	0.164	96.464
CRYPTOCHIRONOMUS/L.	13.108	0.164	96.628
SIMULIIDAE/P.	12.656	0.158	96.786
CHIMARRA/L.	12.656	0.158	96.944
ANCYLIDAE	11.300	0.141	97.085
TANYPODINAE/L.	10.848	0.136	97.221
EUKIEFFERIELLA/L.	10.396	0.130	97.351
HEPTAGENIIDAE/N.	9.944	0.124	97.475
TRICHOPTERA/P.	9.944	0.124	97.599
STENONEMA/N.	9.040	0.113	97.712
ELMIDAE/A.	8.588	0.107	97.820
PARAMETRIOCNEMUS/L.	8.588	0.107	97.927
OCHROTRICHIA/L.	8.136	0.102	98.029
CRICOTOPUS/L.	8.136	0.102	98.130
THIENEMANNIELLA/L.	8.136	0.102	98.232
NAIS BRETSCHERI	7.232	0.090	98.322
TRICHOPTERA/L.	6.780	0.085	98.407
PARALAUTERBORNIELLA/L.	6.328	0.079	98.486
C. (CRICOTOPUS) CYLINDRACUS GRP./L.	5.876	0.073	98.560
CHIRONOMINI/L.	5.876	0.073	98.633
NAIS PARDALIS	5.424	0.068	98.701
DUBIRAPHIA/L.	5.424	0.068	98.769
C. (CRICOTOPUS) FESTIVALIS GRP./L.	5.424	0.068	98.836
POLYPEDILIUM OPHIODES/L.	5.424	0.068	98.904
PRISTINA L. LONGISETA	4.068	0.051	98.955
ABLABESMYIA/L.	4.068	0.051	99.006
LABRUNDINIA/L.	4.068	0.051	99.057
COLLEMBOLA U.	3.616	0.045	99.102
LIMNOPHILA/L.	3.616	0.045	99.147
PARAPHAENOCLADIUS/L.	3.616	0.045	99.192
HEXATOMA/L.	3.164	0.040	99.232
CRYPTOTENDIPES/L.	3.164	0.040	99.271
NANOCLADIUS/L.	2.712	0.034	99.305
POLYPEDILUM (P) TRIP./L.	2.712	0.034	99.339
PSEPHENUS/L.	2.260	0.028	99.367
ISOCHAETIDES CURVISETOSUS	2.260	0.028	99.396
TANYTARSUS/P.	2.260	0.028	99.424

TABLE D-1 (CONT.)

<u>Species Name/Life Stage</u>	<u>Number</u>	<u>Percent</u>	<u>Cumulative Percent</u>
ZAVRELIA GRP./L.	2.260	0.028	99.452
MICROPSECTRA/L.	2.260	0.028	99.480
FOSSARIA	1.808	0.023	99.503
CHAETOGASTER DIAPHANUS	1.808	0.023	99.525
PRISTINA LONGISETA LEIDYI	1.808	0.023	99.548
STENACRON/N.	1.808	0.023	99.571
HYDROPHILIDAE/L.	1.808	0.023	99.593
CHIRONOMIDAE/L	1.808	0.023	99.616
LARSIA/L.	1.808	0.023	99.638
CRICOTOPUS (ISOCLADIUS)/L.	1.808	0.023	99.661
CRICOTOPUS SYVLESTRIS GRP./L.	1.808	0.023	99.684
RHABDOCOELA	1.356	0.017	99.701
AULODRILUS FIGUETI	1.356	0.017	99.718
LIMNODRILUS HOFFMEISTERI	1.356	0.017	99.735
PRISTINA LONGISOMA	1.356	0.017	99.751
ISONYCHIA/N.	1.356	0.017	99.768
GERRIDAE/N.	1.356	0.017	99.785
DYTISCIDAE/L.	1.356	0.017	99.802
C. (ISOCLADIUS) LARICOMALIS GRP./L.	1.356	0.017	99.819
GLYPTOTENDIPES/L.	1.356	0.017	99.836
TANYTARSINI/L.	1.356	0.017	99.853
WAPSA MOBILIS	0.904	0.011	99.864
ORCONECTES S. SANBORNI	0.904	0.011	99.876
ANTOCHA/L.	0.904	0.011	99.887
ORTHOCLADIINAE/L.	0.904	0.011	99.898
C. (CHIRONOMUS) THUMMI (RIPARIUS) GRP./L.	0.904	0.011	99.910
PSEUDOCHIRONOMUS/L.	0.904	0.011	99.921
STICTOCHIRONOMUS/L.	0.904	0.011	99.932
PLEURO CERIDAE	0.452	0.006	99.938
AULODRILUS LIMNOBIUS	0.452	0.006	99.944
LIMNODRILUS CERVIX	0.452	0.006	99.949
PRISTINA BREVISETA	0.452	0.006	99.955
ASTACIDAE	0.452	0.006	99.960
HEXAGENIA/N.	0.452	0.006	99.966
HYDROPTILIDAE/L.	0.452	0.006	99.972
BRACHYCENTRIDAE/L.	0.452	0.006	99.977
LEPTOCERIDAE/L.	0.452	0.006	99.983
TANYPUS/L.	0.452	0.006	99.989
PARATENDIPES/L.	0.452	0.006	99.994
POLYPEDILUM SIMULANS/L.	0.452	0.006	100.000

TABLE D-2 NUMBER OF INDIVIDUALS AND PERCENT COMPOSITION FOR BENTHIC  
MACROINVERTEBRATES COLLECTED, SCIPPO CREEK, AUGUST 1982

Species, Lifestage (a)	Station 1									
	Replicate 1		Replicate 2		Replicate 3		Replicate 4		Replicate 5	
	Number	%	Number	%	Number	%	Number	%	Number	%
CHIRONOMUS, L.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	22.60	0.51
C. (CRICOTOPUS) TREMULUS	0.00	0.00	33.90	1.97	0.00	0.00	22.60	2.60	56.50	1.28
BAETIS, N	350.30	11.61	248.60	14.47	485.90	15.75	90.40	10.39	192.10	4.35
POLYPEDILUM (S.S.) CONVI	22.60	0.75	11.30	0.66	124.30	4.03	0.00	0.00	0.00	0.00
CHIRONOMIDAE, P.	33.90	1.12	45.20	2.63	22.60	0.73	0.00	0.00	79.10	1.79
HYDROPSYCHIDAE, L.	666.70	22.10	124.30	7.24	621.50	20.15	113.00	12.99	350.30	7.93
CHEUMATOPSYCHE, L.	113.00	3.75	56.50	3.29	124.30	4.03	22.60	2.60	56.50	1.28
EPHEMEROPTERA, N	56.50	1.87	124.30	7.24	124.30	4.03	11.30	1.30	11.30	0.26
EMPIDIDAE, L.	169.50	5.62	135.60	7.89	135.60	4.40	45.20	5.19	67.80	1.53
RHEOTANYTARSUS L.	22.60	0.75	33.90	1.97	11.30	0.37	0.00	0.00	0.00	0.00
HYDROPSYCHE, L.	305.10	10.11	56.50	3.29	124.30	4.03	11.30	1.30	22.60	0.51
SIMULIIDAE, L.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
TANYTARSUS L.	79.10	2.62	124.30	7.24	101.70	3.30	113.00	12.99	994.40	22.51
THIENEMANNIMYIA, GRP.	282.50	9.36	158.20	9.21	180.80	5.86	67.80	7.79	271.20	6.14
P. (PHAEOPSECTRA) L.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C. (CRICOTOPUS) BICINCTU	45.20	1.50	45.20	2.63	33.90	1.10	22.60	2.60	56.50	1.28
DICROTENDIPES L.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	372.90	8.44
POLYPEDILUM FALLAX GRP.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
MICROTENDIPES, L.	485.90	16.10	113.00	6.58	146.90	4.76	22.60	2.60	508.50	11.51
CAENIS, N	0.00	0.00	0.00	0.00	0.00	0.00	22.60	2.60	214.70	4.86
DOTHRICEURUM VEJDVSKYA	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
HYDROPTILA, L.	0.00	0.00	56.50	3.29	259.90	8.42	90.40	10.39	226.00	5.12
RHECCRICOTOPUS, L.	0.00	0.00	0.00	0.00	33.90	1.10	0.00	0.00	0.00	0.00
POLYPEDILUM (S.S.) SCALA	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
HYDROPSYCHIDAE, P	11.30	0.37	0.00	0.00	11.30	0.37	0.00	0.00	33.90	0.77
ACARINA	56.50	1.87	0.00	0.00	11.30	0.37	11.30	1.30	0.00	0.00
POLYPEDILIUM ILLINOENSE,	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
IMM TUBIF WITH CAP CHAET	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	11.30	0.26
DIPTERA P.	0.00	0.00	11.30	0.66	45.20	1.47	22.60	2.60	11.30	0.26
NAIS VARIABILIS	0.00	0.00	0.00	0.00	11.30	0.37	0.00	0.00	0.00	0.00
PHYSELLA	0.00	0.00	11.30	0.66	79.10	2.56	0.00	0.00	0.00	0.00
TRICLADIDA	0.00	0.00	0.00	0.00	11.30	0.37	0.00	0.00	0.00	0.00
CRICOTOPUS TRIFASCIA, L.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
RHEOTANYTARSUS, P	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
TRICORYTHODES, N.	56.50	1.87	33.90	1.97	22.60	0.73	0.00	0.00	101.70	2.30
PARATANYTARSUS, L.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
CLADOTANYTARSUS L.	0.00	0.00	33.90	1.97	11.30	0.37	0.00	0.00	214.70	4.86
STENELMIS L.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	45.20	1.02
OTHER SPECIES	259.90	8.61	259.90	15.13	350.30	11.36	180.80	20.78	497.20	11.25
TOTAL	3017.10		1717.60		3084.90		870.10		4418.30	

(a) S.S. = Sensu strictu  
L. = Larvae  
P. = Pupae  
N. = Nymph

Note: Abbreviations and capitalization of species names are due to computer format.

TABLE D-2 (CONT.)

Species, Lifestage (a)	Station 2									
	Replicate 1		Replicate 2		Replicate 3		Replicate 4		Replicate 5	
	Number	Comp.	Number	Comp.	Number	Comp.	Number	Comp.	Number	Comp.
CHIRONOMUS, L.	4926.80	26.06	13469.60	47.43	8825.30	54.54	5265.80	48.80	7345.00	50.54
C. (CRICOTOPUS) TREMULUS	6768.70	35.00	7458.00	26.26	1050.90	6.49	1276.90	11.83	1491.60	10.26
BAETIS, N.	226.00	1.20	226.00	0.80	0.00	0.00	90.40	0.64	45.20	0.31
POLYPEDILUM (S.S.) CONVI	418.10	2.21	2881.50	10.15	1322.10	8.17	1197.80	11.10	689.30	4.74
CHIRONOMIDAE, P.	1039.60	5.50	632.80	2.23	768.40	4.75	587.60	5.45	406.80	2.80
HYDROPSYCHIDAE, L.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
CHEMATOPSYCHE, L.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
EPHEMEROPTERA, N.	0.00	0.00	90.40	0.32	0.00	0.00	135.60	1.26	135.60	0.93
EMPIDIDAE, L.	226.00	1.20	45.20	0.16	0.00	0.00	0.00	0.00	90.40	0.62
RHEOTANYTARSUS L.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
HYDROPSYCHE, L.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
SIMULIIDAE, L.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
TANYTARSUS L.	135.60	0.72	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
THIPNEMANNIMYIA, GRP.	565.00	2.99	0.00	0.00	135.60	0.84	395.50	3.66	226.00	1.56
P. (PHAENOPSECTRA) L.	135.60	0.72	237.30	0.84	1977.50	12.22	237.30	2.20	689.30	4.74
C. (CRICOTOPUS) BICINCTU	0.00	0.00	237.30	0.84	0.00	0.00	79.10	0.73	113.00	0.78
DICROTENDIPES L.	983.10	5.20	485.90	1.71	259.90	1.61	79.10	0.73	339.00	2.33
POLYPEDILUM FALLAX GRP.	135.60	0.72	1197.80	4.22	395.50	2.44	0.00	0.00	113.00	0.78
MICROTENDIPES, L.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
CAENIS, N.	361.60	1.91	203.40	0.72	226.00	1.40	406.80	3.77	271.20	1.87
BOTHRICNEURUM VEJDVSKYA	768.40	4.06	406.80	1.43	361.60	2.23	135.60	1.26	271.20	1.87
HYDROPTILA, L.	45.20	0.24	0.00	0.00	45.20	0.28	0.00	0.00	0.00	0.00
RHECCRICOTOPUS, L.	0.00	0.00	0.00	0.00	0.00	0.00	79.10	0.73	0.00	0.00
POLYPEDILUM (S.S.) SCALA	135.60	0.72	237.30	0.84	0.00	0.00	156.20	1.47	226.00	1.56
HYDROPSYCHIDAE, P.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	45.20	0.31
ACARINA	0.00	0.00	0.00	0.00	0.00	0.00	45.20	0.42	0.00	0.00
POLYPEDILIUM ILLINOENSE,	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
IMM TUBIF WITH CAP CHAET	90.40	0.48	90.40	0.32	0.00	0.00	0.00	0.00	1130.00	7.78
DIPTERA P.	0.00	0.00	0.00	0.00	271.20	1.68	45.20	0.42	0.00	0.00
NAIS VARIABILIS	90.40	0.48	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
PHYSELLA	723.20	3.83	0.00	0.00	45.20	0.28	135.60	1.26	45.20	0.31
TRICLADIDA	0.00	0.00	45.20	0.16	0.00	0.00	0.00	0.00	0.00	0.00
CRICOTOPUS TRIFASCIA, L.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
RHEOTANYTARSUS, P.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
TRICORYTHODES, N.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
PARATANYTARSUS, L.	0.00	0.00	0.00	0.00	0.00	0.00	79.10	0.73	0.00	0.00
CLADOTANYTARSUS L.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
STENELMIS L.	0.00	0.00	45.20	0.16	45.20	0.28	50.40	0.84	271.20	1.87
OTHER SPECIES	1130.00	5.98	406.80	1.43	452.00	2.79	271.20	2.51	587.60	4.04
TOTAL	18904.90		28396.90		16181.60		10791.50		14531.80	

TABLE D-2 (CONT.)

Species, Lifestage (a)	Station 3									
	Replicate 1		Replicate 2		Replicate 3		Replicate 4		Replicate 5	
	Number	%	Number	%	Number	%	Number	%	Number	%
CHIRONOMUS, L.	90.40	1.13	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C. (CRICOTOPUS) TREMULUS	308.50	6.37	474.60	3.99	858.80	5.07	836.20	7.39	271.20	2.34
BAETIS, N.	1118.70	14.02	1231.70	10.36	2079.20	12.27	632.80	5.59	2260.00	19.53
POLYPEDILUM (S.S.) CONVI	452.00	5.67	655.40	5.51	632.80	3.73	553.70	4.89	0.00	0.00
CHIRONOMIDAE, P.	632.80	7.93	1118.70	9.41	678.00	4.00	971.80	8.58	632.80	5.47
HYDROPSYCHIDAE, L.	0.00	0.00	0.00	0.00	361.60	2.13	632.80	5.59	768.40	6.64
CHEMATOPSYCHE, L.	214.70	2.69	214.70	1.81	632.80	3.73	271.20	2.40	316.40	2.73
EPH-MEROPTERA, N.	0.00	0.00	485.90	4.09	904.00	5.33	858.80	7.58	632.80	5.47
EMPIDIDAE, L.	723.20	9.07	1175.20	9.89	1943.60	11.47	700.60	6.19	361.60	3.12
RHEOTANYTARSUS L.	598.90	7.51	1412.50	11.88	1039.60	6.13	0.00	0.00	723.20	6.25
HYDROPSYCHE, L.	418.10	5.24	395.50	3.33	723.20	4.27	339.00	2.99	497.20	4.30
SIMULIIDAE, L.	214.70	2.69	813.60	6.84	2305.20	13.60	282.50	2.50	587.60	5.08
TANYTARSUS L.	305.10	3.82	429.40	3.61	452.00	2.67	553.70	4.89	632.80	5.47
THIENEMANNIYA, GRP.	180.80	2.27	124.30	1.05	361.60	2.13	113.00	1.00	226.00	1.95
P. (PHYENOPSECTRA) L.	56.50	0.71	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C. (CRICOTOPUS) BICINCTU	305.10	3.82	689.30	5.80	587.60	3.47	440.70	3.89	226.00	1.95
DICROTENDIPES L.	33.90	0.42	0.00	0.00	90.40	0.53	0.00	0.00	0.00	0.00
POLYPEDILUM FALLAX GRP.	56.50	0.71	90.40	0.76	150.80	1.07	113.00	1.00	0.00	0.00
MICROTENDIPES, L.	146.90	1.84	124.30	1.05	45.20	0.27	271.20	2.40	0.00	0.00
CAENIS, N.	33.90	0.42	33.90	0.29	45.20	0.27	22.60	0.20	0.00	0.00
BOTHRICNEURUM VEJDOSKYA	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
HYDROTILA, L.	33.90	0.42	180.80	1.52	271.20	1.60	226.00	2.00	90.40	0.78
RHECCRICOTOPUS, L.	180.80	2.27	214.70	1.81	226.00	1.33	497.20	4.39	316.40	2.73
POLYPEDILUM (S.S.) SCALA	90.40	1.13	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
HYDROPSYCHIDAE, P.	361.60	4.53	474.60	3.99	406.80	2.40	22.60	0.20	45.20	0.39
ACARINA	124.30	1.56	180.80	1.52	226.00	1.33	124.30	1.10	226.00	1.95
POLYPEDILUM ILLINOENSE,	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	497.20	4.30
IMM TUBIF WITH CAP CHAET	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
DIPTERA P.	0.00	0.00	146.90	1.24	90.40	0.53	305.10	2.69	271.20	2.34
NAIS VARIABILIS	146.90	1.84	214.70	1.81	316.40	1.87	158.20	1.40	316.40	2.73
PHYSELLA	33.90	0.42	0.00	0.00	45.20	0.27	0.00	0.00	0.00	0.00
TRICLADIDA	146.90	1.84	180.80	1.52	90.40	0.53	33.90	0.30	90.40	0.78
CRICOTOPUS TRIFASCIA, L.	180.80	2.27	0.00	0.00	316.40	1.87	497.20	4.39	0.00	0.00
RHEOTANYTARSUS, P.	0.00	0.00	0.00	0.00	0.00	0.00	937.90	8.28	0.00	0.00
TRICORYTHODES, N.	0.00	0.00	0.00	0.00	0.00	0.00	11.30	0.10	0.00	0.00
PARATANYTARSUS, L.	124.30	1.56	90.40	0.76	226.00	1.33	226.00	2.00	0.00	0.00
CLADOTANYTARSUS L.	0.00	0.00	0.00	0.00	0.00	0.00	113.00	1.00	0.00	0.00
STENELMIS L.	33.90	0.42	0.00	0.00	0.00	0.00	0.00	0.00	45.20	0.39
OTHER SPECIES	429.40	5.38	734.50	6.18	813.60	4.80	576.30	5.09	1536.80	13.28
TOTAL	7977.80		11887.60		16950.00		11322.60		11571.20	

TABLE D-2 (CONT.)

Species, Lifestage <sup>(a)</sup>	Station 4									
	Replicate 1		Replicate 2		Replicate 3		Replicate 4		Replicate 5	
	Number	Comp.	Number	Comp.	Number	Comp.	Number	Comp.	Number	Comp.
CHIRONOMUS, L.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C. (CRICOTOPUS) TREMULUS	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
BAETIS, N.	949.20	15.88	576.30	9.57	836.20	12.50	508.50	10.07	429.40	12.22
POLYPEDILUM (S.S.) CONVI	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
CHIRONOMIDAE, P.	135.60	2.27	271.20	4.50	135.60	2.03	124.30	2.46	67.80	1.93
HYDROPSYCHIDAE, L.	1491.60	24.95	870.10	14.45	1423.80	21.28	0.00	0.00	542.40	15.43
CHEMATOPSYCHE, L.	678.00	11.34	1141.30	18.95	1175.20	17.57	1231.70	24.38	644.10	18.33
EPHEMEROPTERA, N.	160.80	3.02	56.50	0.94	429.40	6.42	598.90	11.86	203.40	5.79
EMPIDIDAE, L.	203.40	3.40	305.10	5.07	158.20	2.36	214.70	4.25	124.30	3.54
RHEOTANYTARSUS L.	565.00	9.45	666.70	11.07	632.80	9.46	666.70	13.20	293.80	8.36
HYDROPSYCHE, L.	678.00	11.34	305.10	5.07	791.00	11.82	508.50	10.07	372.90	10.61
SIMULIIDAE, L.	22.60	0.38	0.00	0.00	0.00	0.00	33.90	0.67	0.00	0.00
TANYTARSUS L.	67.80	1.13	124.30	2.06	0.00	0.00	0.00	0.00	0.00	0.00
THINNEHMANNIYIA, GRP.	45.20	0.76	305.10	5.07	90.40	1.35	0.00	0.00	22.60	0.64
P. (PHENOPSECTRA) L.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C. (CRICOTOPUS) BICINCTU	0.00	0.00	0.00	0.00	22.60	0.34	0.00	0.00	0.00	0.00
DICROTENDIPES L.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
POLYPEDILUM FALLAX GRP.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
MICROTENDIPES, L.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
CAENIS, N.	22.60	0.38	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
BOTHRIDNEURUM VEJDOVSKYA	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	11.30	0.32
HYDROPTILA, L.	90.40	1.51	90.40	1.50	22.60	0.34	33.90	0.67	22.60	0.64
RHEOCRICOTOPUS, L.	22.60	0.38	0.00	0.00	22.60	0.34	0.00	0.00	33.90	0.96
POLYPEDILUM (S.S.) SCALA	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
HYDROPSYCHIDAE, P.	56.50	0.95	0.00	0.00	22.60	0.34	33.90	0.67	11.30	0.32
ACARINA	67.80	1.13	146.90	2.44	67.80	1.01	33.90	0.67	79.10	2.25
POLYPEDILUM ILLINOENSE,	158.20	2.65	214.70	3.56	158.20	2.36	271.20	5.37	90.40	2.57
IMM TUBIF WITH CAP CHAET	0.00	0.00	0.00	0.00	0.00	0.00	33.90	0.67	0.00	0.00
DIPTERA P.	22.60	0.38	33.90	0.56	22.60	0.34	33.90	0.67	0.00	0.00
NAIS VARIABILIS	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
PHYSELLA	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
TRICLADIDA	90.40	1.51	124.30	2.06	158.20	2.36	33.90	0.67	79.10	2.25
CRICOTOPUS TRIFASCIA, L.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
RHEOTANYTARSUS, P.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
TRICORYTHODES, N.	90.40	1.51	124.30	2.06	45.20	0.68	146.90	2.91	22.60	0.64
PARATANYTARSUS, L.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
CLADOTANYTARSUS L.	0.00	0.00	33.90	0.56	0.00	0.00	0.00	0.00	0.00	0.00
STENELMIS L.	22.60	0.38	0.00	0.00	45.20	0.68	0.00	0.00	11.30	0.32
OTHER SPECIES	316.40	5.29	632.80	10.51	429.40	6.42	542.40	10.74	452.00	12.86
TOTAL	5977.70		6022.90		6689.60		5051.10		3514.30	



TABLE D-2 (CONT.)

Species, Lifesage <sup>(a)</sup>	Station 5									
	Replicate 1		Replicate 2		Replicate 3		Replicate 4		Replicate 5	
	Number	Comp.	Number	Comp.	Number	Comp.	Number	Comp.	Number	Comp.
CHIRONOMUS, L.	0.00	0.00	33.90	1.67	0.00	0.00	0.00	0.00	0.00	0.00
C. (CRICOTOPUS) TREMULUS	0.00	0.00	0.00	0.00	0.00	0.00	11.30	0.34	45.20	1.72
BAETIS, N	745.80	33.00	0.00	0.00	361.60	40.51	429.40	12.79	237.30	9.05
POLYPEDILUM (S.S.) CONVI	67.80	3.00	0.00	0.00	0.00	0.00	0.00	0.00	33.90	1.29
CHIRONOMIDAE, P	90.40	4.00	33.90	1.67	22.60	2.53	79.10	2.36	79.10	3.02
HYDROPSYCHIDAE, L.	67.80	3.00	0.00	0.00	79.10	8.86	237.30	7.07	192.10	7.33
CHEUMATOPSYCHE, L.	90.40	4.00	0.00	0.00	67.80	7.59	519.80	15.49	203.40	7.76
EPHEMEROPTERA, N.	237.30	10.50	33.90	1.67	79.10	8.86	949.20	28.28	824.90	31.47
EMPIDIDAE, L.	45.20	2.00	0.00	0.00	22.60	2.53	45.20	1.35	67.80	2.59
RHEOTANYTARSUS L.	135.60	6.00	0.00	0.00	0.00	0.00	67.80	2.02	90.40	3.45
HYDROPSYCHE, L.	90.40	4.00	0.00	0.00	67.80	7.59	372.90	11.11	293.80	11.21
SIMULIIDAE, L.	0.00	0.00	0.00	0.00	0.00	0.00	11.30	0.34	0.00	0.00
TANYTARSUS L.	11.30	0.50	11.30	0.56	11.30	1.27	0.00	0.00	22.60	0.86
THIPNEMANNIMYIA, GRP.	33.90	1.50	0.00	0.00	11.30	1.27	0.00	0.00	0.00	0.00
P (PHAENOPSECTRA) L.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C. (CRICOTOPUS) BICINCTU	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
DICROTENDIPES L.	0.00	0.00	22.60	1.11	0.00	0.00	0.00	0.00	0.00	0.00
POLYPEDILUM FALLAX GRP.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
MICROTENDIPES, L.	56.50	2.50	0.00	0.00	45.20	5.06	0.00	0.00	169.50	6.47
CAENIS, N	11.30	0.50	124.30	6.11	0.00	0.00	0.00	0.00	11.30	0.43
DOTHRIDNEURUM VEJDOSKYA	0.00	0.00	0.00	0.00	11.30	1.27	0.00	0.00	0.00	0.00
HYDROPTILA, L.	0.00	0.00	11.30	0.56	0.00	0.00	11.30	0.34	0.00	0.00
RHEOCRICOTOPUS, L.	22.60	1.00	0.00	0.00	11.30	1.27	22.60	0.67	0.00	0.00
POLYPEDILUM (S.S.) SCALA	293.80	13.00	429.40	21.11	11.30	1.27	0.00	0.00	33.90	1.29
HYDROPSYCHIDAE, P.	11.30	0.50	0.00	0.00	0.00	0.00	0.00	0.00	22.60	0.86
ACARINA	33.90	1.50	0.00	0.00	0.00	0.00	0.00	0.00	22.60	0.86
POLYPEDILUM ILLINOENSE,	0.00	0.00	0.00	0.00	0.00	0.00	22.60	0.67	0.00	0.00
IMM TUBIF WITH CAP CHAET	11.30	0.50	11.30	0.56	0.00	0.00	0.00	0.00	0.00	0.00
DIPTERA P	0.00	0.00	11.30	0.56	0.00	0.00	11.30	0.34	0.00	0.00
MAIS VARIABILIS	0.00	0.00	11.30	0.56	0.00	0.00	0.00	0.00	0.00	0.00
PHYSELLA	11.30	0.50	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
TRICLADIDA	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
CRICOTOPUS TRIFASCIA, L.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
RHEOTANYTARSUS, P.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
TRICORYTHODES, N.	45.20	2.00	0.00	0.00	22.60	2.53	90.40	2.69	67.80	2.59
PARATANYTARSUS, L.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	11.30	0.43
CLADOTANYTARSUS L.	22.60	1.00	305.10	15.00	0.00	0.00	11.30	0.34	11.30	0.43
STENELMIS L.	0.00	0.00	0.00	0.00	11.30	1.27	33.90	1.01	22.60	0.86
OTHER SPECIES	124.30	5.50	994.40	48.89	56.50	6.33	429.40	12.79	158.20	6.03
TOTAL	2260.00		2034.00		892.70		3356.10		2621.60	

TABLE D-2 (CONT.)

Species, Lifestage (a)	Total	
	Number	Comp.
CHIRONOMUS, L.	1599.18	19.99
C. (CRICOTOPUS) TREMULUS	846.60	10.58
BAETIS, N	574.04	7.17
POLYPEDILUM (S.S.) CONVI	362.50	4.53
CHIRONOMIDAE, P.	347.59	4.34
HYDROPSYCHIDAE, L.	341.71	4.27
CHEUMATOPSYCHE, L.	310.98	3.89
EPHEMEROPTERA, N.	286.57	3.58
EMPIDIDAE, L.	280.24	3.50
RHEOTANYTARSUS L.	278.43	3.48
HYDROPSYCHE, L.	254.93	3.19
SIMULIIDAE, L.	170.86	2.14
TANYTARSUS L.	166.79	2.08
THIENEMANNIMYIA, GRP.	151.87	1.90
P. (PHAENOPSECTRA) L.	133.34	1.67
C. (CRICOTOPUS) BICINCTU	116.16	1.45
DICROTENDIPES L.	106.67	1.33
POLYPEDILUM FALLAX GRP.	91.30	1.14
MICROTENDIPES, L.	85.43	1.07
CAENIS, N.	80.46	1.01
DOTHRIONEURUM VEJDOVSKYA	78.65	0.98
HYDROPTILA, L.	72.32	0.90
RHECCRICOTOPUS, L.	67.35	0.84
POLYPEDILUM (S.S.) SCALA	64.64	0.81
HYDROPSYCHIDAE, P	62.83	0.79
ACARINA	58.31	0.73
POLYPEDILIUM ILLINOENSE,	56.50	0.71
IMM TUBIF WITH CAP CHAET	55.14	0.69
DIPTERA P	54.24	0.68
NAIS VARIABILIS	50.62	0.63
PHYSELLA	45.20	0.56
TRICLADIDA	43.39	0.54
CRICOTOPUS TRIFASCIA, L.	39.78	0.50
RHEOTANYTARSUS, P.	37.52	0.47
TRICORYTHODES, N.	35.26	0.44
PARATANYTARSUS, L.	30.28	0.38
CLADOTANYTARSUS L.	30.28	0.38
STENELMIS L.	28.93	0.36
OTHER SPECIES	504.88	6.31
TOTAL	8001.75	

TABLE D-3 ANALYSIS OF VARIANCE AND TUKEY'S STUDENTIZED RANGE TEST RESULTS FOR SPECIES OF CHIRONOMIDAE, SCIPPO CREEK, AUGUST 1982

Chironomus spp.

Dependent Variable: ln count

<u>Source</u>	<u>Df</u>	<u>Sum of Squares</u>	<u>Mean Square</u>	<u>F Value</u>	<u>PR &gt; F</u>
Model	4	157.05	39.26	111.63	0.0001
Error	20	7.03	0.35		
Corrected total	24	164.08			

Tukey's Studentized Range Test

Station (mean ln count)	2	3	5	1	4
	(6.49)	(0.44)	(0.28)	(0.22)	(0)

Chironomus tremulus

Dependent Variable: ln count

<u>Source</u>	<u>Df</u>	<u>Sum of Squares</u>	<u>Mean Square</u>	<u>F Value</u>	<u>PR &gt; F</u>
Model	4	114.16	28.54	62.22	0.0001
Error	20	9.17	0.46		
Corrected total	24	123.34			

Tukey's Studentized Range Test

Station (mean ln count)	2	3	1	5	4
	(5.41)	(3.89)	(0.86)	(0.46)	(0)

Polypedilum convictum

Dependent Variable: ln count

<u>Source</u>	<u>Df</u>	<u>Sum of Squares</u>	<u>Mean Square</u>	<u>F Value</u>	<u>PR &gt; F</u>
Model	4	73.97	18.49	16.63	0.0001
Error	20	22.24	1.11		
Corrected total	24	96.20			

Tukey's Studentized Range Test

Station (mean ln count)	2	3	1	5	4
	(4.55)	(3.15)	(0.86)	(0.67)	(0)

TABLE D-4 ANALYSIS OF VARIANCE AND TUKEY'S STUDENTIZED RANGE TEST RESULTS FOR SPECIES OF HYDROPSYCHIDAE, SCIPPO CREEK, AUGUST 1982

Hydropsyche spp.

Dependent Variable: ln count

<u>Source</u>	<u>Df</u>	<u>Sum of Squares</u>	<u>Mean Square</u>	<u>F Value</u>	<u>PR &gt; F</u>
Model	4	48.99	12.25	18.43	0.0001
Error	20	13.29	0.66		
Corrected total	24	62.28			

Tukey's Studentized Range Test

Station (mean ln count)	4 (3.81)	3 (3.73)	5 (2.19)	1 (1.88)	2 (0)

Cheumatopsyche spp.

Dependent Variable: ln count

<u>Source</u>	<u>Df</u>	<u>Sum of Squares</u>	<u>Mean Square</u>	<u>F Value</u>	<u>PR &gt; F</u>
Model	4	55.07	13.77	26.02	0.0001
Error	20	10.58	0.53		
Corrected total	24	65.65			

Tukey's Studentized Range Test

Station (mean ln count)	4 (4.43)	3 (3.32)	5 (2.19)	1 (1.91)	2 (0)

Early Instar Hydropsychidae

Dependent Variable: ln count

<u>Source</u>	<u>Df</u>	<u>Sum of Squares</u>	<u>Mean Square</u>	<u>F Value</u>	<u>PR &gt; F</u>
Model	4	40.19	10.05	4.54	0.0090
Error	20	44.26	2.21		
Corrected total	24	84.45			

Tukey's Studentized Range Test

Station (mean ln count)	4 (3.60)	1 (3.29)	3 (2.35)	5 (2.00)	2 (0)

TABLE D-5 ANALYSIS OF VARIANCE AND TUKEY'S STUDENTIZED RANGE TEST RESULTS PERFORMED ON SPECIES OF BAETIDAE, SCIPPO CREEK, AUGUST 1982

Baetis sp.

Dependent Variable: ln count

<u>Source</u>	<u>Df</u>	<u>Sum of Squares</u>	<u>Mean Square</u>	<u>F Value</u>	<u>PR &gt; F</u>
Model	4	23.30	5.83	5.71	0.0031
Error	20	20.40	1.02		
Corrected total	24	43.71			

Tukey's Studentized Range Test

Station (mean ln count)	3 (4.77)	4 (4.04)	1 (3.09)	5 (2.89)	2 (1.98)
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Early Instar Baetidae

Dependent Variable: ln count

<u>Source</u>	<u>Df</u>	<u>Sum of Squares</u>	<u>Mean Square</u>	<u>F Value</u>	<u>PR &gt; F</u>
Model	4	15.50	3.87	2.24	0.1009
Error	20	34.58	1.73		
Corrected total	24	50.08			

Tukey's Studentized Range Test not performed since ANOVA results were nonsignificant.

TABLE D-6 RESULTS OF A  $\chi^2$  TEST PERFORMED ON THE NUMBER OF BENTHIC  
MACROINVERTEBRATE TAXA COLLECTED AT EACH STATION

	Station				
	<u>1</u>	<u>2</u>	<u>3</u>	<u>4</u>	<u>5</u>
Number of taxa(a)	58	43	69	46	70
Expected number (based on Station 1)	--	58	58	58	58
$\chi^2$ contribution(b)	--	3.88(c)	2.09	2.48	2.48

(a) Number of unique taxa/life stages by combining five replicate samples for each station.

(b) For individual stations, the 1 degree of freedom  $\chi^2$  with  $P > \chi^2 = 0.05$  is 3.84.

(c) Significantly different from the expected value at Station 1 ( $P \leq 0.05$ ).

Note: For all stations combined, the calculated  $\chi^2$  with 4 Df = 10.93 ( $PR > \chi^2 = 0.028$ ).

TABLE D-7 LIST OF FISH SPECIES AND FAMILIES COLLECTED SCIPPO CREEK,  
AUGUST 1982(a)

<u>Family</u>	<u>Scientific Name</u>	<u>Common Name</u>
Cyprinidae (minnows)	<u>Notropis photogenis</u>	Silver shiner
	<u>Notropis chrysocephalus</u>	Striped shiner
	<u>Semotilus atromaculatus</u>	Creek chub
	<u>Rhinichthys atratulus</u>	Blacknose dace
	<u>Notropis spilopterus</u>	Spotfin shiner
	<u>Pimephales notatus</u>	Bluntnose minnow
	<u>Ericymba buccata</u>	Silverjaw minnow
	<u>Campostoma anomalum</u>	Stoneroller
	<u>Notropis stramineus</u>	Sand shiner
Catostomidae (sucker)	<u>Catostomus commersoni</u>	White sucker
	<u>Hypentelium nigricans</u>	Northern hog sucker
Centrarchidae (sunfish)	<u>Ambloplites rupestris</u>	Rock bass
	<u>Micropterus dolomieu</u>	Smallmouth bass
Percidae (perch)	<u>Etheostoma blennioides</u>	Greenside darter
	<u>E. caeruleum</u>	Rainbow darter
	<u>E. flabellare</u>	Fantail darter
	<u>E. nigrum</u>	Johnny darter
	<u>E. zonale</u>	Banded darter

(a) Names follow Robins et al. 1980.

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