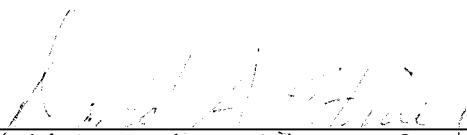
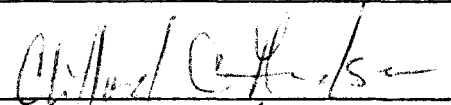


To the Graduate Council:

I am submitting herewith a dissertation written by Virginia Rose Tolbert entitled "Effects of Strip Mining Related Disturbance on the Benthic Insect Communities of Selected Streams in the New River Basin of East Tennessee." I recommend that it be accepted in partial fulfillment of the requirements for the degree of Doctor of Philosophy, with a major in Ecology.


David A. Etnier, Major Professor

We have read this dissertation
and recommend its acceptance:



Accepted for the Council:


Vice Chancellor
Graduate Studies and Research

EFFECTS OF STRIP MINING RELATED DISTURBANCE ON THE
BENTHIC INSECT COMMUNITIES OF SELECTED STREAMS
IN THE NEW RIVER BASIN OF EAST TENNESSEE

A Dissertation
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Virginia Rose Tolbert

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ABSTRACT

Four streams in the New River Basin in East Tennessee (one undisturbed and three disturbed) were sampled monthly to determine the effects of contour strip mining for coal on benthic insect communities. In addition to sampling for benthic insects, various physical parameters were measured at the time of sampling to help determine factors that cause changes in benthic community composition in streams disturbed by strip mining. Samples from each stream were analyzed monthly, seasonally, and for the total sampling period to determine effects on the number of species and individuals and on species diversity. For these three factors data were analyzed using analyses of variance, Student-Newman-Keuls means separation tests, and the Shannon diversity index. Benthic communities in the disturbed streams showed significant reductions in species, individuals and species diversity with mining disturbance. In order to determine which factors were primarily responsible for determining stream differences, multivariate discriminant analysis, using both independent and dependent factors, was employed. The variables found to be most discriminating in determining differences among streams were rainfall, stream flow, and turbidity. Three of the seven taxa shown to be discriminating variables in determining stream differences were significantly different between the control and disturbed streams.

Results clearly demonstrated the overriding influence of physical factors associated with rainfall and runoff, and resultant increased stream flow on benthic communities in streams disturbed by strip mining activity.

TABLE OF CONTENTS

CHAPTER	PAGE
I. INTRODUCTION	1
Rationale	1
II. LITERATURE REVIEW	5
Biological Effects of Acid Mine Drainage	7
Erosion and Sedimentation	10
Biological Effects of Siltation	15
III. STUDY AREA	19
Water Quality	28
IV. DATA COLLECTION AND ANALYSIS	33
Biological Data	33
Biological Analysis	35
Physical Data	36
Biological and Physical Data Analysis	37
Velocity Tolerance	39
Heavy Metal Content	42
V. RESULTS	45
Taxa	49
Individuals	54
Species Diversity	55
Physical Results	55
Biological and Physical Results	59
Velocity Tolerance	69
Heavy Metals	71
VI. DISCUSSION	75
Mortality	78
Litter	78
Change in Species Composition	79
Recolonization	79
Taxa and Individuals	80
VII. CONCLUSIONS	87

LITERATURE CITED	91
APPENDIX	102
VITA	104

LIST OF TABLES

TABLE		PAGE
1.	Summary of Characteristics of the Study Watersheds and Their Mining History	23
2.	Summary of Physical Stream Characteristics	26
3.	Taxa and Number of Individuals Present in the Four Study Streams in the New River Watershed	46
4.	Percent Composition of Individuals by Order for Each of the Four Study Streams	48
5.	Summary of Analysis of Variance and Student-Newman-Keuls Means Separation Test for Total Individuals, Total Taxa, and Species Diversity	51
6.	Results of the Analysis of Variance and Student-Newman-Keuls Means Separation Tests for the Major Orders in the Four Study Streams	53
7.	Comparison of Monthly Species Diversities for the Four Study Streams	56
8.	Mean Values for the Physical Parameters Included in This Study	57
9.	Summary of Discrimination Variables in Order of Selection for Determining Stream Differences	65
10.	Summary of Analysis of Variance and Student-Newman-Keuls Means Separation Test for the First Seven Taxa Selected by Discriminant Analysis	66
11.	Stream Classification Resulting from Discriminant Analysis	68
12.	Results of Laboratory Analysis to Determine at What Stream Velocity Taxa of Benthic Insects Are Dislodged from the Substrate	72
13.	Heavy Metal Content from Various Taxa Analyzed from the Four Study Streams	73
14.	Heavy Metal Content of Insect Tissue from Each of the Four Study Streams	103

LIST OF FIGURES

FIGURE	PAGE
1. Map of the Study Area in the New River Basin Showing the Major Streams in the Area	20
2. Map of the Coal Producing Regions of the Eastern United States	21
3. Generalized Stratiographic Sequence of the Pennsylvanian Coal Bearing Strata of the Study Area	24
4. Velocity Tolerance Flume	40
5. The Total Number of Individuals and Taxa from Each Stream for the Sampling Period August 1975 Through July 1976	50
6. Monthly Stream Flow Values for the Four Study Streams . . .	58
7. Differences in Suspended Solids for the Four Study Streams	60
8. Five Day Averages for Rainfall Preceding Sampling Dates	61
9. Group Centroids (Group Means) from Discriminant Analysis of the Four Streams	62
10. Summary of Discriminant Analysis F-matrix for Significant Differences Among the Four Streams	63
11. Standardized Discriminant Function Coefficients Showing the Location of Group Centroids Based on the Discriminating Functions	70

CHAPTER I

INTRODUCTION

Rationale

With increased emphasis on coal utilization (Kendrick, 1977), the amount of coal mined in the United States in the next decade is estimated to increase 50 to 200% (Freeman, 1974). Surface mining would account for at least 67% (Freeman, 1974; Appalachian Regional Commission, 1969). Surface mine permit requirements and reclamation legislation have become more stringent with increased public awareness of degraded water quality. As a result, more information is needed on the effects of surface mining on water quality and stream biota.

The importance of aquatic biota in predicting water pollution has been recognized since the early 1900's. Forbes and Richardson (1913, 1919) studied changes in the Illinois River resulting from stream pollution, and Jewell (1922) studied the fauna on an acid stream. These as well as other authors have studied the biological effects of water pollution with an increasing awareness of the importance of aquatic biota in predicting water quality (Purdy, 1926; Bartsch, 1948; Patrick, 1949; Warren, 1971; Cairns and Dickson, 1971; Olive, 1976; and others). The importance of benthic communities in determining water quality has been recognized by Wilhm and Dorris (1968), Mackenthun (1966), Anderson et al. (1965) and Tarzwell and Gaufin (1953). These communities were defined by Reid (1961) as a local assemblage of populations of bottom dwelling species maintained in an area delineated by environmental conditions.

Wilhm and Dorris (1968) and Mackenthun (1966) have proposed that sampling and examination of benthic organisms may provide a better interpretation of long term water quality than standard physical and chemical analysis or toxicity studies, since the latter methods indicate stream conditions only at the time of sampling. Cairns and Dickson (1971), Wilhm and Dorris (1968), and Mackenthun (1966) indicate that the benthic community should be examined because this community is indicative of both past and present water quality and conditions on the stream bottom. Sampling benthic communities can provide population comparisons at particular times and at different distances from sources of disturbance. Tarzwell and Gauvin (1953) indicated that populations of benthic invertebrates provide a more reliable criterion of disturbance than just the presence of a given species. In temperate freshwater streams, benthic communities are usually comprised of populations of dipterans, odonates, plecopterans, ephemeropterans, trichopterans, coleopterans, and megalopterans as well as other arthropods and annelids. Studies of benthic communities are particularly valuable since individual benthic organisms are relatively sessile, have long life histories (one year or longer), and are fairly easy to sample because of their limited mobility (Mackenthun, 1966; Cairns and Dickson, 1971).

The importance of coal mine drainage as a source of water pollution was reported in the 1930's by Carpenter and Herndon (1933) and Hodge (1938). Since the Appalachian region northward from West Virginia is affected primarily by acid mine drainage, the majority of studies on coal mine drainage have dealt with this factor. Numerous studies

(Brezina et al., 1970; Hyde, 1970; Herricks and Cairns, 1974; Cole et al., 1977) have dealt with the effects of acid mine drainage on water quality. Studies by Roback and Richardson (1969), Dills and Rogers (1974), and Herricks and Cairns (1974) have dealt with the effects of acid mine drainage on benthic organisms. Only a few articles from Kentucky and Tennessee have dealt even remotely with nonacid mine drainage effects on fresh water benthic communities. Carter (1964) and Hensley (1970) found Ephemeroptera and Trichoptera almost entirely lacking in mining disturbed streams while these orders comprised 28% of the total benthic fauna from an undisturbed stream in Kentucky. Branson and Batch (1972), studying the effects of strip mining on fishes in Kentucky, found a 90% reduction in total benthic population size and number of species as a result of increased strip mining related siltation. Talak (1977), studying the degree of recovery exhibited by benthic insects in several streams in the Cumberland Mountains of Tennessee, found that after fifteen to twenty years, the insect fauna recovered but never obtained the original community composition.

In the southern Appalachian region, studies have been conducted by Minear and Tschantz (1976), Plass (1976), Rose (1975), Curtis (1971, 1972, 1974), Collier et al. (1964, 1970), Ward (1977), Tung (1975) and Miller (1977) concerning various aspects of physical and chemical alterations of water quality by surface mining.

This study is an analysis of the impact of contour coal mining on benthic insect communities relative to population size, species richness, species diversity, and the ability of benthic communities to be

discriminating factors in determining differences between streams disturbed and undisturbed by mining activity. This study was conducted in the New River Basin of the Cumberland Mountains of Tennessee and is part of an Appalachian Resources Council project at the University of Tennessee to determine the effects of surface mining for coal on (1) water quality, (2) movement of metals through terrestrial and aquatic systems, (3) stream hydrology, and (4) stream biota.

CHAPTER II

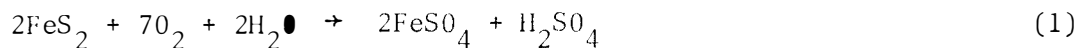
LITERATURE REVIEW

Acid mine drainage is one of the most adverse effects resulting from surface mining for coal. In the Appalachian region approximately 5,700 miles of waterways are continuously polluted by acid mine drainage. This pollution results in \$3.5 million in added annual costs to industrial water users, municipal water supplies, navigation, and public facilities. The general environmental and aesthetic degradation of affected areas, as well as destruction of aquatic life and deterrent to waterbased recreation may well exceed these other more readily measured costs (Appalachian Regional Commission, 1969). Altered water quality resulting from all types of mine drainage has occurred in 10,500 miles of Appalachian streams. Of this total, 5,700 miles were affected by acid drainage with most of the affected streams occurring from West Virginia and Maryland northward. By 1974 there were 6,300 miles of streams affected by acid mine drainage. This is 93.5% of the total acid drainage affected streams for the nation (HRD-Singer, 1974).

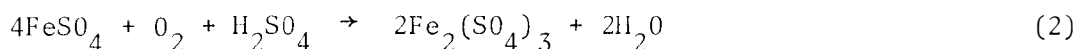
In the Appalachian region 72% of the acid mine drainage originates in underground mines, 12% from surface mining, 10% from a combination of surface and underground mines, and 6% from other mine related sources. Of the amount from underground sources, 80% is from abandoned mines (Appalachian Regional Commission, 1969).

Acid mine drainage originates as a result of the oxidation of sulfide minerals (primarily iron sulfide). The source of this iron

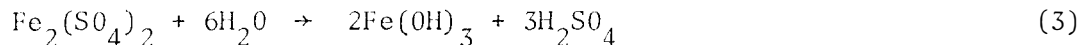
sulfide is from pyritic materials associated with coal seams. With the exposure of iron sulfide to oxygen in a moist environment, ferrous sulfate and sulfuric acid are produced.



The ferrous sulfate is further oxidized (2) to produce ferric sulfate.



The ferric sulfate is further oxidized (3) to produce ferric hydroxide (a precipitate) and more sulfuric acid (Herricks and Cairns, 1974).



These reactions can result in a pH as low as 2.0 in receiving waters, depending on the amount and composition of the pyritic material in the coal and overburden, the amount of contact time between the pyrite and water, the amount of available oxygen, and the amount of water present for reaction with the pyrite (Yeasted and Shane, 1975; Appalachian Regional Commission, 1969).

Acid mine drainage is not considered a serious problem in Tennessee, since sulfur concentrations in coal and the associated strata are lower and the proportion of alkaline materials are higher in the exploited Pennsylvanian strata of the southern Appalachian region (Elmore, 1967). This is supported by Curtis (1972), Larson et al. (1976), Plass (1976), and Minear and Tschantz (1976) who found that both pH and alkalinity often increase in streams in this area after strip mining disturbance. In streams undisturbed by mining the low pH is controlled by nutrients and acidity leached from vegetation; since runoff from surrounding watersheds acquires only a small mineral load and therefore has a low alkalinity. After mining disturbance, alkalinity increases as a result

of leaching of formerly inaccessible alkaline materials from the disturbed surface areas of the watershed. Increased alkalinity results in increased acid neutralizing capacity of mine drainage and a higher pH (Larson et al., 1976).

Biological Effects of Acid Mine Drainage

Studies of the effects of acid mine drainage can be divided into two groups based on the type of strip mining used and the terrain. In flatter locations, area strip mining is used whereby successive pits are formed. Each pit is filled by the overburden removed from the next pit. As a result of no additional fill for the final pit, "strip pit" lakes are formed. Contour strip mining results most frequently in stream pollution, since contour strip mining is carried out on generally steep mountain sides.

Effects of acid mine drainage are discussed in a review article by Smith and Frey (1971). Studies by Dinsmore (1958), Riley (1950), and Burner and Leist (1953) of the ecology of strip mine formed lakes indicated physical alteration of both lake substrate and acidity and increased community alteration with time since lake formation.

There are numerous studies on the effects of acid mine drainage on stream systems. Parsons (1957), Herricks and Cairns (1974), and Dills and Rogers (1974) found benthic populations decreased as a result of acid drainage, but they found recovery occurring with increased distance downstream from the source of acid drainage. Patrick (1956), Roback and Richardson (1969), Cairns et al. (1971) also found reduced benthic populations and changes in community structure as a result of acid mine drainage.

Although these authors all found reduced community diversity as a result of acid mine drainage, the community compositions differed with the different studies. In Pennsylvania, Roback and Richardson (1969) found Plecoptera, Ephemeroptera, and Odonata completely eliminated from study streams receiving acid mine drainage. In Alabama, Dills and Rogers (1974) found that members of these orders were present but in reduced numbers. They also found the benthic fauna in the disturbed streams to be dominated primarily by chironomids, ceratopogonids, and megalopterans. Herricks and Carins (1974) found Ephemeroptera and Plecoptera but not Trichoptera reduced as a result of acid mine drainage. Parsons (1957) also found these orders reduced but found an insect community of several dipteran species and the megalopteran Sialis sp. well adapted to acid mine drainage. The adaptation of Sialis sp. to acid drainage is supported by findings of Tarter (1975). The Appalachian Regional Commission's (1969) study of acid mine drainage presents a table showing that from pH 5.0-7.0 there is a diverse benthic fauna with Trichoptera, Ephemeroptera, Plecoptera and Simuliidae (Diptera) present. Below pH 4.5, the fauna changes to primarily Trichoptera, Odonata, and Diptera (Chironomidae). Roback (1974) showed that below a pH of 4.5 the benthic fauna was dominated by Coleoptera, Diptera, and Hemiptera with Megaloptera and Trichoptera comprising the remaining fauna.

Bell and Nebeker (1969) and Bell (1971) found that most aquatic insects emerged successfully at a pH of 4.0 to 5.9 and that as pH decreased emergence success also decreased. The trichopterans, Hydropsyche betteni Ross and Brachycentrus americanus (Banks), were

found to be most tolerant of low pH. Plecoptera and Odonata were moderately tolerant, and the ephemeropteran Ephemerella subvarians (McDonnough) was fairly sensitive with a TL_{50} (30 day tolerance limit where 50% of the test individuals survive) of pH 5.38 and 50% emergence success of pH 5.9. Although the studies of acid drainage streams cited above correspond fairly well with those findings of pH tolerance by Bell and Nebeker (1969) and Bell (1971), there are differences in community composition that are the result of disturbance related factors other than pH.

Dills and Rogers (1974) and Herricks and Cairns (1974) found increases in ionic content with decreased pH as a result of acid drainage. The former found a high negative correlation between hydrogen ion concentration and benthic species diversity; however, they indicated that the ionic content may exert as great an influence on benthic organisms as pH. Herricks and Cairns (1974) experimentally considered acid as an individual problem and found benthic community recovery associated with both time and distance from the source of disturbance. They also considered a disturbed stream and found benthic recovery associated with distance from the source of disturbance. They concluded that secondary biological stress may occur as a result of heavy metal toxicity.

Several authors have considered benthic insects to be the most important indicators of heavy metal pollution (Nehring, 1976; Rehwoldt et al., 1973). This is because (1) insects concentrate metals in relation to the proportion of metals in the water, (2) they concentrate the metal

over a predictable time period, and (3) they are more tolerant of heavy metals than fish (Nehring, 1976). Clubb et al. (1975) found that during periods when there is a high metal content in the water, greater quantities of metals are concentrated, but during periods of reduced metal content there is a linear rate of loss which may serve to lower or prevent mortality. Both Warnick and Bell (1969) and Rehwoldt et al. (1973) found that benthic insects are more able to withstand heavy metal input than are fish.

From the above studies on acid mine drainage which show depressed pH, increased ionic content, and increased or decreased turbidity depending on the pH, it is evident that the effects of contour strip mining on benthic communities vary with the geology of the area and with the geographical areas studied. From these studies, it is also evident that pH is generally considered the controlling factor of benthic community structure since lower pH levels are found in most coal producing regions and other factors are often thought to be a resulting effect of the acid (Curtis, 1971). Sulfate content (Herricks and Cairns, 1974; Biesecker and George, 1966) and specific conductance (Pickering and Musser, 1970) have been proposed as indicative of mine drainage since pH indicates little about the chemical nature of the water which can vary, depending on the geology, among streams of the same pH.

Erosion and Sedimentation

Acid production from coal mining is primarily the result of underground mining. In contrast the primary adverse effect of surface mining is spoil bank erosion and siltation (Greene, 1975). Approximately

50% of the total sediment found in waterways is classified as resulting from land-disturbing activities associated with mining, construction, agriculture and silviculture, 30% from natural or geologic sources and 20% from municipal, industrial and individual sources (Grissinger and McDowell, 1970). Whenever mining cuts are made to expose coal seams, the overburden or rubble removed from over the coal seam is pushed over the edge of the cut and down the mountainside. These spoil banks are a major source of erodable materials, although access and haul roads in some instances may contribute as much erodable material as the spoil bank (Curtis, 1972). These findings are supported by Collier et al. (1970) and Reed (1977) who have also found considerable erodable material as a result of road construction. The soil type, length and degree of slope, climate, and amount of rainfall all affect the amount of erosion resulting from surface mining (Greene, 1975). The extent of the watershed disturbed and the extent of reclamation carried out in the watershed should also be added to this list since these would influence the amount of erodable materials available. The degree and length of slope are probably two of the most important factors in determining erodable material since these two factors would determine, in the absence of heavy rainfall and water accumulation within the spoil bank, the stability of the spoil bank. As the steepness of the slope and amount of rainfall increase, erosion and sedimentation become a more serious problem since the loosened spoil material is more unstable (Goldberg, 1972; Collier et al., 1970) and subject to gully erosion.

In areas disturbed by contour strip mining along steep slopes, it is not unusual for total sediment yields to be at least a thousand times greater than from similar unmined watersheds (Tschantz, 1977). In the Cumberland Mountains of East Tennessee, Minear and Tschantz (1976) and Tschantz (1977) have found that the median particle size removed from the spoil banks, carried in the streams, and deposited behind the weirs has increased steadily with time since mining disturbance ceased. Changes in particle size distribution were the result of increased velocity and turbulence in storm runoff. Tschantz and Overton (1978) found that with moderate storms silt deposited on the streambed was dislodged. With the advent of another more intense storm, larger underlying particles were then removed. These findings are supported by Collier et al. (1970) who also found increased particle size. The increase in particle size was a result of slower movement toward the streams by the larger particles and in some cases the proximity of spoil banks to the stream margins. Collier et al. (1970) found that the particle size increased sooner after mining in areas where the spoil bank was near the stream margin. Higher values for extreme suspended solids in the water column correspond to days during which rainfall occurred or days immediately following a rainfall (Minear and Tschantz, 1976).

In eastern Kentucky, Curtis (1972) discussed the importance of siltation from improperly maintained haul roads and from strip mined areas. He found an erosional rate of 5.9 tons per acre per year in disturbed watersheds as compared to .7 tons per acre per year in

undisturbed watersheds. In a Virginia watershed disturbed in area by approximately 11% as a result of road construction, Reed (1977) found that erosion averaged 151 tons per acre per year. This was two thousand times that expected from forested land in that basin. Collier, Pickering, and Musser (1964) found an annual sediment yield of 20 to 30 tons per square mile prior to mining activity. Since mining activity began, the annual sediment yield has ranged from 617 to 3,010 tons per square mile or sixty-nine times the amount from the control stream. Curtis (1972) concluded that larger storms were responsible for increased sediment but the maximum concentrations depended primarily on the extent of mining activity and disturbance.

In studying the impact of strip mining on water quality in the New River Basin, the source of 70% of the coal produced in Tennessee, Minear and Tschantz (1976), Tschantz (1977), and Bowers (1977) observed sedimentation similar to that found by Curtis (1972) and Collier et al., (1964) in Kentucky. In the disturbed streams, suspended solids regularly exceeded 1000 mg/l and at times exceeded 10,000 mg/l following rainfall days. In one of the disturbed streams, the suspended solids concentration in a peak storm flow was greater than that predicted for an urban storm of the same intensity (Ward, 1977). In comparison, the suspended solids were consistently below 25 mg/l in the control stream regardless of season or precipitation (Tschantz, 1977). Collier et al. (1970) found that in the disturbed streams there were seasonal differences in sedimentation as the result of extended periods of rain during the winter, and brief intense storms during the summer yielded higher sediment concentrations.

In contrast they found that no seasonal variability in the amount of sediment occurred in the control stream. The differences were attributed to the fact that the unprotected (nonforested) strip mined areas were more sensitive to form, intensity, and duration of precipitation and to antecedent moisture conditions.

Minear and Tschantz (1976) and Tschantz and Overton (1978) found an increase in median particle size carried into the disturbed streams and deposited behind the weirs with increased time since mining ceased. Changes in size particle distribution were the result of increased velocity and turbulence changes in storm runoff. Tschantz and Overton (1978) found that with moderate storms silt deposited on the streambed was dislodged with the advent of another more intense storm. The larger sediment exposed when the silt was removed was then carried by the increased stream velocity.

In studies of streams not receiving acid mine drainage, it is possible to examine factors in addition to siltation that might cause changes in benthic communities. Studies, covering a seven year period in Breathitt County, Kentucky, conducted by Curtis (1971, 1972) and Dyer and Curtis (1977), showed that, as a result of strip mining disturbance, ionic content increased following mining. Ionic concentrations of sulfate, calcium, and magnesium showed the greatest increase, but aluminum, manganese, iron, and zinc were also found to increase. In eastern Tennessee, Minear and Tschantz (1976) found an increase in pH in the streams disturbed by mining as a result of alkaline material being exposed to weathering. Curtis (1972) also found increased pH as a result of mining.

Biological Effects of Siltation

Branson and Batch (1972), studying the effects of siltation from strip mining in Kentucky, found a high level of siltation — as much as 15 to 30 times that of an undisturbed stream. This increased siltation eliminated fishes from headwater streams and reduced reproduction of fishes downstream. They also found that benthic food organisms were reduced by at least 90%. Batch (personal communication) found ephemeropteran and plecopteran faunas altered and numbers of individuals and species reduced in streams disturbed by strip mining siltation. Hensley (1970) found ephemeropteran, plecopteran and trichopteran species reduced by acid mine drainage and siltation. Tolbert (1977) and Tolbert and Vaughn (1978) found reduced numbers of taxa and individuals in streams disturbed by contour strip mining in East Tennessee.

Hynes (1960) described two principal ways that faunas of streams and rivers may be affected by inert solids. First, when solids are suspended in the water, they may eliminate plant and algal growth by reducing light penetration. This eliminates the food of herbivorous organisms and the autochthonous detritus produced from plant and algal break up (Hynes, 1960; Chutter, 1969). Second, when inert solids settle out of the water, they smother algal growth, kill plant roots and mosses and alter the substrate by clogging the interstices between substrate particles. As long as allochthonous materials are available as an energy source (Hynes, 1970; Cummins et al., 1973), communities can remain virtually the same in the absence of autochthonous materials. When allochthonous materials are removed as a food source by increased current resulting from runoff (Hynes, 1970), community alteration occurs.

Cordone and Kelly (1961), reviewing the literature on the effects of silt and sand, found that in many instances the density of the streambed fauna was considerably reduced by sedimentation. Tebo (1955), studying the effects of siltation resulting from logging, found that there was a statistically significant reduction of bottom dwelling organisms below the mouth of a logged watershed. Taff and Shapovalov (1935), as reported by Cordone and Kelly (1961), found that streams affected by silt from mining activity always had fewer organisms than streams in unmined areas. Turbidity and solids deposition have been found to be the primary factors associated with siltation that affect benthic organisms (Bartsch and Schilpp, 1953).

According to Hynes (1970) substrate is largely controlled by stream velocity. Stream velocity determines the distribution of substrate particles, deposits and carries away silt, transports and shreds leaf detritus, and is necessary for respiration by benthic organisms (Rabeni and Minshall, 1977; Hynes, 1970; Reice, 1974). If the velocity is fairly constant throughout the year, the substrate, no matter what type, is usually stable. If the current is slow part of the year, sand and silt from erosion will build up deposits on the substrate. These deposits are then flushed downstream or to the stream margins with increased flow or flooding. Tebo (1955) found that the alteration of the substrate by silt accumulation resulted in decreased substrate stability with benthic organisms being subject to decimation by flood waters. Branson and Batch (1972) found this to be true also for amphibians when they found salamanders entombed under rocks after the streambed was covered with clay accumulations of two to six inches from strip mine runoff.

Chutter (1969) found that elmids (Coleoptera) and Caenidae (Ephemeroptera) as well as chironomids (Diptera) were adversely affected by silt and sand, particularly during the summer months. He also found that Simulium sp. (Diptera) move away from areas of silt deposition. Hynes (1970) stated that Glossosoma sp. (Trichoptera) occur only where its food gathering mechanism is not impeded by sand or silt. Chutter (1969) found some species to be particularly sensitive to silt abrasion and others to be intolerant of siltation because of its interference with food acquisition. On stony substrates, the presence of silt reduces and changes the fauna (Savage and Rabe, 1973; Sprules, 1947). This is especially true for intolerant species of Plecoptera, Ephemeroptera, and Trichoptera. With siltation the proportion of chironomid individuals in the benthic fauna increased (Sprules, 1947). Moffett (1936) and Jones (1951) also found chironomids to be the predominant fauna in disturbed streams. Rabini and Minshall (1977) also found changes in benthic fauna as a result of siltation.

In general the fauna of clean, stony streams is richer than that of silty reaches in terms of both number of species and in total biomass (Hynes, 1970). This difference may be the result of several factors. Silt accumulates on the substrate clogging the interstices between substrate particles. This clogging reduces the number and kinds of available habitats as well as the autochthonous food supply (Hynes, 1970), thus reducing the benthic fauna. Species generally select substrates based on substrate stability (Lauff and Cummins, 1964; Scott, 1958). As shown by Tebo (1955) and Hynes (1970) these stable areas are unavailable

in streams receiving silt loads. With variations in stream current, resulting from increased runoff, the unstable silt accumulations are disturbed. Movement of eroded materials and dislodged substrate result in abrasion to some species (Savage and Rabe, 1973; Chutter, 1969). Harker (1953), Harrison and Elsworth (1958), and Hynes (1970) found benthic individuals crushed by moving substrate. Larimore (1972), Elliot (1967), and Elliot and Minshall (1968) found increased drift with increased runoff from rainfall and Brusven (1974) found drift to increase as the concentration of suspended solids in the water increased. Thus, with erosion and resultant siltation of streams, the various life stages of benthic organisms are reduced.

CHAPTER III

STUDY AREA

The 955 sq km (382 sq mi) New River Basin is located entirely within Scott, Morgan, Anderson, and Campbell counties of eastern Tennessee (Figure 1). This area, which is part of the Appalachian coal producing region extending from central Pennsylvania into northwestern Alabama (Figure 2), produces approximately 70% of the coal mined in Tennessee (Larson et al., 1976; Minear and Tschantz, 1976). The New River flows northward and joins Clear Fork near New River, Tennessee to form the Big South Fork of the Cumberland River. The significance of the New River has increased with the enactment of PL 93-251 in 1974, which authorized the U. S. Army Corps of Engineers to establish the Big South Fork National River and Recreational Area (Minear and Tschantz, 1976).

The New River Basin is characterized by generally rugged terrain, moderate temperature (13°C or 55°F annual average), and greater than 127 cm (50 in.) of rainfall annually. Elevation ranges from 332 m (1,090 ft.) to greater than 1,006 m (3,000 ft.) above mean sea level with an average slope of 14 degrees. Slopes of the streambeds range from 7 to 26% (Tung, 1975). The extreme relief of the area results in the loss of most precipitation as runoff. As a result, most of the smaller streams in the area cease flowing periodically during the summer months because of lack of groundwater recharge (Gairola, 1947; Minear and Tschantz, 1976).

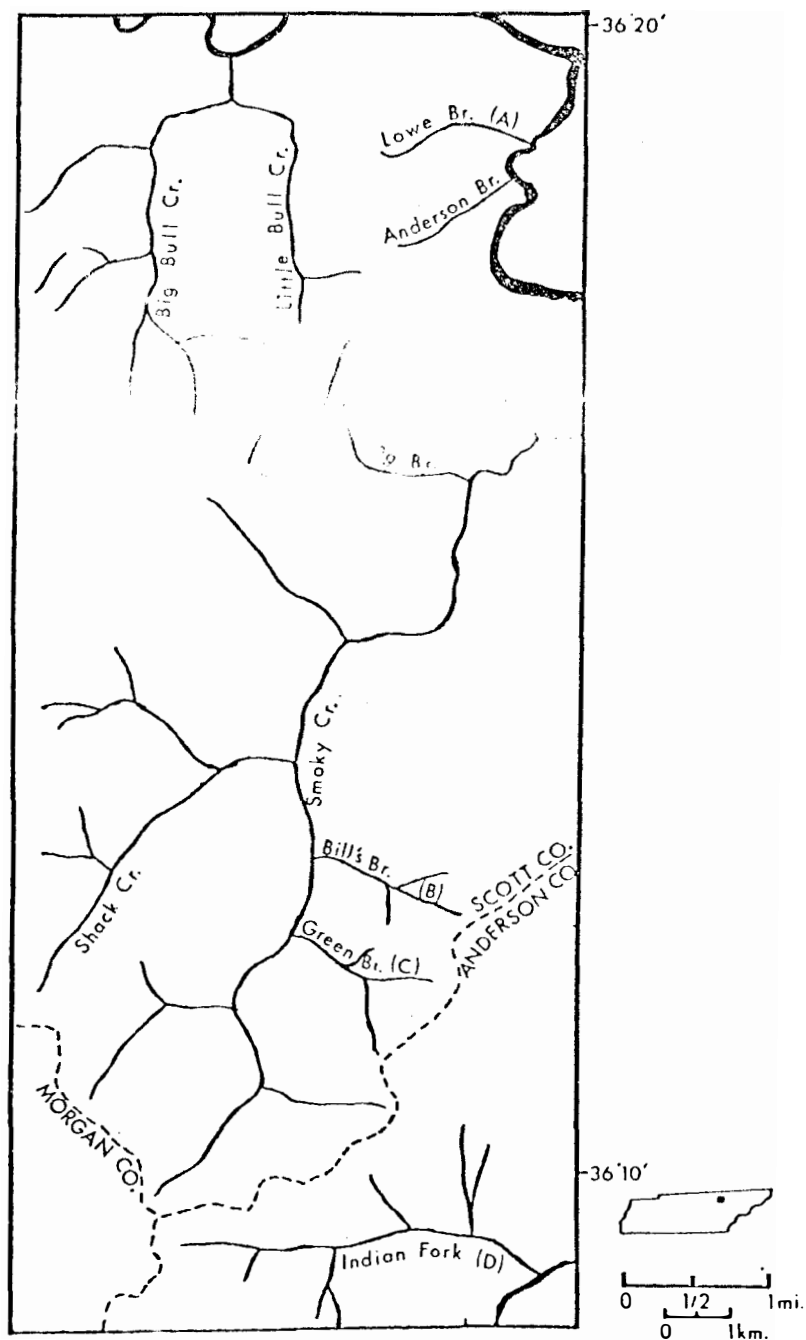


Figure 1. Map of the study area in the New River Basin showing the major streams in the area. The four study streams are indicated as follows: Lowe Branch (A), Bill's Branch (B), Green Branch (C), and Indian Fork (D).

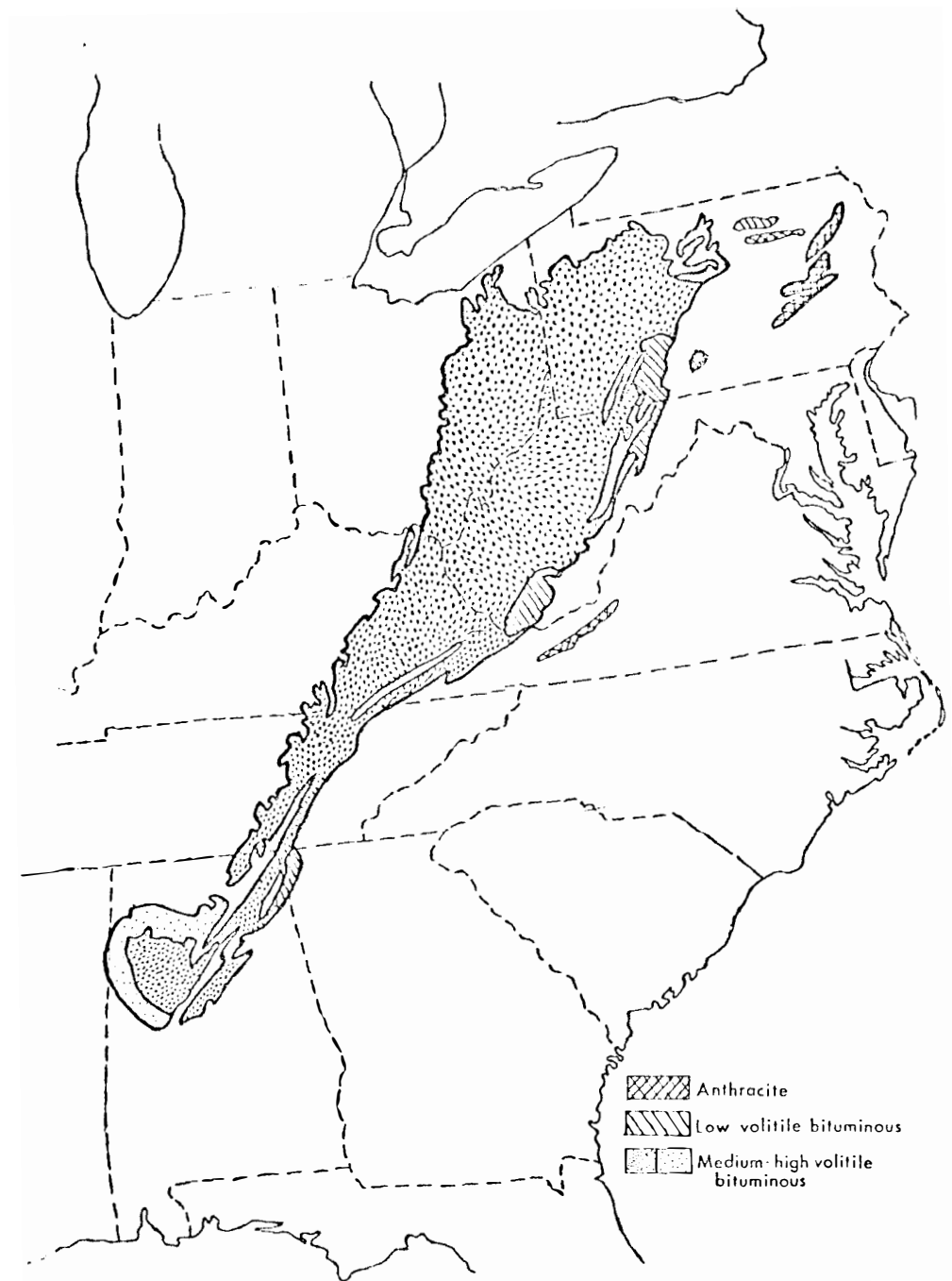


Figure 2. Map of the coal producing regions of the Eastern United States.

Surface coal mining began in the New River Basin in the early 1940's and has continued to the present. This has resulted in approximately 5% of the total area of the basin being disturbed by 1975 (Tung, 1975). In some of the small, primary watersheds within the basin, considerably greater disturbance has occurred (See Table 1 for examples).

Geologically, the New River fluvial system drains the Northern Cumberland Plateau of eastern Tennessee and is contained in the Wartburg Basin, which is a physiographic subprovince of the plateau. Within the New River Basin, exposed rock is of Pennsylvanian age and consists largely of conglomerate, sandstone, siltstone, and shale with smaller amounts of coal (Tung, 1975). The lower parts of the middle Pennsylvanian sequence contain primarily thick sandstone layers with lesser amounts of coal. The upper parts of the sequence contain greater amounts of coal, thicker shale layers, and reduced layers of sandstone.

Those coal seams of the middle Pennsylvanian sequence of primary mining interest in the New River Basin are Big Mary at approximately 686 m (2,250 ft.) elevation and the Pewee seam at about 793 m (2,600 ft.) elevation. Other seams mined in conjunction with these two primary seams are Windrock, Walnut Mountain and Red Ash (Figure 3). Descriptions of these coal seams and associated strata are contained in Briggs and Rule (1976) and Minear and Tschantz (1976).

In order to obtain information on streams not influenced by mining activity within the New River Basin, primary watersheds were chosen. The four watersheds under study (Lowe Branch, Bill's Branch, Green Branch, and Indian Fork) are located in Anderson and Scott counties within the

Table 1. Summary of Characteristics of the Study Watersheds and Their Mining History.

<u>Stream</u>	% Slope of Streambed	Area of Watershed (Hectares)	Area Disturbed (Hectares)	% Area Disturbed	Mining Activity began ended
Lowe Branch	12.6	238	--	--	-- --
Bill's Branch	26.0	174	17.6	9.8	1974 - 1975
Green Branch	17.6	357	86.2	24.1	1972 - 1975
Indian Fork	8.4	1,119	211.0	18.9	1950's - current

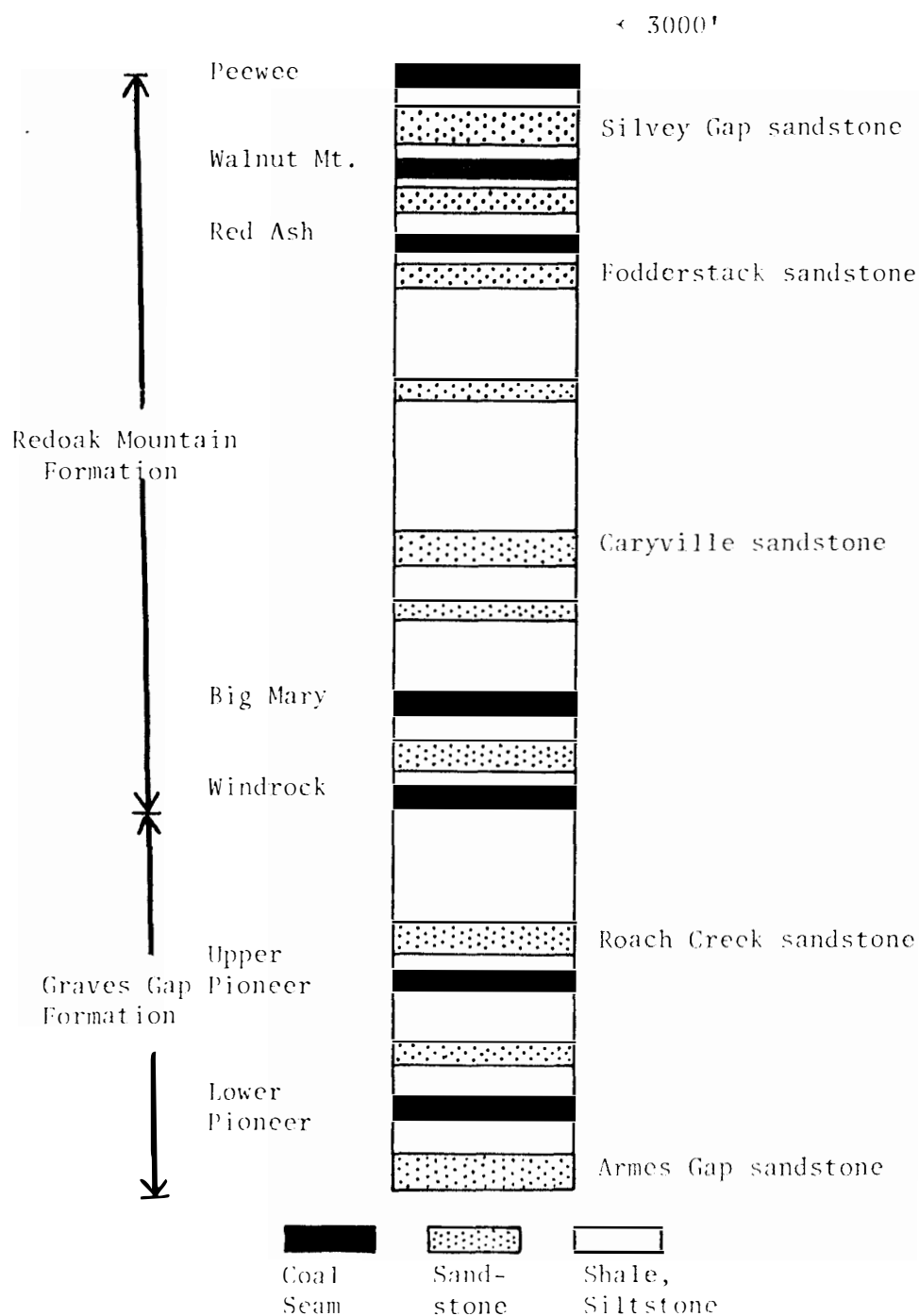


Figure 3. Generalized stratigraphic sequence of the Pennsylvanian coal bearing strata of the study area.

New River basin. Lowe Branch is the only stream in a watershed undisturbed by strip mining. The locations of these watersheds are shown in Figure 1 (p. 20) with brief descriptions in Table 1 (p. 23). The study watersheds for the project were chosen based primarily on projected land use, particularly with respect to planned strip mining, in order to insure an undisturbed control stream throughout the study. The watersheds are geologically similar, and with the exception of Indian Fork, which is sparsely populated near its mouth, access is limited. Based on the geology and the fact that basin discharge results solely from rainfall and ground water, background water quality of the streams is relatively uniform and of "near pristine" characteristics (Minear and Tschantz, 1976). The study of water quality in these study streams had been reported by Minear and Tschantz (1976), Tschantz (1977), and Tschantz and Overton (1978); metal movement through both terrestrial and aquatic systems by Rule (1975), and stream hydrology by Tschantz (1977) and Tschantz and Overton (1978).

Lowe Branch (Figure 1), a first order stream (Horton, 1945; Leopold et al., 1964) undisturbed by strip mining, served as the control stream. The stream is characterized by a substrate composed of rock, gravel, and sand with abundant allochthonous material. The current is moderate to slow most of the year with flow in the lower reaches ceasing during parts of the summer. During periods of heavy rainfall the stream velocity shows little increase (maximum flow 1.6 cfs, Tschantz, 1977). The stream width varies from 1 to 2 meters and the depth from 5 to 20 cm with the average width being 1.5 m and the average depth 12 cm (Table 2).

Table 2. Summary of Physical Stream Characteristics.

Stream	Width (m)	Depth (cm)	Substrate	Stream Margins	Current
Lowe Branch	1-2	5-20	Rocks, gravel, and debris underlain with sand	Mixed hardwood forest	Moderate to slow
Bill's Branch	1-2.5	5-30	Large rock, rock and gravel underlain with sand and silt. Silt-algal cover on rocks.	Mine tailings and mixed hardwood forest	Moderate
Green Branch	1-3	8-40	Large rock, rock, and gravel cemented to substrate with clay. Silt-algal cover on rocks.	Mine tailings and mixed hardwood forest	Moderate
Indian Fork	2-3	5-35	Rock and gravel cemented to substrate with clay, some sand and silt, extensive precipitate.	Mine tailings, some cleared areas, and mixed hardwood areas	Moderate to swift

The substrate of Bill's Branch, a first order stream, is primarily large rock, rock, and gravel underlain with sand and silt. There is a silt-algal cover on the rocks and layers of silt among the rocks (Table 2, p. 26). The current is moderate, except during periods of rainfall when the velocity may exceed 9.5 cfs (U. S. Geol. Survey, 1977), and stream width ranges from 1.0 to 2.5 m and depth from 5 to 30 cm. The average width is 2.0 m and depth 18 cm.

Green Branch (Figure 1, p. 20), a first order stream, is characterized by a substrate of rock and gravel cemented together by clay particles, a few large rocks, and a silt-algal cover on the rocks. The current is moderate most of the time but may exceed 22.6 cfs (Tschantz and Overton, 1978) during periods of rainfall. The average stream depth is 16 cm and the average width 2.5 m (Table 2).

Indian Fork (Figure 1), a first order stream, has a rock and gravel substrate with considerable amounts of clay and small gravel particles among the rock and gravel. Most of the rocks in the stream are cemented to the streambed either by deposited silt or they are the only substrate particles that have not been dislodged by increased stream flow resulting from rainfall. During periods of rainfall the stream velocity may frequently exceed 26.7 cfs (Tschantz and Overton, 1978). There is little allochthonous material accumulated on the substrate of either Indian Fork or Green Branch. The material that is available is found in leaf packs caught on branches which extend into the stream. There is considerable deposition of ferric hydroxide on the substrate and adhering to clay particles accumulated in the stream (Perhac, 1974); this precipitate imparts an orange color to the substrate and the water. Sources of the

ferric hydroxide and acidic waters of Indian Fork are primarily open auger holes produced in the latter stages of strip mining, and leakage from abandoned deep mines, including some that are sealed at the mouth (Minear and Tschantz, 1976). There is also accumulated ferric hydroxide in Bill's Branch and Green Branch but not enough to influence the color of the water or substrate. Indian Fork has an average depth of 15 cm and average width of 2 m. The stream current is moderate to swift (Table 2, p. 26). Indian Fork has been influenced by mining activity longer (approximately 25 years) than the other two disturbed streams (Table 1, p. 23). The exposure of a nearly solid firm bottom of cemented rocks and gravels at times in the disturbed streams is the result of increased stream flow during storms which scour the streambed, dislodge most loose substrate materials, and wash them downstream (Tschantz, 1977).

Water Quality

In the New River Basin as well as parts of eastern Kentucky (Curtis, 1972), and West Virginia (Plass, 1976), the pH and alkalinity increase following strip mining disturbance. This is in contrast to the generally low pH and alkalinity found in undisturbed watersheds. The State of Tennessee has set the minimum pH for mining and industrial discharge at 6.0. In the New River Basin ground water and surface water accumulate little mineral content so that alkalinity is low. As a result, the undisturbed streams are in marginal compliance or sometimes actually in violation of state pH standards. Water quality data for the four streams under study are available from related parts of the total Appalachian Resources Council project on the New River Basin.

Minear and Tschantz (1976) found little variation in water quality parameters with time and little difference among unmined watershed streams in contrast to greater differences in water quality among the disturbed watershed streams. They also determined that extensive disturbance, particularly the resultant generation of unconsolidated spoil materials, promote leaching of previously inaccessible minerals. This increased leaching of minerals was determined to be the result of increased contact time between diverted surface water and subsurface spoil materials. This diversion and retention of surface water within the spoil bank results in significant increased dry weather flow in streams disturbed by mining (Curtis, 1972; Tschantz, 1977). The dry weather flow in the disturbed streams results from the gradual release of spoil bank seepage and bench impoundment of water (Minear and Tschantz, 1976).

The water quality variables reported by Minear and Tschantz (1976) reflect a relationship between the extent and type of disturbance and/or time since disturbance. Bill's Branch, which was disturbed for one year (1974-1975), has lower water quality than the undisturbed stream but considerably better water quality than was found in the other two disturbed streams. The pH and alkalinity fluctuations in Indian Fork are attributed to the acid sources in this watershed resulting from auger holes and abandoned deep mines. When stream flow is high, these sources are not sufficient to lower pH below acceptable limits. However, with decreased pH and resultant drop in alkalinity, pH oscillations are greater than in the other disturbed streams (Minear and Tschantz, 1976).

Curtis (1972), Plass (1976), and Minear and Tschantz (1976) have found increases in the levels of calcium and magnesium in streams following mining disturbance. These ions have been found to increase during summer months when water temperature is higher and to decrease within two to three years after mining ceased. Minear and Tschantz (1976) found that for the disturbed streams in the New River Basin, the levels of calcium and magnesium were considerably higher on a percentage and absolute basis than the control streams. Lowe Branch, the control stream, was classified as a soft water stream, Bill's Branch as soft but with increasing hardness, Green Branch as moderately hard to hard, and Indian Fork as hard to very hard (Minear and Tschantz, 1976).

Sulfate concentrations have been proposed as an indicator of acid mine drainage (Herricks and Carins, 1974); however, sulfate has been found by Curtis (1971; 1972), Plass (1976), and Minear and Tschantz (1976) to increase in strip mining disturbed streams where acid mine drainage was not found. Sulfate concentrations have been found to increase with the extent of disturbance and to decrease with time since disturbance (Dyer and Curtis, 1977; Minear and Tschantz, 1976). Curtis (1972), Plass (1976) and Minear and Tschantz (1976) have found lags in sulfate production following disturbance and long term production of sulfates following disturbance, which result in less acid production in the southern Appalachian region. Generally the control stream had iron and manganese levels considerably below those found in the disturbed streams. Minear and Tschantz (1976) found that 88% of the samples from Bill's Branch, 88% from Green Branch and 98% of the samples from Indian

Fork exceeded the state drinking water standards (.05 mg/l) for manganese, but did not exceed the mining permit standards. For iron concentrations the three disturbed streams exceeded the mining permit levels 43.5, 47.6, and 95.4% of the time (Bill's Branch, Green Branch, and Indian Fork). These three streams exceeded the state drinking water standards 78.3% of the time (Bill's Branch), 73.8% (Green Branch), and 100% of the time (Indian Fork). Most of the iron found in the disturbed streams was found as total iron concentration usually as a precipitate; while soluble iron comprised only a small part of the total iron.

In the undisturbed watersheds the suspended solids load remained below 25 mg/l and never exceeded 50 mg/l. Suspended solids ranged from 2 to 10,160 ppm in the disturbed streams and from 1 to 38 ppm in the control stream. The average suspended solids for each stream were Lowe Branch 6 ppm, Bill's Branch 1452.3 ppm, Green Branch 2045.1 ppm, and Indian Fork 329.4 ppm for the total sampling period. The turbidity measures (JTU's) ranged from 4 to 1000 units in the disturbed streams and from 0 to 4 in the control stream. The average turbidity for the streams were Lowe Branch 0.3 JTU's, Bill's Branch 83.8 JTU's, Green Branch 236.16 JTU's, and Indian Fork 44.4 JTU's.

Based on the findings of Minear and Tschantz (1976), Tschantz (1977), Rose (1975), Rule (1975), and Minear (1978) for the four watersheds under study, it can be seen that water quality variations in the undisturbed streams are minimal and essentially independent of stream-flow and antecedent climatological conditions. In contrast, the disturbed streams, particularly Indian Fork, show extreme oscillations in concentrations of

individual variables from one sampling period to the next. For Indian Fork and to some extent Green Branch, the greatest changes in water quality variables coincide with rainfall events. Minear and Tschantz (1976) showed that with increased area of the watersheds disturbed, rainfall resulted in increased levels of sediment being carried into the stream. With increased time since mining disturbance the concentration of various water quality parameters increased in a systematic fashion, before some parameters such as sulfate, aluminum and chloride decreased two to three years after disturbance ceased.

CHAPTER IV

DATA COLLECTION AND ANALYSIS

Biological Data

From July 1975 through July 1976, Surber samples were taken on a monthly basis, weather and roads permitting, from four streams in the New River Basin. A 15 threads/cm mesh net size was chosen since in the disturbed streams since finer meshed nets became so clogged with coarse sand, coal, and silt that samples could not be taken. The Surber sampler was modified in that the lower edge of the frame was rounded and flexible and could be pressed firmly against the substrate. This edge conformed more to the rocky substrate of running water areas than the typical Surber sampler and prevented organisms from escaping under the bottom edge of the frame and net. Monthly, eight 0.2 m^2 samples were taken from each stream using the modified Surber sampler.

The criterion employed to determine benthic sampling adequacy was established as a standard error to mean ratio ($SE/\bar{x}$) of less than 10% where SE is equal to the Standard Error and $\bar{x}$ is equal to the mean of the samples from one collection date. An individual sample size of 0.2 m^2 with eight samples per stream was determined to be necessary for the $SE/\bar{x}$ to be less than 10% of the mean (Southwood, 1966) and to meet sample adequacy for this study. Numbers were chosen at random from a random numbers table and were used to determine the location of sampling sites in each stream for each sampling date. Benthic samples were taken

from running water sections nearest the determined random site in each of the four streams. Running water areas were chosen for sampling since running water was necessary to wash dislodged organisms into the net, and a rock and gravel substrate has been found to be the most suitable habitat for the largest variety of benthic organisms (Sprules, 1947; Mackenthun, 1966; Hynes, 1970; Rabini and Minshall, 1977). Restricting sampling to random sites within one particular habitat (stratified random sampling) enabled better comparisons between disturbed and undisturbed streams. This was the case since similar substrate types for each sample reduced the differences among samples within each stream. The running water habitat in this study was defined as a water current of at least 2 cm/s passing over rocks and gravel (≥ 2.0 cm in diameter). Samples were not taken from the same site in the streams in any two consecutive months, since consecutive sampling could bias the number of organisms collected. However, this departure from randomness was not considered important since the probability of sampling the same site twice was very small given the length of the streams sampled.

The benthic samples and associated retained substrate were preserved in the field in 70% ethanol, then separated in the laboratory from the collected substrate materials using a Bausch-Lomb binocular microscope. The benthic invertebrates were then identified to species where possible using keys for Plecoptera (Frison, 1935; Claassen, 1931; and Ross and Ricker, 1971), Trichoptera (Ross, 1944; Flint, 1964; Wiggins, 1977; and Schuster and Etnier, in press), Ephemeroptera (Berner, 1950; Burks, 1953; Edmunds et al., 1976), and Diptera, Coleoptera, and Megaloptera (Usinger, 1974).

Biological Analysis

The benthic communities of the four streams were first compared in terms of the number of species, total number of individuals, and the species diversity. Species diversity was calculated using the Shannon diversity index (Shannon and Weaver, 1963) where

$$H' = -\sum P_i \text{Log} P_i$$

where P_i is the number of individuals of each species divided by the total number of individuals of all species. This is one of the more widely used indices employed to estimate environmental stress in aquatic communities (Wilhm, 1967; Wilhm and Dorris, 1968; Cairns et al., 1971; and Stoneburner et al., 1975). Maximum H' values are obtained when each species contains the same number of individuals, and minimum H' values are obtained when all individuals belong to the same species. This index was used since samples were taken randomly from running water; thus meeting the criteria set forth by Pielou (1969) that samples be random from a larger parent population and be representative of the larger population. Species diversities were determined by month, by season, and for the total sampling period for each of the four streams. Diversity values in this study are based on eight 0.2 m^2 samples/stream/month, since this sample size and number were necessary to adequately sample populations in the various streams. The values for the different species were then pooled since pooled samples tend to result in a less variable measurement of diversity (Wilhm, 1967).

Both monthly and seasonal data were analyzed using one-way analysis of variance (Sokal and Rohlf, 1969) to determine if there were significant

differences among streams either monthly or seasonally for the number of taxa, the number of individuals, or for species diversity. Where significant differences occurred either in the monthly or seasonal analyses, Student-Newman-Keuls means separation tests (Sokal and Rohlf, 1969) were used to determine which streams were significantly different from each other.

Physical Data

In addition to the eight samples taken to determine the benthic community composition and differences among streams for each collection date, replications of various physical parameters were measured concurrently at each sampling site.

Measurements of pH were made in the field at the time benthic samples were taken, using a Gibson portable ion meter, in close proximity to the benthic samples in each stream. Water temperature and dissolved oxygen were determined at the time of sampling using a YSI (Yellow Springs Instrument Co.) Temperature-Oxygen meter, Model #51. Micro-velocity measurements were taken as close to the substrate as possible (2.5 cm above the substrate), using a Teledyne-Gurley Pigmy meter, Model #65, to determine approximate velocity where organisms were living. Turbidity measurements were made in the field at the time of sampling with a Hach Chemical Engineers Kit using percent transmittance for measurement. For each modified Surber sample, the percent of the sampling area covered by litter, rock, gravel, and sand were estimated in the field.

In addition to the above parameters measured at the time of sampling, all streams have continuous digital stream flow gauges and digital rainfall

monitors operated by the U. S. Geological Survey. Other parameters such as alkalinity, total, suspended, and dissolved solids, pH, turbidity, sulfate, iron, manganese, calcium, and magnesium were monitored on a bi-weekly basis using standard methods by members of the Environmental Engineering Program, Department of Civil Engineering, University of Tennessee, Knoxville.

Biological and Physical Data Analysis

In order to determine which environmental parameters were responsible for benthic community differences, multivariate analysis (Tatsuoka, 1971; Nei, et al., 1976) were employed. The Student-Newman-Keuls means separation test enables multiple comparisons of means in a stepwise manner to determine the source of the significant differences (Harris, 1975).

In order to determine differences among the various disturbed streams and the control stream based on both biological and environmental differences, multivariate statistical analyses were performed. Multivariate statistics provide both descriptive and inferential techniques to analyze experimental data which involve sets of variables either as predictors or as measures of performance (Harris, 1975). By the use of multivariate analysis, all possible effects are considered in one test thereby reducing the experimental error rate that occurs with multivariate significance tests using the same data. One of the best methods of multivariate analysis for field data is discriminant analysis, although this method is not yet widely used. Cody (1968) used discriminant analysis to reduce multidimensional habitat data for breeding birds

to a single function. By using abundance of different herb species as variates, Norris and Barkham (1970) were able to discriminate among several beech woodlands. Green (1971) used multiple discriminant analysis to identify functional niche separation of molluscs among several physical and chemical variables. Dueser, et al. (1976) used discriminant functions based on structural forest characteristics to define niche partitioning among four species of small mammals and Mann (1977) used discriminant analysis to define niche partitioning among forest herbs.

Discriminant analysis is appropriate for the analysis of stream relationships in relation to environmental factors for a number of reasons. First, it reduces the variables to new independent (orthogonal) linear additive functions of the original variables (Cooley and Lohnes, 1971). These functions become axes describing a new space in which the original values of the variables for each species can be plotted. Since the axes are derived to maximize differences between groups and since the discriminant functions are orthogonal to one another, mean group differences (centroids of groups) are more easily identified. Secondly, since the coefficients of the original variables used in the discriminant functions can be ranked as to their contribution to the functions, ecological interpretation of the axes is usually possible. Third, discriminant analysis does not assume linearity of ecological gradients as do principal components and cluster analysis (Ihm and Groenewoud, 1975). It assumes that environmental variables used in the analysis have normal distributions (Ihm and Groenewoud, 1975). Fourth, if the

underlying statistical assumptions can be met, the analysis provides a statistic similar to the F ratio of analysis of variance to differentiate between group centroids. A significant F ratio for overall discrimination indicates that there is a significant difference in the group means of the streams studied in relation to the environmental variables measured.

Although discriminant analysis assumes normality of variates, this assumption is not actually justified by most data sets. Harris (1975) discussed the robustness of normal-curve based F tests which can be considered valid for even U-shaped populations and concluded that this robustness probably extends to multivariate tests also.

In order to determine differences between streams both the dependent (benthic samples) and independent (environmental) variables were used. The within group differences of these variables were minimized and the between group differences maximized using this technique. This enables determination of factors that result in group differences.

Velocity Tolerance

As a result of observations of increased stream flow following rainfall and increased deposition of sediment materials, laboratory studies were undertaken to determine what velocities various benthic species could withstand before being dislodged and carried downstream. A sixty foot plexiglass flume one foot wide and one foot deep was used to determine velocity tolerance (Figure 4). This flume is part of the Civil Engineering hydrology laboratory. It extends from a holding tank which has a continuous water source and gate to control flow rate to a second tank where the water is collected and weighed to determine

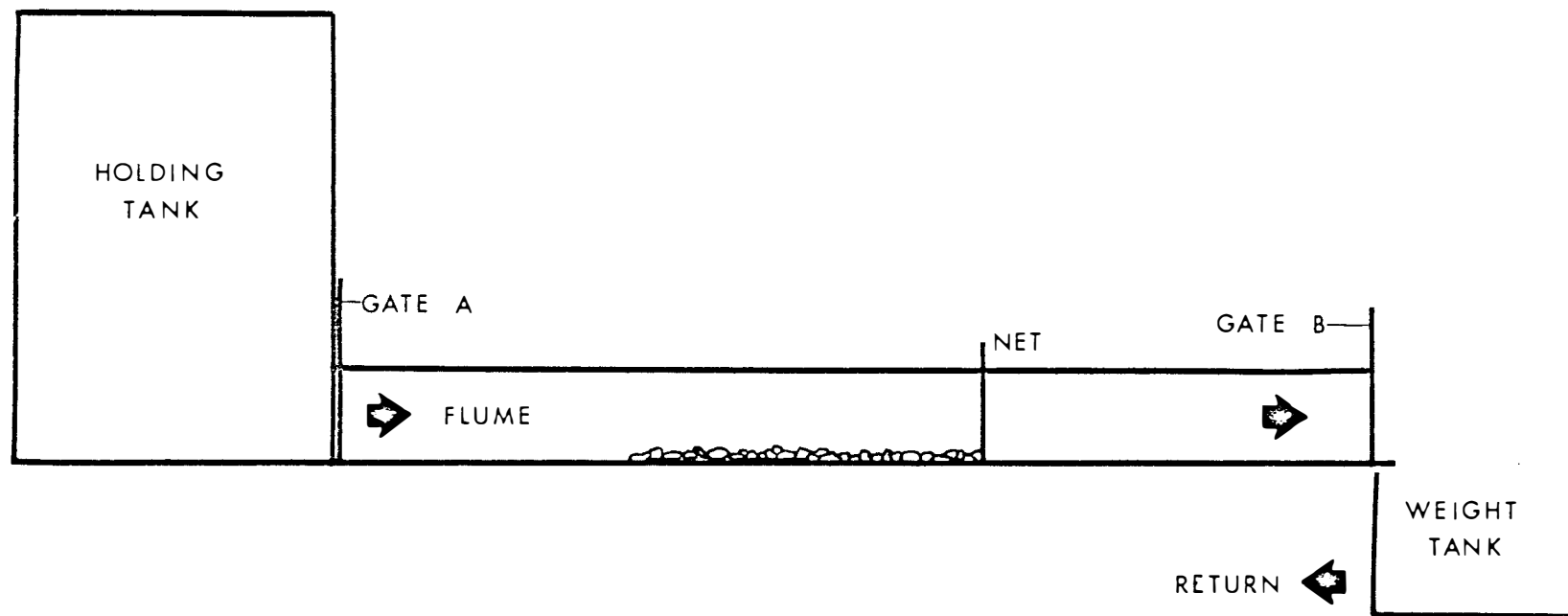


Figure 4. Velocity tolerance flume.

velocity. The water is then pumped through the system and back to the holding tank. The velocity of the stream is controlled electronically by increasing or decreasing the flow from the holding tank with gate A (Figure 4) and by increasing or decreasing the opening of gate B thus controlling the amount of water leaving the stream.

Substrate materials (rock $\geq$ 2.5 cm, gravel $\geq$ 1 cm, and gravel $>$.5 cm) were placed in the stream covering the bottom to a depth of approximately 5 cm. The difference in substrate sizes allowed the benthic organisms to "choose" their preferred substrate. Water depth in the stream varied with the experimental velocities used. However, the substrate materials were covered to a depth of at least 8 cm at all times. Nets were fitted securely to the interior margins of the flume so that no organisms could escape. As a back-up precaution against rock or gravel being carried back through the system to the holding tank, a screen was fastened securely across the lower end of the flume.

Benthic invertebrates were collected from the running water areas of a natural stream the day of sampling, transported to the laboratory in gallon coolers to maintain stream temperature, then gradually acclimated from stream temperature (8°C) to the study stream temperature (12°C) to reduce the effects of transfer to the study stream. An oxygen level of 10 mg/l was maintained by aeration in the study stream. The benthic organisms were allowed to acclimate in the study stream and "choose" their substrate for thirty minutes at a velocity of 10 cm/s prior to beginning experimentation. Individuals of Ephemerella sp., Isonychia sp., Baetis sp., Stenonema sp., Heptagenia sp., Nemoura sp.,

and Isoperla sp. were used in the experiment because they were large enough to identify without a microscope and since enough individuals could be obtained for the tests. Individuals of the retreat case building hydropsychids were not used in the laboratory velocity analyses since a thirty minute acclimation period was not adequate for them to construct their nets. Adequate numbers for other genera could not be obtained to carry out the experiment the same day that individuals were collected.

Velocities were measured in the study stream using the Teledyne-Gurley Pigmy meter. Velocity was gradually increased from 11 cm/s to 30 cm/s and held at that point for fifteen minutes. At the end of that time the net was checked for dislodged organisms and the number of individuals and genus of each was noted. This pattern was repeated for velocities of 60 to 120 cm/s. Preliminary tests had shown that more disturbance and drifting of the benthic began to occur at 85 cm/s so that velocity was increased in 10 cm/s increments from 60 to 130 cm/s following the same pattern as originally established. Above 130 cm/s velocity measurements could not be obtained.

Data were analyzed for means, standard error, and range so that difference in drift rate for the different taxa could be obtained.

Heavy Metal Content

Samples were collected from each of the four streams in September and April in order to determine if there were differences in metal content in benthic insect tissue from the undisturbed and disturbed streams. One and one-half hours sampling time were spent in each of the

streams for each of the sampling periods. A minimum of 25 m² running water was sampled in each of the disturbed streams and 10 m² in the control stream in an attempt to secure adequate numbers of individuals of the various taxa to total approximately 1 gram wet weight of body tissue. It was impossible to obtain adequate weights of some taxa for analysis, because of the scarcity and small size of individuals in the disturbed streams. Insects were sampled using a kick net and Surber type sampler in order to obtain as many individuals as possible using both techniques. The insects were separated from the substrate in the field by sieving samples through 2.5 cm and 1.25 cm hardware cloth onto screen wire where individuals were hand collected. Individuals were stored dry from each stream and frozen in the field using dry ice. Samples were returned to the laboratory, sorted by taxa, and weighed using an Ainsworth type 12 balance. All individuals of the same taxon from the same stream were weighed together for one sample. The samples were placed in acid washed teflon vials and dissolved in 10 ml concentrated Nitric Acid (2N). Samples were allowed to digest for two days and were then analyzed in the Environmental Engineering Laboratory using the Perkins 300 flame spectrophotometer according to standard methods for the various ions. Determinations of Pb, Fe, and Zn content were made, since these were the ions that showed major increases following strip mining activity (Minear and Tschantz, 1976; Upham, 1975; Curtis, 1972).

These analyses were performed to determine if heavy metals were factors influencing faunal differences in the control and mining-disturbed

streams based on differences in concentrations in insect tissues. The concentration of metals in insect tissues from this study were compared with those found by Warrick and Bell (1969), Nehring (1976), Rehwoldt et al. (1973), Chubb et al. (1975) to determine if metals were a cause of faunal differences among the streams.

CHAPTER V

RESULTS

Based on the total sampling period, August 1975 through July 1976, the major orders of aquatic insects were represented by similar numbers of taxa in this study (Table 3). Many species of these major taxa are intolerant of surface mining related changes (Branson and Batch, 1972; Hynes, 1970) that occur in the three disturbed streams (Table 3). Insect orders containing species sensitive to environmental disturbance (Roback, 1974) were most abundant in Lowe Branch (Table 3). Five species of Ephemeroptera (mayflies), six species of Plecoptera (stoneflies), seven species of Trichoptera (caddisflies), and two species of Coleoptera (beetle) were found only in Lowe Branch, the control stream (Table 3). Of the forty-five taxa collected, only ten were found in all four streams, and all but five were found in the control stream (Table 3).

The percentage of the total benthic fauna was determined for each aquatic order (Table 4) to facilitate comparison of the four streams. From this it was found that numbers of the dominant insect orders changed from the undisturbed stream, Lowe Branch, to the disturbed streams. In the control stream, Ephemeroptera comprised 57.9%, Plecoptera 26.8%, and Diptera (true flies) 10.4% of the total individuals. Ephemeroptera made up 65.9% of the total individuals from Bill's Branch (Table 4). This was the largest percent composition of any order in any

Table 3. Taxa and Number of Individuals Present in the Four Study Streams in the New River Watershed.

	Lowe Br.	Bill's Br.	Green Br.	Indian Fork
Ephemeroptera				
Baetidae				
<u>Baetis</u> spp.	338	319	41	9
<u>Ephemerella</u> (<u>walkeri</u> group)	201	12	1	-
<u>Isonychia</u> sp.	-	61	-	-
<u>Paraleptophlebia</u> sp.	58	-	-	-
<u>Pseudocloeon</u> sp.	2	-	-	-
Heptageniidae				
<u>Epeorus</u> sp.	36	43	-	-
<u>Heptagenia</u> (<u>flavescens</u> group)	212	-	-	-
<u>Heptagenia</u> (<u>maculipennis</u> group)	-	55	1	1
<u>Stenonema</u> <u>annexum</u> Traver	229	-	-	-
<u>Stenonema</u> <u>pulchellum</u> (Walsh)	-	55	-	2
Ephemeridae				
<u>Hexagenia</u> sp.	5	1	-	-
Plecoptera				
Perlidae				
<u>Acroneuria</u> <u>abnormis</u> Newman	18	8	3	-
<u>Phasganophora</u> <u>capitata</u> (Pictet)	2	-	-	-
Peltoperlidae				
<u>Peltoperla</u> sp. (probably <u>maria</u>)	8	12	21	-
Leuctridae				
<u>Leuctra</u> <u>sibleyi</u> Claassen	52	4	11	1
<u>Leuctra</u> <u>carolinensis</u> Claassen	20	-	-	-
Perlodidae				
<u>Isogenus</u> (<u>Cultus</u>) <u>decisus</u> Walker	2	-	-	-
<u>Isoperla</u> <u>clio</u> (Newman)	317	30	13	3
Chloperlidae				
<u>Sweltsa</u> <u>mediana/onkes</u>	10	-	-	-
<u>Chloroperla</u> <u>terna</u> Frison	29	2	2	-
Nemouridae				
<u>Nemoura</u> (<u>Amphinemura</u>) <u>nigritta</u> Provancher	7	6	9	6
Capniidae				
<u>Allocaenia</u> sp.	90	26	12	-
Tanyopterygidae				
<u>Taeniopteryx</u> <u>metaqui</u>	3	1	1	-
Trichoptera				
Rhyacophilidae				
<u>Glossosoma</u> <u>nigrior</u> Banks	1	2	1	-
<u>Rhyacophila</u> sp.	1	-	3	-

Table 3 (continued)

	Lowe Br.	Bill's Br.	Green Br.	Indian Fork
Hydropsychidae				
<u>Hydropsyche ventura</u> Ross	14	3	-	-
<u>Diplectrona modesta</u> and <u>D. metaqui</u>	49	51	55	6
Philopotamidae				
<u>Dolophilodes distincta</u> (Walker)	9	-	-	-
Limnephilidae				
<u>Neophylax</u> sp.	38	4	-	-
Diptera				
Tipulidae				
<u>Tipula</u> sp.	41	17	9	3
<u>Eriocera</u> (probably <u>fultonensis</u>)	9	-	-	-
Chironomidae				
Chironominae	99	46	105	19
Tanypodinae	56	52	79	12
Simuliidae				
<u>Simulium</u> (probably <u>vittatum</u>)	2	-	11	-
Dixidae				
<u>Dixa</u> sp.	3	-	10	-
Stratiomyiidae				
<u>Stratiomyia</u> sp.	-	-	1	-
Ceratopogonidae				
<u>Atrichopogon</u> sp.	-	-	1	-
Unidentified dipteran taxa				
A	4	1	-	-
B	1	-	-	-
C	1	-	-	-
D	1	-	-	-
Coleoptera				
Elmidae				
<u>Stenelmis</u> sp.	36	10	5	4
Psephenidae				
<u>Psephenus herricki</u>	53	2	-	-
Dryopidae	3	2	-	-
Megaloptera				
Sialidae				
<u>Sialis</u> sp.	1	-	-	-
<u>Corydalus cornutus</u>	16	1	-	1
Hemiptera				
Veliidae	6	1	1	1
Total Individuals	2084	829	405	77
Total Taxa	42	29	24	14

Table 4. Percent Composition of Individuals by Order for Each of the Four Study Streams.

<u>Order</u>	<u>Stream</u>			
	Low Branch	Bill's Branch	Green Branch	Indian Fork
Ephemeroptera	51.9	65.9	10.6	15.6
Plecoptera	26.8	7.8	17.8	13.0
Diptera	10.4	14.0	53.6	44.2
Trichoptera	5.4	7.5	16.5	10.4
Coleoptera	4.4	1.7	1.6	5.2
Megaloptera	.8	.1	---	1.3
Hemiptera	.3	.2	.2	10.4
Other	.1	---	.5	4.1
Total Individuals	2084	829	405	77

of the four streams; although the number of individuals of the order was less than from the control stream. Diptera had the second and Plecoptera the third most abundant number of individuals in Bill's Branch. In Green Branch the most abundant individuals were Diptera (53.6%). Individuals of Plecoptera and Trichoptera were the second and third most abundant in this stream. Diptera were also most abundant in Indian Fork with Ephemeroptera and Plecoptera being the second and third most abundant orders.

The total number of individuals of all taxa collected from all four study streams was approximately 3400. Lowe Branch, the control stream, had the greatest number of individuals (2084) of all taxa and the greatest number of taxa (42) (Figure 5). Lowe Branch had 2.5 times as many individuals as Bill's Branch, and 27 times as many individuals as Indian Fork. Indian Fork, whose watershed has been disturbed the longest by mining activity, had the fewest number of individuals (77) and the fewest taxa (14) of all four streams (Figure 5).

Taxa

Analysis of variance (ANOVA, Sokal and Rohlf, 1969) of taxa for the total sampling period showed a significant difference in the number of taxa between the four streams ($p > .001$, Table 5). Means from the analysis of variance were then analyzed in a stepwise fashion using Student-Newman-Keuls means separation test (Sokal and Rohlf, 1969). The numbers of taxa from the control stream, Lowe Branch, were significantly different from the three disturbed streams (Table 5). Bill's Branch and Green Branch, which have been disturbed for a short period (1 and 3 years

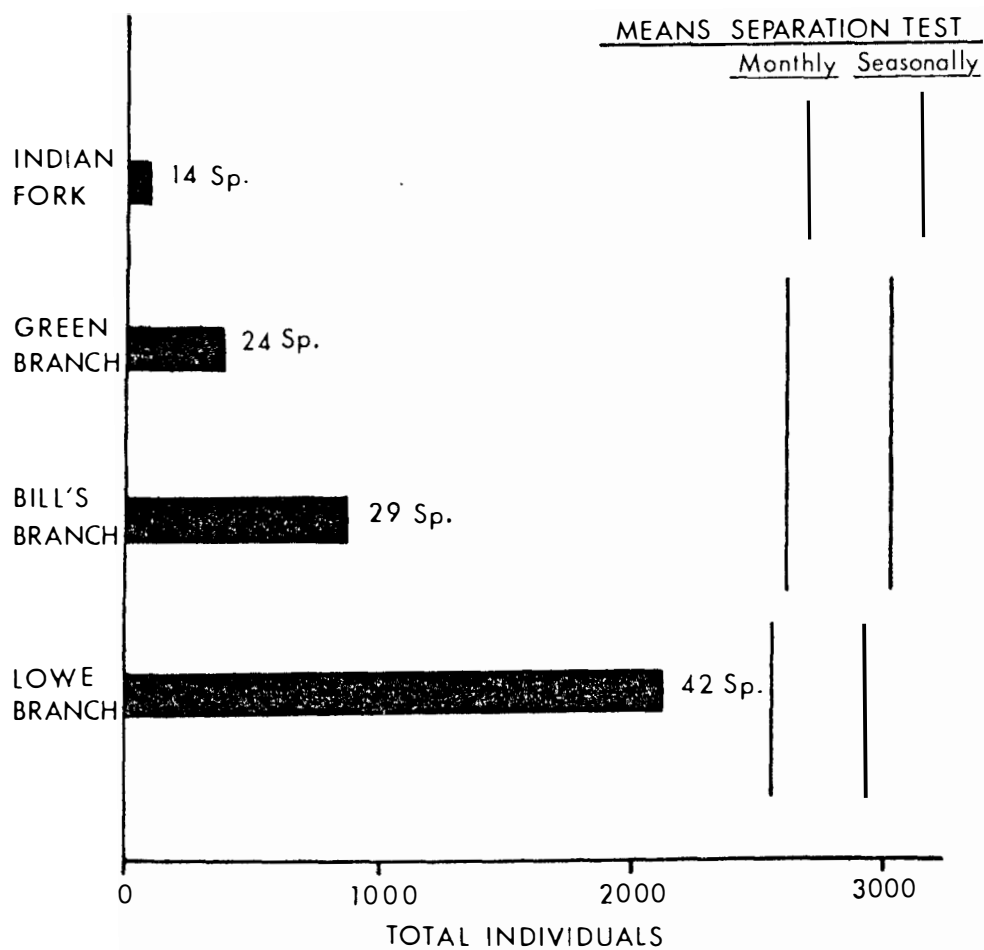


Figure 5. The total number of individuals and taxa from each stream for the sampling period August 1975 through July 1976. These numbers are based on eight samples per stream for ten sampling dates for Lowe Branch, Bill's Branch, Green Branch, and nine sampling dates from Indian Fork. Streams joined by vertical lines were not significantly different ($p < .05$) from Student-Newman-Keuls Mean Separation Test for species diversity.

Table 5. Summary of Analysis of Variance and Student-Newman-Keuls Means Separation Test for Total Individuals, Total Taxa, and Species Diversity. Underlined Streams Indicate Streams That Are Not Significantly Different from One Another. All Analyses Were Significant at Least at the .01 Level.

Variable	F-Test	Stream ¹			
		LB	BB	GB	IF
Total Individuals					
Seasonally	5.29	<u>LB</u>	<u>BB</u>	GB	IF
Total time	6.73	<u>LB</u>	<u>BB</u>	<u>GB</u>	<u>IF</u>
Total Taxa					
Seasonally	9.12	<u>LB</u>	<u>BB</u>	<u>GB</u>	IF
Total time	32.61	<u>LB</u>	<u>BB</u>	<u>GB</u>	<u>IF</u>
Species Diversity					
Seasonally	7.40	<u>LB</u>	<u>BB</u>	<u>GB</u>	<u>IF</u>
Total time	19.17	<u>LB</u>	<u>BB</u>	<u>GB</u>	<u>IF</u>

¹LB = Lowe Branch (control)

BB = Bill's Branch

GB = Green Branch

IF = Indian Fork

respectively), were not significantly different for numbers of taxa, but were significantly different from Lowe Branch and Indian Fork.

The number of taxa from the four streams were also analyzed seasonally using analysis of variance and were found to be significantly different ($p > .01$, Table 5). Student-Newman-Keuls means separation test showed Lowe Branch and Green Branch were not significantly different seasonally for total taxa and Green Branch did not have significantly different taxa from Indian Fork. Indian Fork, seasonally, had significantly different taxa than Lowe Branch and Bill's Branch (Table 5).

Of the four major aquatic orders, three were found to be significantly different in terms of the number of taxa in the four streams (Table 6). Analysis of variance results showed Ephemeroptera to be significantly different ($p > .01$) both seasonally and for the total sampling period. Student-Newman-Keuls means separation test for both seasonal and total time analysis showed no significant difference in the number of Ephemeroptera or Plecoptera among streams (Table 6). For the total sampling period, there was a significant difference in the number of dipteran individuals. There was no significant difference in the number of trichopteran individuals either seasonally or for the total sampling period between the streams. The control stream was significantly different only from Indian Fork. The three disturbed streams were not significantly different from each other (Table 6) in terms of the number of dipterans.

In summation, results of analyses of variance and Student-Newman-Keuls means separation tests for the total taxa found in the four

Table 6. Results of the Analysis of Variance and Student-Newman-Keuls Means Separation Tests for the Major Orders in the Four Study Streams.

Taxa	F-Test	P >	Streams			
Emphemeroptera						
Total time	6.56	.01	LB	BB	GB	IF
Seasonally	7.93	.01	LB	BB	GB	IF
Plecoptera						
Total time	4.76	.01	LB	BB	GB	IF
Seasonally	3.04	NS				
Diptera						
Total time	4.6	.01	LB	BB	GB	IF
Seasonally	3.06	NS				
Trichoptera						
Total time	2.01	NS				
Seasonally	1.82	NS				

NS = not significantly different.

streams, indicated that Lowe Branch, the control stream had significantly more taxa of benthic organisms than any of the three disturbed streams when considered both seasonally and for the total time period. When considered both seasonally and for the total sampling period, the numbers of taxa from Bill's Branch and Green Branch were very similar. Seasonally the number of taxa from Green Branch and Indian Fork were similar.

Results of the Student-Newman-Keuls means separation tests showed that of the four major insect orders (Ephemeroptera, Plecoptera, Diptera, and Trichoptera) only the number of taxa belonging to the order Trichoptera were significantly different among streams.

Individuals

The total number of individuals from each stream was also analyzed seasonally and for the total sampling period using analysis of variance and Student-Newman-Keuls means separation test. Seasonally there were significant differences in the number of individuals between streams (Table 5, p. 51). The number of individuals from Lowe Branch and Bill's Branch was not significantly different. However, the number of individuals from the control stream was significantly different from the other two disturbed streams. The total number of individuals in the three disturbed streams was not significantly different from each other seasonally. For the total sampling period the number of individuals of all taxa was not significantly different in Lowe and Bill's branches (Table 5). They were, however, significantly different from the other two disturbed streams. Green Branch and Indian Fork were not significantly different from each other (Table 5).

Species Diversity

As a result of decreased numbers of species and individuals of many species in the disturbed streams, species diversities in the disturbed streams were reduced monthly (Table 7), seasonally (Table 5, p. 51), and for the total sampling period (Table 5). Species diversities of Lowe Branch were consistently higher than those of any of the three disturbed streams. Diversities of Bill's Branch and Green Branch were consistently very similar for all periods analyzed. For all periods analyzed, diversities of Indian Fork were lowest.

Species diversities among streams were compared seasonally and for the total sampling period using one-way analysis of variance. Seasonally the species diversities were significantly different ($p > .05$). Student-Newman-Keuls means separation test (Table 5) showed Lowe Branch, the control stream, to be significantly different from all the disturbed streams. The species diversities of Bill's Branch and Green Branch were not significantly different from each other but both were significantly different from Indian Fork (Table 5).

Physical Results

Mean values were determined for the eight physical parameters used in this study (Table 8). This table shows that generally values for pH, temperature, oxygen, and microvelocity varied little from stream to stream. Mean stream flow also varied little among streams; however, for different sampling dates there were considerable differences in stream flow among streams (Figure 6). There were significant differences ($p > .01$) in the amount of rainfall, litter, and turbidity between the

Table 7. Comparison of Monthly Species Diversities for the Four Study Streams.

Month	Lowe Branch	Bill's Branch	Green Branch	Indian Fork
August	2.32	1.90	1.65	1.47
September	1.92	2.03	1.92	1.11
October	1.88	1.69	1.65	0.00
November	2.25	1.74	1.16	NS
December	2.10	1.13	.89	0.00
February	2.11	1.02	1.63	.77
April	2.30	2.08	1.86	.98
May	2.10	.70	1.25	1.24
June	2.32	1.43	1.46	.65
July	2.89	2.15	1.44	.85

Table 8. Mean Values for the Physical Parameters Included in This Study.

Parameter	Streams			
	Lowe Branch	Bill's Branch	Green Branch	Indian Fork
pH	6.79	6.93	7.07	6.47
Stream Flow (cm/s) ¹	.59	.42	.07	.50
Rainfall (inches) ¹	.53	1.77	1.28	6.05
Temperature (°C)	13.08	13.32	12.11	13.64
Oxygen	7.61	7.02	8.20	7.31
Litter (%)	55.43	37.21	35.72	18.39
Microvelocity	19.95	21.45	21.19	19.50
Turbidity (% transmittance)	96.66	90.19	87.00	71.08

¹Data from Department of Civil Engineering.

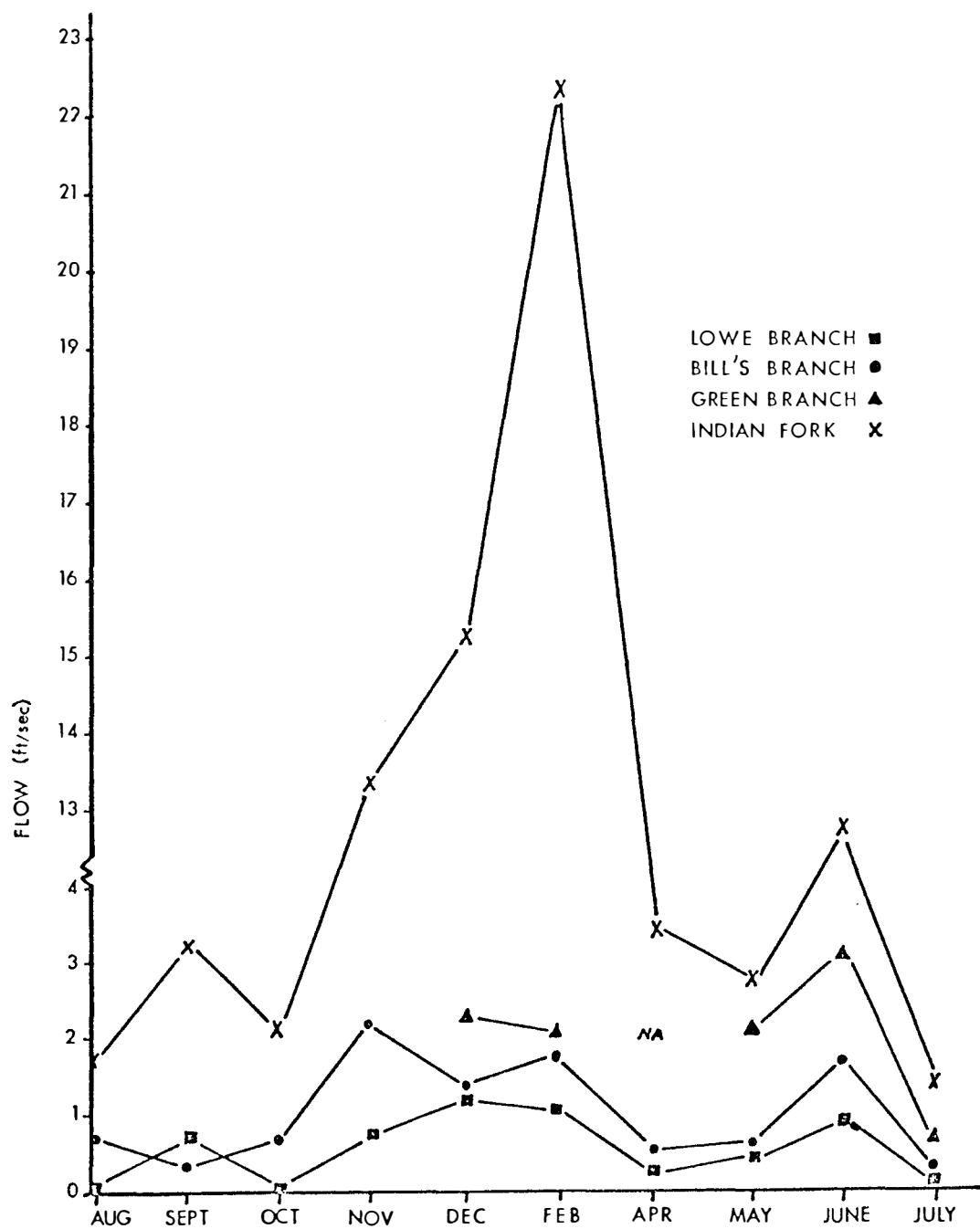


Figure 6. Monthly stream flow values for the four study streams.

streams. The monthly values for turbidity (suspended solids) and rainfall are shown in Figures 7 and 8. Turbidity, as a measurement of suspended solids, was used to indicate differences in siltation in the four streams. Litter, allochthonous material carried into the stream, was a measurement of food available in the streams.

Biological and Physical Results

After determining by use of analysis of variance and Student-Newman-Keuls means separation tests that there were significant differences among streams, the next step was to determine what factors caused these differences in taxa, number of individuals and species diversities among streams. From discriminant analysis (Nei et al., 1976; Tatsuoka, 1971) where both independent (physical variables) and dependent (biological variables) are entered simultaneously into the analysis, stream differences were again found to be significant. Streams were separated in the same manner as by Student-Newman-Keuls means separation tests for total taxa and species diversity (Table 5, p. 51). Lowe Branch and Indian Fork were separated as distinct groups which were more different from each other than from the other two disturbed streams. The group centroids (means) for Bill's Branch and Green Branch were shown by the analysis to be the same (Figure 9). However, from the F-tests (Figure 10) all groups were shown to be significantly different.

In this analysis variables were chosen that minimized Wilk's lambda, a measure of group discrimination that considers differences between group centroids and cohesion within the groups, and maximized F among groups (Nei et al., 1976). Of the fifty-four variables entered into the

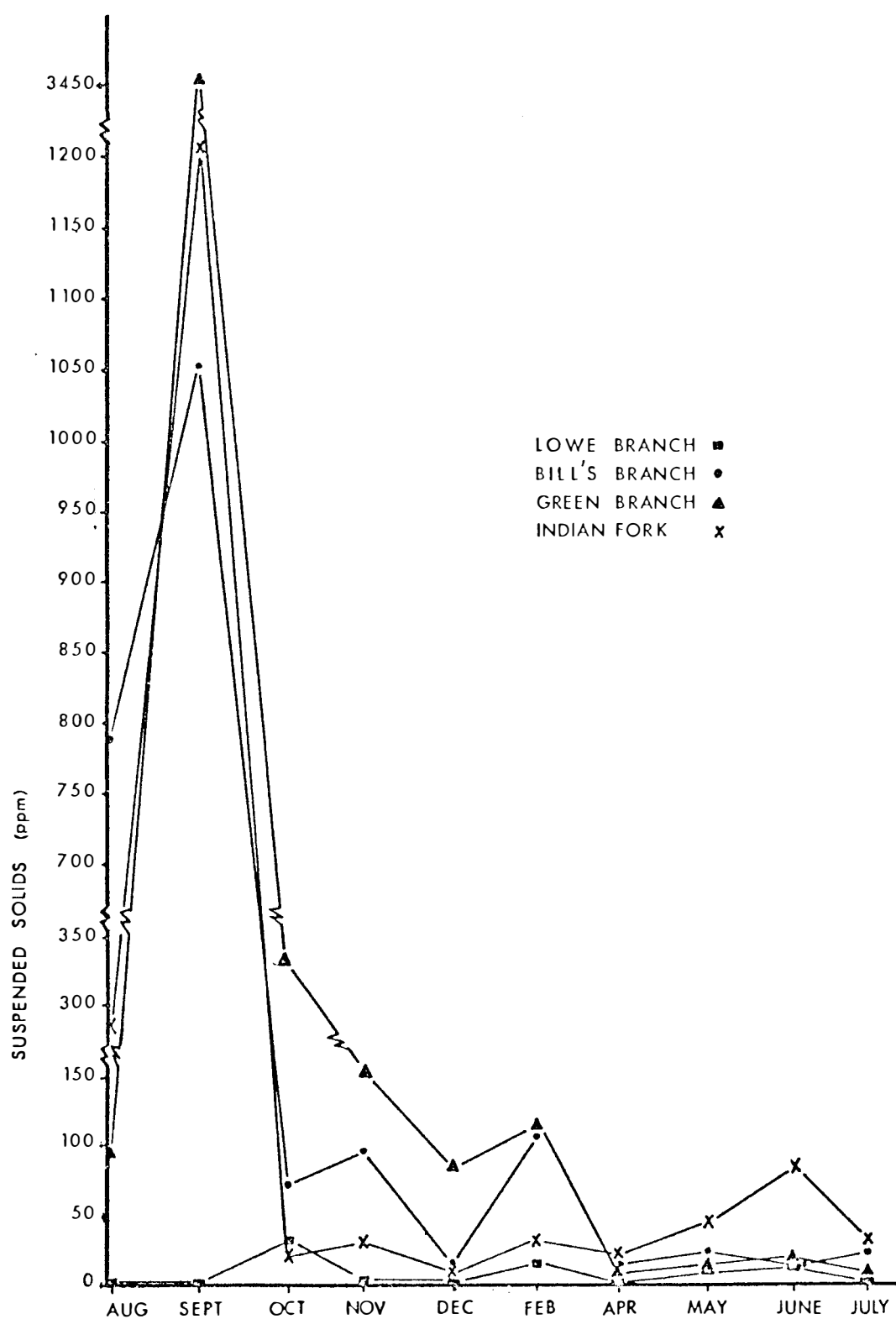


Figure 7. Differences in suspended solids for the four study streams.

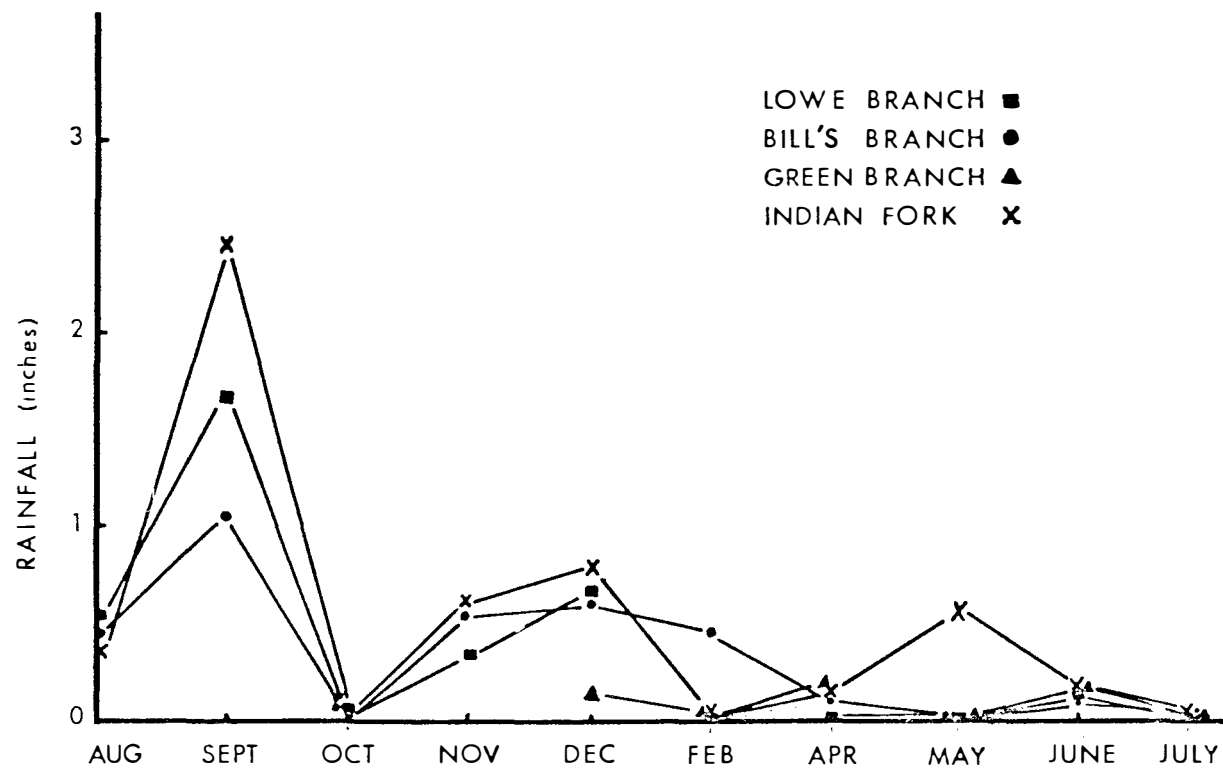


Figure 8. Five day averages for rainfall preceding sampling dates.

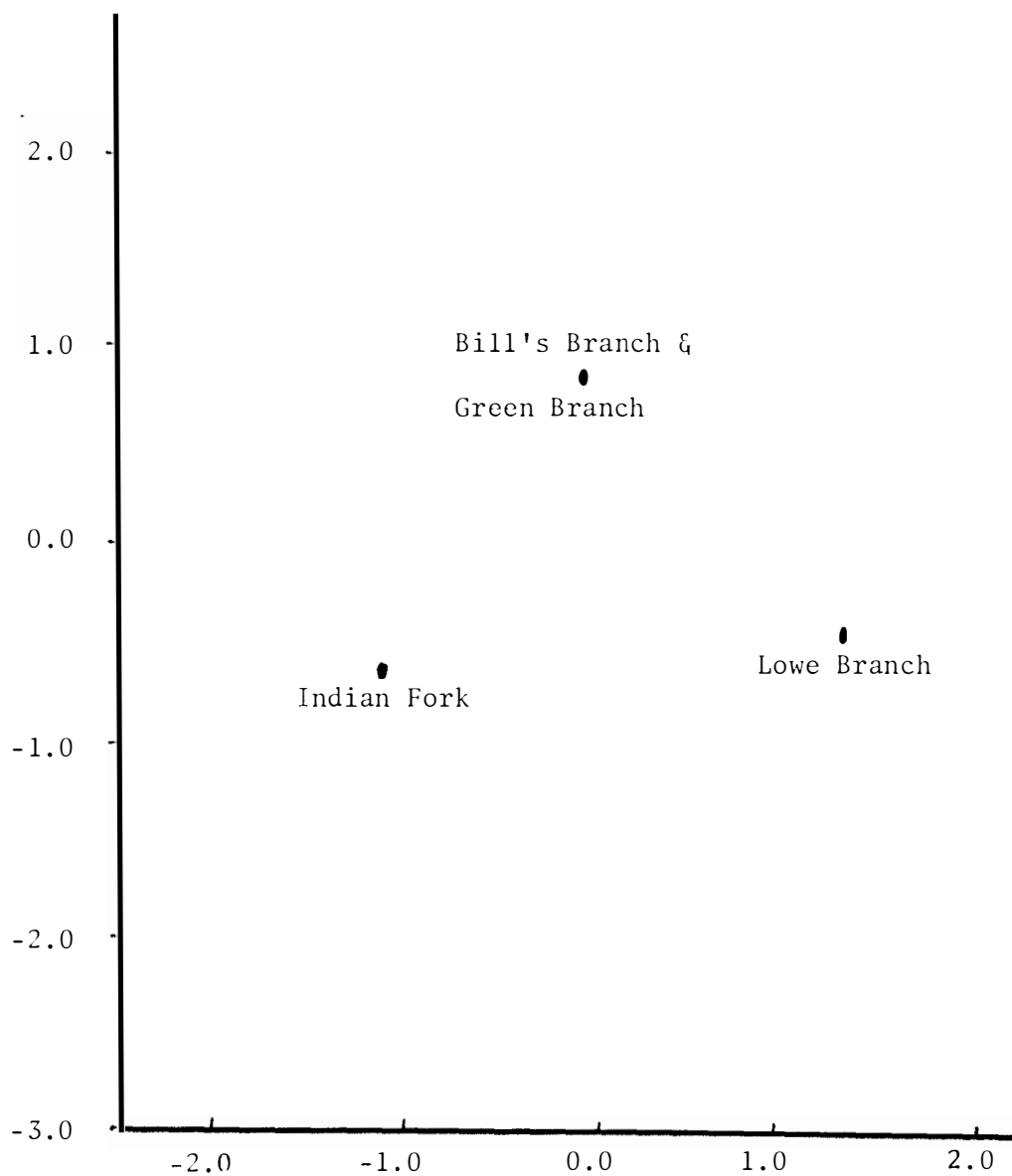


Figure 9. Group centroids (group means) from discriminant analysis of the four streams.

	Indian Fork	Bill's Branch	Green Branch
Bill's Branch	9.86176*		
Green Branch	10.21111*	3.31159*	
Lowe Branch	20.98024*	10.82376*	11.29271*

Degrees of freedom 28, 281.

Figure 10. Summary of discriminant analysis F-matrix for significant differences among the four streams. An * indicated significant differences between streams.

analysis, only twenty-eight were chosen as adding significant information (Table 9). Of the first ten variables chosen, seven were species that were found distributed fairly evenly across sampling dates in the control stream and as scattered individuals or absent from the disturbed streams. The three environmental parameters chosen as discriminating among streams summarized disturbance as a result of sedimentation and rainfall. Turbidity and stream flow provided the best environmental explanation for the differences among streams.

Of the seven taxa explaining the greatest differences among the four streams, three, Heptagenia (flavescens group), Paraleptophlebia sp., and Dolophilodes distincta were found only from Lowe Branch, the control stream. Only Heptagenia (flavescens group) and Paraleptophlebia sp. had distributions such that analysis of variance could determine differences among streams (Table 10). The number of individuals of both Heptagenia (flavescens group) and Paraleptophlebia sp. were significantly different in Lowe Branch than in the three disturbed streams. Dolophilodes distincta occurred in very low numbers from only two sampling dates in Lowe Branch. Of the remaining seven most discriminating taxa that were found in the control stream and at least one of the disturbed streams, Ephemerella sp., and Leuctra sibleyi Claassen were significantly different among streams (Table 10). In Lowe Branch and Bill's Branch the numbers of Ephemerella sp. were not significantly different. However, there was a significant difference between these two streams and the other two disturbed streams (Table 10). For Leuctra sibleyi Claassen the number of individuals from Lowe Branch was significantly different from the three disturbed streams.

Table 9. Summary of Discrimination Variables in Order of Selection for Determining Stream Differences.

Variable	Variable Name	Wilk's Lambda	F to Enter
X8	Turbidity	.72352	39.23304
X3	Stream Flow	.43664	67.23414
Y2	<u>Ephemerella</u> sp.	.37597	16.45942
Y15	<u>Leuctra sibleyi</u>	.32262	16.81220
X2	Rainfall	.28839	12.02862
Y7	<u>Heptagenia (flavescens gr.)</u>	.26357	9.50801
Y1	<u>Baetis</u> spp.	.24825	6.21383
Y4	<u>Paraleptophlebia</u> sp.	.23590	5.25473
Y27	<u>Dolophilodes distincta</u>	.22338	5.60190
Y33	<u>Simulium</u> sp.	.21257	5.07196
X6	Litter	.20281	4.77767
X4	Temperature	.19174	5.71800
Y14	<u>Peltoperla</u> sp.	.18352	4.41732
Y25	<u>Hydropsyche</u> spp.	.17651	3.90817
Y8	<u>Heptagenia (maculipennis gr.)</u>	.17008	3.70418
Y5	<u>Pseudocloeon</u> sp.	.16482	3.11555
Y42	<u>Psephenus herricki</u>	.15961	3.17842
Y32	<u>Tanypodinae</u>	.15507	2.83690
Y20	<u>Nemoura nigritta</u>	.15103	2.58929
Y31	<u>Chironominae</u>	.14736	2.39803
Y46	<u>Veliidae</u>	.14394	2.27867
Y10	<u>S. pulchellum</u>	.14107	1.94398
Y22	<u>Tanypteryx metaqui</u>	.13860	1.70170
Y45	<u>Corydalus</u> sp.	.13647	1.48082
Y41	<u>Stenemelis</u> sp.	.13471	1.23610
Y21	<u>Allocapnia</u> sp.	.13328	1.01354
Y29	<u>Tipula</u> sp.	.13172	1.11215
Y19	<u>Sweltsa mediana/onkes</u>	.13030	1.02459

Table 10. Summary of Analysis of Variance and Student-Newman-Keuls Means Separation Test for the First Seven Taxa Selected by Discriminant Analysis. Two Streams Underlined by a Single Line Indicate No Significant Differences Among Streams.

Taxa	F-test	P	Streams			
<u>Ephemerella</u> sp.	3.766	.05	<u>LB</u>	<u>BB</u>	<u>GB</u>	<u>IF</u>
<u>Leuctra sibleyi</u>	18.380	.001	<u>LB</u>	<u>BB</u>	<u>GB</u>	<u>IF</u>
<u>Heptagenia</u> (<u>flavescens</u> gr.)	6.98	.01	<u>LB</u>	<u>BB</u>	<u>GB</u>	<u>IF</u>
<u>Baetis</u> spp.	2.78	.05 ¹				
<u>Paraleptophlebia</u> sp.	5.57	.001	<u>LB</u>	<u>BB</u>	<u>GB</u>	<u>IF</u>
<u>Dolophilodes distincta</u>	.97	.05 ¹				
<u>Simulium</u> sp.	2.62	.05 ¹				

¹Nonsignificant

One step in discriminant analysis looks at the groups (streams) and determines if the samples within each group are correctly classified or whether some are more like another group. The results of this part of the analysis showed the number and percent of the samples correctly classified and the number and percent of the samples that were more like another group. The groups to which misclassified samples should belong were given as part of the analysis. Based on twenty-eight variables chosen as contributing significant information (Table 9, p. 65), samples taken from the four streams were correctly classified 73.08% of the time (Table 11). Samples from Lowe Branch were correctly classified as belonging to that group 71.3% of the time. All incorrectly classified samples from Lowe Branch belonged to Bill's Branch (15%) or Green Branch (13.8%). Samples from Bill's Branch were correctly classified 77.5% of the time with most of the remaining samples being classified into Green Branch (20%) and only 2.5% as belonging to the control stream. Green Branch samples were correctly classified 86.3% of the time (Table 11). Of the remaining samples, 10% were more like Bill's Branch and 3.8% were more similar to Lowe Branch. Samples from Indian Fork were correctly classified as belonging to that group 55.6% of the time (Table 11). All incorrectly classified samples belonged to Green Branch (27.8%) or Bill's Branch (16.7%). None of the incorrectly classified samples from this disturbed stream were classified as more similar to Lowe Branch, the control stream (Table 11). Of the four streams the largest percent of samples classified into the stream from which they were taken were from Green Branch (86.3%), Bill's Branch (77.5%), and Lowe Branch (71.3%,

Table 11. Stream Classification Resulting from Discriminant Analysis. The Number of Observations Correctly Classified for Each Stream and the Percent of the Observations Correctly Classified Are Given.

Stream	# of Cases	Predicted Stream Membership			
		Indian Fork	Bill's Branch	Green Branch	Lowe Branch
Lowe Branch	72	40 55.6%	12 16.17%	20 27.8%	0 0.0%
Bill's Branch	80	0 0.0%	62 77.5%	16 20.0%	2 2.5%
Green Branch	80	0 0.0%	8 10.0%	69 86.3%	3 3.8%
Indian Fork	80	0 0.0%	12 15.0%	11 13.8%	57 71.3%

Percent of stream observations correctly classified - 73.08%.

Table 10, p. 66). From Green Branch and Bill's Branch, the samples indicated as being incorrectly classified were primarily classified into one or the other of the two streams (Table 11).

From the discriminant analysis, discriminant functions were produced that explained the difference in the groups in N dimensional space. From this analysis, three discriminant functions were calculated that best separated the four streams (Figure 11). On the X-axis, the four streams were separated primarily by differences in turbidity (an estimation of suspended solids). The streams were separated on the Y-axis by differences in stream flow and on the Z-axis by the differences in the recorded rainfall for five days prior to each sampling date. The differences in these three physical parameters for the four streams are shown in Figures 6, 7, and 8 (pp. 58, 60, and 61 respectively).

In summary the results of discriminant analysis showed that the streams were separated on the basis of three discriminant functions (Figure 11). The factor that best explained stream differences on the X-axis was turbidity, on the Y-axis stream flow explained the greatest difference, and on the Z-axis rainfall best explained the differences in the four streams. From the stream classification (Table 11) 73.08% of the samples were classified correctly and analysis showed that those streams incorrectly classified were most similar to the next most closely classified stream.

Velocity Tolerance

From studies to determine velocities at which individuals are dislodged, it was found that the mean velocity at which Ephemerella sp.

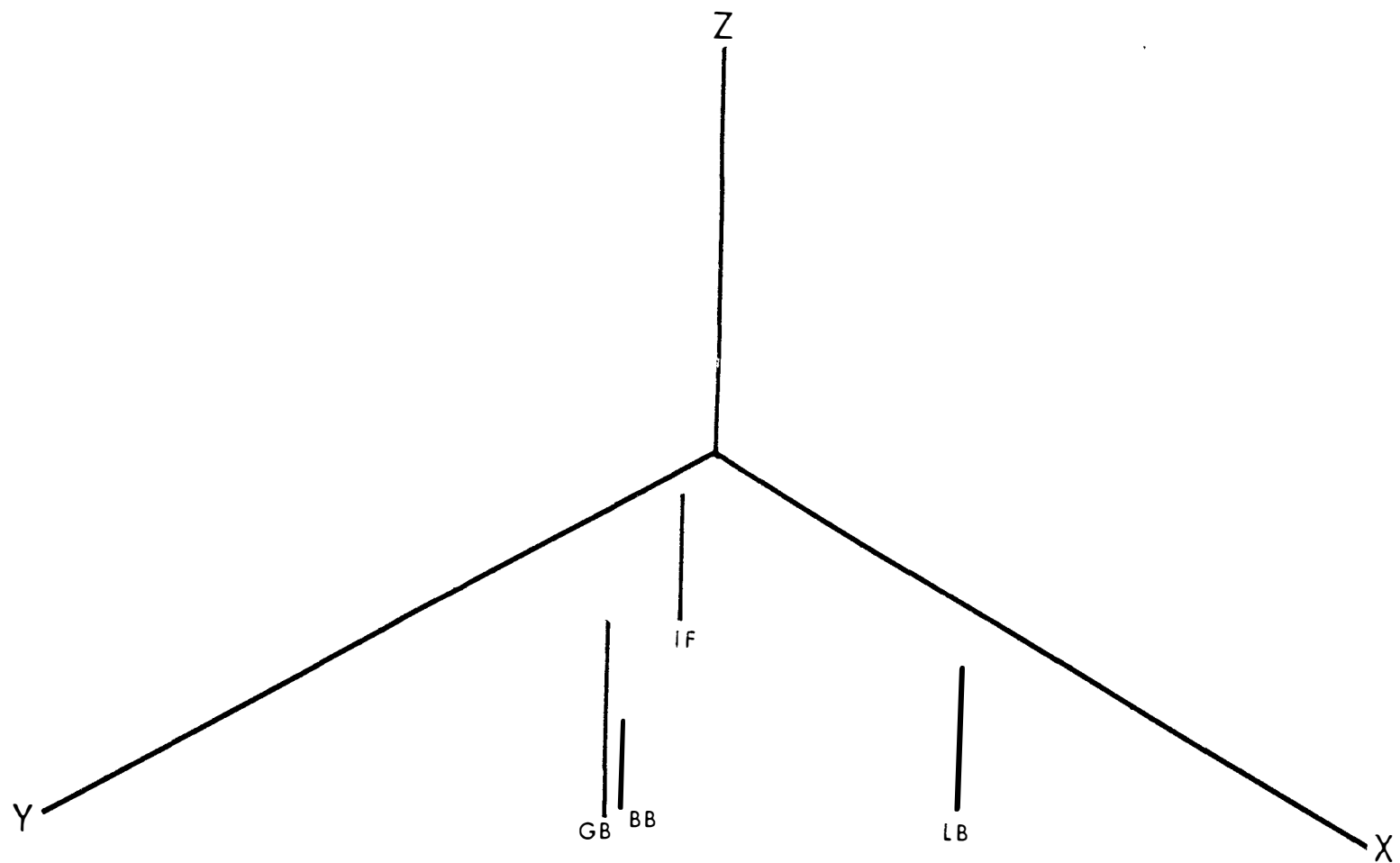


Figure 11. Standardized discriminant function coefficients showing the location of group centroids based on the discriminating functions. X-axis = turbidity, Y-axis = stream flow, and z-axis = rainfall.

were dislodged was 111.08 cm/s and that the range of tolerance was 30-130 cm/s (Table 12). Baetis was found to be dislodged by velocities ranging from 70-80 cm/s while Isoperla was found to withstand a mean velocity of 106.25 cm/s. Stenonema and Isonychia had similar velocity tolerances (118.00 and 120.00 cm/s, Table 12). The mean velocity necessary to dislodge Simulium individuals was 106.15 cm/s with a range of 80-130 cm/s, and the mean velocity that dislodged Nemoura was 106.67 cm/s (Table 12).

Heavy Metals

Tissue samples from as many insect genera as could be reasonably obtained were analyzed for metal content of Pb, Zn, and Fe. These metals were chosen since from the water quality section of the total project, these metals were found in increased levels in the disturbed streams. Lead was measured in parts per billion (ppb) using furnace spectrophotometry since levels were so low that detection using flame spectrophotometry could not be adequately carried out. Both Zn and Fe were analyzed as parts per million (ppm) using the flame spectrophotometry. Since these methods take multiple measurements of metal content and give an average measurement of the metal content in the body tissue, statistical analysis of the results could not be performed.

Individual taxa analyzed are shown in Table 13 and the metal content (ppb or ppm) for each of the three metals analyzed are shown for each of the four streams. The weight of the individual taxa analyzed and the metal content for each taxa are shown in the Appendix. From analysis of the insect tissues, it was found that the levels of metals

Table 12. Results of Laboratory Analysis to Determine at What Stream Velocity Taxa of Benthic Insects Are Dislodged from the Substrate.

Taxa	N	# Dislodged	$\bar{x}$ cm/s	Std. Error	Range	SE/ $\bar{x}$ (%)
<u>Empemarella</u> sp.	45	37	111.08	4.00	30-130	3.6
<u>Baetis</u> spp.	8	4	75.00	2.87	70-80	3.8
<u>Isoperla</u> sp.	8	8	106.25	7.30	70-130	6.9
<u>Stenonema</u> sp.	5	5	118.00	3.74	110-130	3.2
<u>Isonychia</u> sp.	4	3	120.00	10.00	100-130	8.3
<u>Simuliwn</u> sp.	15	13	106.15	5.83	80-130	5.5
<u>Nemoura</u> sp.	3	3	106.67	3.33	100-110	3.1

Table 13. Heavy Metal Content from Various Taxa Analyzed from the Four Study Streams.

Metal	Taxa	Lowe Br.	Bill's Br.	Green Br.	Indian Fork
Pb	<u>Isonychia</u> sp.	24.9615	4.5205	.4326	NA ¹
	<u>Isoperla clio</u>	15.4520	34.5979	29.2925	NA
	<u>Diplectrona modesta</u>	2.9286	2.4547	5.0471	.2553
	<u>Baetis</u> spp.	.6368	4.2694	.2079	.7860
	<u>Epeorus</u> sp.	53.5955	7.0677	NA	NA
	<u>Neophylax</u> sp.	3.0984	NA	123.5395	NA
	<u>Psephenus herricki</u>	4.7328	.3184	NA	NA
	<u>Tipula</u> sp.	7.9167	46.3990	65.9552	30.5572
	<u>Stenonema</u> sp.	NA	4.5205	NA	3.4592
	<u>Heptagenia</u> sp.	NA	.1388	NA	.2753
	<u>Leuctra</u> sp.	NA	.6799	NA	.6799
	<u>Allocapnia</u> sp.	NA	.4671	NA	.3615
Zn	<u>Isonychia</u> sp.	2.2670	.9053	.2671	NA
	<u>Isoperla clio</u>	4.7776	24.3070	7.1605	NA
	<u>Diplectrona modesta</u>	.2245	.1819	.3096	.0968
	<u>Baetis</u> spp.	.1819	1.6713	.1819	.0543
	<u>Epeorus</u> sp.	4.5648	1.1181	NA	NA
	<u>Neophylax</u> sp.	.4373	NA	4.8627	NA
	<u>Psephenus herricki</u>	.3947	.0543	NA	NA
	<u>Tipula</u> sp.	1.5011	2.9053	3.5436	2.9904
	<u>Stenonema</u> sp.	NA	1.0330	NA	.2245
	<u>Heptagenia</u> sp.	NA	1.6287	NA	.3947
	<u>Leuctra</u> sp.	NA	.0968	NA	.0968
	<u>Allocapnia</u> sp.	NA	.2671	NA	.1819
Fe	<u>Isonychia</u> sp.	45.0986	16.8187	19.3896	
	<u>Isoperla clio</u>	24.5314	42.5277	52.8113	NA
	<u>Diplectrona modesta</u>	10.3914	10.3914	16.8187	21.9605
	<u>Baetis</u> spp.	12.9623	11.6768	10.3914	9.1059
	<u>Epeorus</u> sp.	78.5204	23.2459	NA	NA
	<u>Neophylax</u> sp.	11.6768	NA	18.1041	NA
	<u>Psephenus herricki</u>	21.9605	10.3914	NA	NA
	<u>Tipula</u> sp.	34.8150	73.3786	245.6293	140.222
	<u>Stenonema</u> sp.	NA	15.5332	NA	11.6768
	<u>Heptagenia</u> sp.	NA	19.3896	NA	11.6768
	<u>Leuctra</u> sp.	NA	7.8205	NA	5.2496
	<u>Allocapnia</u> sp.	NA	7.8205	NA	9.1059

¹NA - not adequate numbers for analysis, or no individuals obtained.

in tissue of the same taxa differed either very little among streams or in some cases were higher in the control stream than in the disturbed streams. Therefore, the effects of heavy metal accumulation in insect tissues were determined to be of little importance in determining differences in benthic communities among the four streams.

CHAPTER VI

DISCUSSION

The results of this study to determine the effects of strip mining related disturbance on benthic insect communities showed that there are definite differences between disturbed and undisturbed streams. There were significant differences among streams for the number of taxa, number of individuals, and species diversity. The major factors found to influence benthic communities in streams draining strip mined watersheds were variable stream flow, rainfall, and turbidity. In these study streams, acid mine drainage, except for ferric hydroxide accumulations in Indian Fork resulting from auger holes and deep mines, was not found to be significant. In fact, in those streams disturbed by strip mining, pH was found to increase following mining disturbance (Table 8, p. 57). Other chemical pollutants, particularly heavy metals, were not shown to be important factors in determining stream differences, since there were no clear patterns in metal accumulation between the control and disturbed streams.

In order to understand the effects of the factors found to be important in discerning stream differences, it seems necessary to first consider each factor individually before considering any effect on the benthic communities. In order to understand the differences in the benthic communities among streams, one needs to examine the dynamic and very dramatic changes that occur in stream flow, stream volume, streambed scouring, and siltation which occur during storm water flow periods.

During periods of heavy rainfall, the rate of runoff from watersheds disturbed by strip mining is accelerated as a result of reduced vegetation on steep areas. In Green Branch, for example, where 24% of the area has been disturbed by mining, little vegetational recovery has occurred. In this watershed, spoil banks are in such close proximity to the stream margins that runoff reaches the stream soon after rainfall. When large volumes of runoff reach the stream soon after rainfall occurs, large amounts of sediment are carried into the stream and carried along by the increased stream flow. Under storm conditions the suspended solids load for Green Branch has been modeled to be 10,500 lb/acre (Tschantz and Overton, 1978). In contrast, the suspended solids load for Lowe Branch, the control stream, was calculated at 0 to 4.32 lb/acre. Loads for Bill's Branch and Indian Fork were determined to be 580 and 586 lb/acre respectively.

From simulated hydrographs (storm flow-cfs) and loadographs (tons of sediment) for Lowe Branch with modeled 24% disturbance, peak storm flow was calculated to increase 15% and watershed runoff to increase 40%. In addition, the sedimentation loss would change from 4 tons/acre to 1500 tons/acre for one storm period. The storm flow would increase from 14 cfs to 22 cfs with 24% disturbance (Tschantz and Overton, 1978).

With mining disturbance, the time to flow peak following rainfall decreases one to two hours. In Green Branch, peak stream flow occurs 5.5 hours after rainfall initiation as compared to 7.6 hours for Lowe Branch. Disturbed streams may increase rapidly from 1-3 meters wide and 10-15 cm deep with a moderate velocity to a stream 10 meters in width

and 3 or more meters deep with a velocity of as great as 600 cm/s. When this occurs boulders as large as 4-5 meters in diameter may be dislodged from the streambed and carried considerable distances downstream. Following storm flow, it is not unusual to find boulders 1 meter in diameter moved >100 m downstream and areas which were previously deep pools (50 cm) changed to areas with depths of 5-10 cm. Following heavy rainfall, the stream channel in the disturbed streams may be completely altered, often moving from the right to left bank or vice versa. As the flow decreases following these storms, suspended particles carried by the streams are deposited on the streambed and accumulate in areas of slower flow.

With periods of heavy rainfall and resulting increase in stream velocity, particles which are dislodged or carried in the streams scour the streambed. As the streambed is scoured, so are the benthic organisms living in or on the substrate. With the scouring of the streambed by the increased stream flow and suspended particulate matter, the streambed may be removed so that often the only substrate remaining is tightly cemented together by clay. As a result, benthic organisms that drift into the scoured area from headwater regions of the stream are restricted to the substrate deposited as stream flow decreases.

As mentioned above, as a result of discriminant analysis to determine which factors, both biological and physical, determine stream differences, the three factors measured that explain the greatest differences in the four study streams were turbidity, rainfall, and stream flow. Turbidity was used as a measure of suspended solids, since

turbidity increased as the amount of suspended solids carried in the water column increased. These three factors are closely related in their influence on benthic communities in the four streams.

Mortality

As a result of increased drift in response to stream perturbation, insects are susceptible to mortality from substrate abrasion and from movement of particles >2.5 cm. Observations of substrate particles and individual organisms showed several of the larger individuals being crushed or mutilated by substrate movement.

Litter

Another physical factor found to be important in determining differences among streams and the benthic communities of the streams was allochthonous litter accumulation. Since all four streams had vegetation along their margins, it was assumed that all streams had equal opportunities for litter input. Also, since similar running water habitats were sampled in each stream, there should have been similar amounts of litter accumulated in each stream. However, from determination of the percent litter composition of each sample, it was found that there was a significantly greater amount of litter present in the control stream. The difference in the amount of litter in the control vs the disturbed streams appears to be the result of differences in runoff and stream flow. From observations of litter and substrate particles in conjunction with the laboratory velocity tolerance studies, it was observed that litter was dislodged from the substrate at 70-80 cm/s. Allochthonous materials have been shown by numerous workers (Minshall, 1967; Wallace

et al., 1970; Egglshaw, 1964; Nelson and Scott, 1962; and Rabini and Minshall, 1977) to be the primary source of food and energy for benthic organisms and to be an important factor in determining the distribution of benthic individuals. As a result, litter coupled with moderate runoff from rainfall, moderate stream flow, and a low sedimentation rate, are important factors in providing optimal habitat requirements in the control stream. In the disturbed streams the amount of litter available as a food source ranged from 18.39 to 37.21% as compared to 55.43% litter in the control stream.

Change in Species Composition

Tolbert and Vaughn (1978) and Tolbert (1978) found that the factors mentioned above, rainfall runoff, stream flow, sedimentation, and litter are the most important factors in determining differences in the benthic communities of the four streams. Stream velocity interacts with all other factors in determining differences in the benthic communities of the four streams. In the streams in this study area, the effects of substrate particle distribution, silt deposition, and transport and shredding of leaf detritus, are magnified by the rapid changes in rainfall runoff and resultant increase in stream volume, which at times increases by several orders of magnitude.

Recolonization

As long as the disturbed streams are subjected to periods of increased runoff and increased stream flow that dislodges substrate materials and carries erosional materials from the surrounding watershed, recovery of insect communities will occur slowly if at all. In those streams continually perturbed by storm disturbance factors, the major

fauna will be those taxa that have short life cycles or are adapted to withstand disturbance. In Indian Fork, the predominant insect fauna is comprised primarily of individuals of the family Chironomidae. Usually recolonization occurs from headwater reaches of streams or from relic populations surviving in the streams. However, in these disturbed streams receiving several major rainfall related disturbances per spring and fall season, recovery is slow since recovering communities are frequently redislodged by storm water flow. Talak (1977), also in the New River basin, found that stream insect recovery generally requires as much as 20 years and that from his study the faunas of some streams showed no indications of recovery since in some instances there were no close sources of benthic faunas for repopulation.

Taxa and Individuals

From this study, both the number of species and the total number of individuals from the disturbed streams were significantly less than the control stream. In Green Branch, the largest proportion of individuals were Chironominae. These individuals were found primarily in the upper reaches of the stream which has been disturbed by only a single mining cut and has less silt accumulation than the lower reaches of the stream. This could account for fewer individuals in the downstream section which has been disturbed by two mining cuts and is in close proximity to an unreclaimed spoil bank. Indian Fork has been disturbed to the extent that only chironomids were collected to any extent (31 individuals). These individuals comprise nearly half the total number of individuals collected from this stream. Both seasonally and for the total sampling

period, the diversities of the three disturbed streams were significantly less than that of the control stream.

The predominance of members of the order Ephemeroptera in Lowe Branch was not surprising, since members of this order are abundant in shallow, flowing streams where they are found clinging to the undersides of rocks or in debris lodged in the stream. Individuals of Baetis spp., Stenonema annexum Traver, and Heptagenia (flavescens group) are particularly suited to such areas since they are herbivorous and these areas can provide adequate food to support two generations per year. Stenonema annexum requires clean water streams for survival (Lewis, 1974) and was found only from Lowe Branch.

The large percentage of plecopterans in Lowe Branch is the result of several factors. Most stonefly nymphs are found on rocks or between the gravel and rocks of the streambed (Roback, 1974). In Lowe Branch, there is little if any silt accumulation among the bottom rubble, so there are many habitats available for occupancy. The greater abundance of Leuctra spp. and Allocaenia sp. in this stream is the result of unimpeded water movement carrying dissolved oxygen through the streambed gravel. In the disturbed streams the interstices between gravel are clogged by silt which reduces water movement and oxygen transport. In Indian Fork, Nemoura nigritta nymphs have been found covered with silt. These nymphs live under rocks and in gravel areas where silt is trapped, and their notable pubescence also tends to trap silt particles.

A moderate current that does not dislodge litter fragments that fall into the stream allows food for many plecopterans to remain, either

as detritus or as a food source for herbivorous individuals which then become food sources for carnivorous Isoperla and Acroneuria. These individuals feed on chironomids, simuliids, and mayflies. Tipulids, as well as other dipteran larvae which are found in leaf packs, feed on decaying leaves, wood, algae, and diatoms (Robach 1974), and are abundant in this stream. Chironomids feed on diatoms which were found to be more abundant and to have more species in Lowe Branch than in the disturbed streams (Vaughn, 1978). These individuals, as well as simuliids, are "vacuum" feeders that take anything of reasonable size into the digestive track (Roback, 1974). These dipterans were found in greatest numbers in Lowe Branch and from the upper sampling site on Green Branch since these were the only locations that provided the necessary unsilted substrate for optimum feeding and for simuliid attachment. Case building and retreat building Trichoptera were most abundant in the control stream because the rocks were free of silt deposits which would interfere with case attachment and food gathering. Scott (1958) found Glossosoma sp. only in areas where silt did not occur since silt interfered with their food gathering mechanisms. Hydropsyche larvae were reduced in numbers in Bill's Branch and absent from the other two disturbed streams (Table 3), probably as a result of silt and sand interfering with or clogging their food gathering nets. Since there are abundant leaf packs, diatoms, and allochthonous material available as food, as well as numerous herbivorous, detritivorous, and carnivorous benthic insect species present, there is adequate food being both produced and broken down in Lowe Branch to be used by a great variety of benthic species.

Bill's Branch had 69% as many species and 37% as many individuals relative to Lowe Branch, the control stream. Of the individuals present, 66% belonged to the order Ephemeroptera (Table 5, p. 51). In this order, Baetis spp. comprised better than half of those individuals found. These species live on or under stones and rocks, in riffles, or among debris and move frequently from place to place. They feed on small particles of plants and detritus on the surface of rocks and leaves (Edmunds, et al., 1976). It is probable, since these individuals move about frequently, that the silt and algal cover on the rocks in this stream and Green Branch, trap enough food particles to support them. Another possible explanation for the large number of Baetis spp. in this stream may be the result of reduced competition as a result of other species being eliminated (Hynes, 1970). Isonychia sp. was found only in Bill's Branch where individuals live in debris or leaves caught in areas of swift flow. These organisms face into the current and filter food from the flowing water (Berner, 1950). Since they possess strong fore claws, they are well adapted to clinging tenaciously to the substrate and can withstand all but the heaviest flows. Individuals of Heptagenia (maculipennis group), Stenonema pulchellum (Walsh) and Epeorus sp. make up the majority of the remaining individuals. All three species are flattened dorso-ventrally and hold tenaciously to the under surfaces of rocks (Edmunds, et al., 1976; Berner, 1959). These characteristics make them difficult to dislodge from the substrate. Diptera, comprised primarily of Chironominae (Table 3, p. 46) was the second most abundant order in this stream (Table 4, p. 48). Since these are primarily "vacuum" feeders on algae, particularly diatoms, as well as organic debris in the

water, they could obtain adequate food from particles washed into and carried in the stream.

Green Branch had slightly less than half the number of individuals when compared with Bill's Branch as well as fewer species (Figure 5, p. 50). More than half (53.6%) of the individuals present in Green Branch were dipterans of which chironomids were most abundant. Most of the individuals of the subfamily Chironominae were found in December and August indicating probably two generations per year (Roback, 1974), one that overwinters and emerges in early spring, and the other that develops during the summer and emerges in the fall. Three genera of Plecoptera, Peltoperla, Isoperla, and Allocaonia (Table 3, p. 46), comprised the majority of the second most abundant order (17.8%). All three genera were found in greatest numbers at the upper sampling site, which was below a single mining cut. Diplectrona modesta and Diplectrona metaqui Banks (Trichoptera) occurred in greatest numbers at this site. The greater abundance of this species, as well as Rhyacophila sp. and the plecopteran genera mentioned above at this upper site, was most probably the result of less silt accumulation, moderate stream flow, and resultant greater food availability, when compared to increased stream flow as well as larger stream size at the downstream sampling site. In addition, more individuals were available as food sources for Isoperla clio and Rhyacophila sp. at the upper site. Large boulders and dense vegetation on the land surface between the mine cut and the upper site seem to catch most of the gravel and silt eroding from this mined area. This

prevents most of the silt usually associated with strip mining from moving into the stream, clogging the interstices of stones and covering food sources. Downstream, the stream passes through spoil banks and over-burden near the stream margins. The close proximity of these porous earth masses to the stream margin, the large area of the watershed disturbed ($\sim 25\%$, Table 1, p. 23), and the lack of reclamation, combine to provide large amounts of silt which are washed into the stream. The close proximity of the spoil bank also reduces the distance runoff has to travel before entering the stream. As a result, rainfall runoff and resultant increased stream flow as well as increased silt loads, may occur sooner and more drastically following rainfall (Tschantz, 1977).

The number of individuals from Indian Fork is only 4% of the total number from Lowe Branch and is 21% less than the number from Green Branch (Table 5, p. 51). Dipterans are the most abundant taxa followed by the ephemeropteran genus Baetis, which is found on and under rocks and debris. The decreased number of individuals in Indian Fork is due primarily to the increased flow and sediment carried and deposited in the stream as a result of increased runoff, the reduced amount of litter available as a food source, and to some extent, the large amount of ferric hydroxide deposited on the streambed. This ferric hydroxide accumulates on the bodies of benthic organisms, possibly interfering with respiration and food gathering mechanisms.

There was a progressive decrease in the number of individuals and species from the control stream, Lowe Branch, to a moderately disturbed stream, Bill's Branch and then to a more disturbed and longer disturbed

stream, Green Branch. The further decrease in the number of individuals from Indian Fork, which has been disturbed for the longest period of time, indicates that the amount and duration of the disturbance determines to a large extent, the insect fauna present in the streams.

. Although numerous researchers have demonstrated the importance of acid mine drainage, heavy metals, and other changes in water chemistry associated with strip mining in other coal producing areas, this study clearly demonstrates the overriding influence of physical factors associated with rainfall and runoff, and resultant increased stream flow for the Cumberland Mountain region. The aquatic system in the New River Basin, unlike many which are subjected to chronic pollution from industrial sources, municipal sewage outfalls, etc., is a periodically stressed system. Within this area, the source of stress to benthic communities is physical rather than of a chemical nature.

CHAPTER VII

CONCLUSIONS

In the New River Basin, streams affected by disturbance related to strip mining were found to have reduced numbers of taxa, individuals, and species diversities when compared to an undisturbed stream. In streams disturbed by contour strip mining, the principal physical factors affecting benthic insect communities were siltation of the streambed and increased stream flow. With the significant disturbance of 8 to 24% of the area of the watersheds and the resultant removal of vegetation from steep slopes by contour strip mining, the rainfall retaining capacity of the watersheds is reduced. As a result rainfall runoff makes its way rapidly into the streams. Particularly in those streams where the spoil banks are in close proximity, mobilized sediment particles of various sizes are carried into the streams. Within less than 6 hours following rainfall initiation, individual stream size may increase from 1-3 m in width and 10 cm deep to 10 m in width and 3 cm in depth. As a result boulders as large as 4 m in diameter may be moved many meters downstream. With increased stream flow from runoff, these sediment particles as well as mobilized substrate particles and benthic organisms are dislodged and carried downstream. As a result the number of individuals and taxa and the community diversity are reduced in the disturbed streams.

It was found from the laboratory velocity studies that the increased velocities resulting from rainfall in the disturbed watersheds were

greater than the velocities found to dislodge benthic organisms and substrate particles. Laboratory and field observations showed that particles larger than 2 cm in diameter were dislodged by storm flow and that insects were frequently crushed by these moving particles. From sediment transport and storm flow studies performed in support of this project, it is obvious that both the sedimentation rate and storm water flow in the mining disturbed streams are of such magnitude as to override the influence that other physical or chemical parameters might have on benthic insects.

Acid mine drainage was not found to be a problem in this area since in all cases the pH increased in the streams with mining disturbance and resultant exposure of limestone materials to weathering. Heavy metals and other chemical pollutants did not increase sufficiently to exceed state water quality standards or exert an important influence on the benthos. Only in Indian Fork was there a ferric hydroxide accumulation, from auger holes and deep mine sources, great enough to be of any significance. With the effects of substrate alteration as a result of increased stream flow and sediment deposition, these effects were considered of little relative importance.

Water chemistry among the four streams was not appreciably different. Although there were increases in Ca, Mg, Mn, and alkalinity as a result of strip mining these elements have not been shown to have any effect on benthic insects. Temperature, oxygen content, and micro-velocity for the four streams were very similar and therefore did not exert an influence in the differences in benthic insect communities in

the four streams. Differences in the availability of litter as an allochthonous food source in the disturbed streams as compared with the control stream exerted a significant influence in determining differences in the benthic communities in the disturbed streams. However, these differences were secondary to those of rainfall, stream flow, and turbidity.

The physical factors of stream flow, rainfall, and turbidity as a measure of sedimentation were the primary factors in determining the benthic communities of the four study streams. The control stream, Lowe Branch, had consistently moderate flow with only slight elevation in stream flow as a result of rainfall runoff. Siltation in this stream was very low such that there was minimal accumulation of the substrate to interfere with the attachment of benthic individuals. The moderate flow resulted in a stable substrate and abundant litter available as food sources. This is in contrast to increased levels of siltation of the streambeds of the disturbed streams and the instability of the substrate as a result of increased storm water flow dislodging substrate particles. In the disturbed streams there was little litter available as a food source. Laboratory studies demonstrated that velocities of 70-80 cm/s removed litter accumulations, showing that in the disturbed streams storm water velocities exceed the levels necessary to dislodge litter.

The results of this study indicate that in storm water dominated systems physical perturbation are primarily responsible for disturbance of benthic communities. The results show that the differences in

benthic communities are primarily the result of the effects of rainfall runoff, increased stream flow and sedimentation. The results of this study also indicate that in acid mine drainage studies the effects of rainfall runoff, increased stream flow, and sedimentation may be at least as important in determining differences in benthic communities as the effects of low pH.

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APPENDIX

Table 14. Heavy Metal Content of Insect Tissue from Each of the Four Study Streams.

Taxa	Weight	Pb ppb	Zn ppm	Fe ppm
Lowe Branch				
1. <u>Isonychia</u> sp.	.42168	24.9615	2.2670	45.0986
2. <u>Corydalis</u> <u>cornutus</u>	.07109	2.2918	.5649	11.6768
3. <u>Hexagenia</u> sp.	.04711	3.4592	.3947	15.5332
4. <u>Diplectrona</u> sp.	.01529	2.9286	.2245	10.3914
5. <u>Isoperla</u> <u>clio</u>	.53197	15.452	4.7776	24.5314
6. <u>Baetis</u> spp.	.01022	.6368	.1819	12.9623
7. <u>Pseudocloeon</u> sp.	.03508	3.4592	.4373	14.2477
8. <u>Isoperla</u> imm.	.17529	13.3294	1.2032	27.1023
9. <u>Epeorus</u> sp.	.99821	53.5955	4.5648	78.5204
10. <u>Psephenus</u> <u>herricki</u>	.08626	4.7328	.3947	21.9605
11. <u>Neophylax</u> sp.	.05524	3.0984	.4373	11.6768
12. <u>Tipula</u> sp.	.56128	7.9167	1.5011	34.8150
Indian Fork				
13. <u>Baetis</u> spp.	.00907	.7860	.0543	9.1059
14. <u>Heptagenia</u> sp.	.03468	.2753	.3947	11.6768
15. <u>Stenonema</u> sp.	.01329	3.4592	.2245	11.6768
16. <u>Allocaonia</u> sp.	.01083	.3615	.1819	9.1059
17. <u>Leuctra</u> sp.	.01186	.6799	.0968	5.2496
18. <u>Diplectrona</u> sp.	.01642	.2553	.0968	21.9605
19. <u>Tipula</u> sp.	.32776	30.5572	2.9904	140.2222
Bill's Branch				
20. <u>Hydropsyche</u> sp.	.18900	.9050	.0968	70.3914
21. <u>Tipula</u> sp.	2.52691	46.3990	2.9053	73.3786
22. <u>Psephenus</u> <u>herricki</u>	.00829	.3184	.0543	10.3914
23. <u>Isoperla</u> <u>clio</u>	1.31580	34.5979	24.307	42.5277
24. <u>Leuctra</u> sp.	.1775	.6799	.0968	7.8205
25. <u>Ephemerella</u> sp.	.01903	.1698	.2671	7.8205
26. <u>Isonychia</u> sp.	.13700	4.5205	.9053	16.8187
27. <u>Stenonema</u> sp.	.19595	4.5205	1.0330	15.5332
28. <u>Peltoperla</u> sp.	.02225	1.5489	.3522	10.3914
29. <u>Eperous</u> sp.	.10824	7.0677	1.1181	23.2459
30. <u>Baetis</u> spp.	.11604	4.2694	1.6713	11.6768
31. <u>Heptagenia</u> sp.	.13877	3.8373	1.6287	19.3896
32. <u>Allocaonia</u> sp.	.02118	.4671	.2671	7.8205
33. <u>Diplectrona</u> sp.	.2983	2.4547	.1819	10.3914
Green Branch				
34. <u>Rhyacophila</u> sp.	.02153	1.3313	.1819	10.3914
35. <u>Diplectrona</u> sp.	.05091	5.0471	.3096	16.8187
36. <u>Tipula</u> sp.	.61577	65.9552	3.5436	245.6293
37. <u>Isoperla</u> <u>clio</u>	.95325	29.2925	7.1605	52.8113
38. <u>Baetis</u> spp.	.01577	.2079	.1819	10.3914
39. <u>Isonychia</u> sp.	.2066	.4326	.2671	19.3896
40. <u>Neophylax</u> sp.	.2837	123.5395	4.8627	18.1041

VITA

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