SPECIES DIVERSITY OF BENTHIC MACROINVERTEBRATES IN A STREAM RECEIVING DOMESTIC AND OIL REFINERY EFFLUENTS

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PREFACE

The objectives of the present study of community structure of benthic macroinvertebrates in a stream receiving domestic and oil refinery effluents were to (1) determine seasonal environmental changes; (2) investigate effects of physico-chemical conditions on benthic macroinvertebrate populations; (3) relate quantitative composition of benthic macroinvertebrate populations to various degrees of pollution; (4) apply methods derived from information theory to benthic macroinvertebrate community structure.

This investigation is part of a larger survey of Skeleton Creek. Concomitant studies include a determination of primary productivity by diurnal oxygen curve analysis, a survey of community structure of fish populations and a study of the effects of organic enrichment on morphological characteristics of fish.

Dr. Troy C. Dorris served as major advisor. Drs. Calvin G. Beames, William A. Drew, Roy W. Jones and Rudolph J. Miller served on the advisory committee and criticized the manuscript. Dr. Bernard C. Patten, Oak Ridge National Laboratory, criticized the manuscript. Dr. Robert Morrison directed writing of the computer program for species diversity calculations. Verification of invertebrate determinations were made by Drs. R. O. Brinkhurst, W. D. Day, O. S. Flint, E. J. Kormondy, H. G. Nelson, S. S. Roback, W. W. Wirth and Rev. H. B. Herrington. Ray K. Baumgardner helped make field collections. The assistance of all these

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I. INTRODUCTION

Two approaches typically are used in the study of natural communities. One approach emphasizes biomass and production and is concerned with assemblages of organisms in terms of matter and energy. The second approach emphasizes community structure and analyzes communities as complexes of individuals belonging to different species with definite ecological requirements. Community structure has been described in terms of species frequency, species per unit area, spatial distribution of individuals and numerical abundance of species (Hairston, 1959).

Community structure of benthic macroinvertebrate populations has frequently been used to evaluate conditions in streams receiving organic effluents. Bottom organisms are particularly suitable for such studies because their habitat preference and low motility cause them to be directly affected by substances which enter the environment. Chemical surveys indicate stream conditions only at the time of sampling, but benthic macroinvertebrate populations can be indicative both of present and past environmental conditions (Farrell, 1931). Butcher (1955) has shown that chemical and biological data differ widely when several types of pollution are involved and stated that pollution should be defined by biological conditions instead of chemical standards.

Different species have different ranges of tolerance to organic wastes (Richardson, 1928). A number of investigators have attempted to classify bottom organisms according to pollution tolerance (Richardson, 1928; Gaufin and Tarzwell, 1952; Schiffman, 1953; Surber, 1953; Mackenthun, Van Horn and Balch, 1956; Gaufin, 1958). Frequently, there exists an appreciable lack of agreement as to the true status of many of these organisms because of regional differences in species and environment (Gaufin, 1958). Difficulties arise in using specific organisms as indicators of pollution, because organisms that may occur in large numbers in an enriched zone may exist in smaller numbers in cleaner zones (Gaufin and Tarzwell, 1952). Conversely, some clean water forms may occur in polluted zones. Environmental conditions other than pollution also may influence the distribution of organisms (Needham, 1938).

Use of associations or populations of benthic invertebrates provides a more reliable criterion of organic enrichment than mere occurrence of a specific species (Gaufin and Tarzwell, 1956). However, such analyses usually involve long lists or descriptions of associations which are often cumbersome to use. Measures which summarize community structure clearly and briefly would be valuable in evaluating effects of organic enrichment.

Diversity indices permit summarization of large amounts of information about numbers and kinds of organisms (Patten, 1962). Such parameters express the distribution of individuals into species. According to Hairston (1959), the first important attempt to interpret animal community structure from the relationship between numbers of individuals and species was that of Fisher, Corbet and Williams (1943). Fisher concluded that the logarithmic series provided an adequate description of the data and proposed a constant, α, as an expression of diversity. Preston (1948) stated that the frequency distribution of an animal population is nearer a lognormal distribution; that is, frequency distributions of random samples of ecological assemblages approximate the form of a normal curve drawn on a logarithmic base. Williams (1953) compared the two methods and concluded that the distribution of species with different numbers of individuals is more nearly lognormal than logarithmic. Hairston and Byers (1954) attempted the analysis of populations of soil arthropods by both methods and concluded that both indices of diversity were related to sample size, which makes it difficult to compare different communities.

Linear relationship between the number of species and logarithm of the area studied was probably first noted by Gleason (1922). Margalef (1951) considered the area studied proportional to the number of individuals and used this relationship as a basis for a measure of community diversity.

Margalef (1956) proposed analysis of natural communities by methods derived from information theory. Diversity and information may be considered equal for practical purposes and can be calculated directly from the sample. Unlike many expressions for describing community structure, Margalef's index of diversity includes numbers of individuals representing each species. Maximum diversity exists if each individual belongs to a separate species, and minimum diversity exists if all individuals belong to the same species. In most communities the distribution of individuals into species lies between these extremes and diversity is intermediate. Diversity is usually expressed in bits. One bit represents the information required to specify one of two equally probable states.

Unequal abundance of individuals per species in a mixed population constitutes repetition for common species. This repetition is redundancy. According to Margalef's concept, redundancy represents the position of a community between maximum and minimum diversity extremes. Patten, Mulford and Warinner (1963) defined redundancy as a measure of the extent to which dominance (abundance) is expressed by one or more species.

Communities must transmit their structure (information) through time to perpetuate themselves. Communities generally are adapted to extant environmental conditions. Changes in environment ordinarily will be accompanied by changes in biota, and information content (diversity) and redundancy will be altered. Weaver (1949) stated that in the process of transmitting information, extraneous elements may be added. Unwanted additions or changes in the transmission constitute noise and reduce useful information. In an ecosystem changes in environmental conditions may have the effect of noise.

If excessive amounts of effluents are discharged into a stream, a high level of noise may be created. Some species may be unable to survive and others may persist in reduced numbers. With the resulting reduced competition, certain species may be able to attain great abundance. Margalef (1961) pointed out that in the event of a sudden increase of nutrients, the different species are led to take full advantage of their respective rates of increase. As such rates differ widely, the result is a decrease in diversity and one or a few species may attain great abundance. The species composing the resultant community are capable of transmitting information and the effluent would not have the effect of noise to this community. "Noise is thus purely relative to some given recipient ..." (Ashby, 1961).

Large numbers of individuals and small numbers of species ordinarily are found in enriched areas of streams receiving organic wastes. Since several species may be superabundant, a large probability exists that an individual observed during sampling belongs to a species previously recognized. Thus, considerable repetition of information exists and redundancy is high. Information per individual is low and would be reflected in a low index of diversity. Only a few samples would be necessary to describe this low-information system and additional samples would be superfluous. Downstream clean water areas are characterized by smaller numbers of individuals and larger numbers of species. There is less repetition of information per individual. Thus, information per individual is greater and redundancy is lower than in enriched areas. More samples are needed to adequately describe this system. Stream areas between these two extremes would probably have intermediate values of diversity and redundancy. Thus, information and redundancy possess features which make them useful measures of community structure in a polluted situation.

II. DESCRIPTION OF AREA

General Description

Skeleton Creek is a permanent stream located in a transition zone between the humid prairie and the subhumid plain. The stream originates near Enid, Oklahoma; flows southeasterly for 70 miles through Garfield, Kingfisher and Logan Counties; and empties into the Cimarron River 5 miles north of Guthrie, Oklahoma (Fig. 1). Stream elevation is 1,244 ft at Enid and 910 ft at the mouth, with an average gradient of 6 ft/mile (Fig. 2). Skeleton Creek and its tributaries are well defined and have carved wide shallow valleys ranging in depth from 40 to 75 ft. Small areas of steep land occur along the edges of the stream valleys where drainageways are cutting back into the plain.

The Skeleton Creek drainage basin is a mixed-grass prairie association and comprises approximately 400,000 acres. Over 80% is cultivated or open pasture and range (Soil Conservation Service, 1958). Topography of the drainage basin is flat to gently undulating, with broad divides. The flood plain area comprises 50,000 acres of which 40,000 acres are cultivated. The channel is inadequate to carry runoff from storms producing more than two inches of rainfall. Minor floods covering 20% of the watershed occur about every other year, while major floods occur approximately every 10 years. Erosion of agricultural uplands appears to have been relatively light; however, flood plain erosion has been moderately severe.

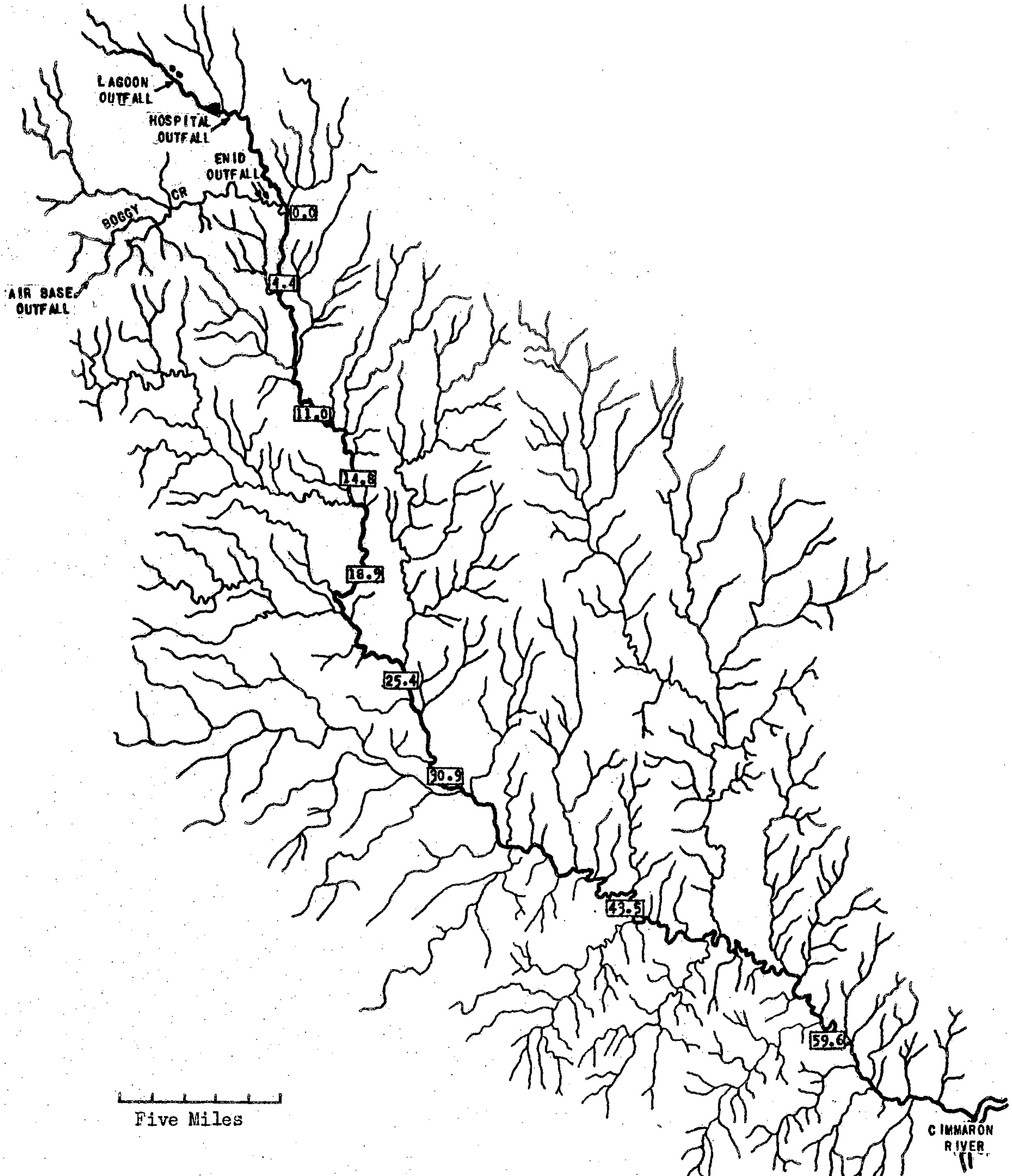

Fig. 1. Skeleton Creek drainage basin.

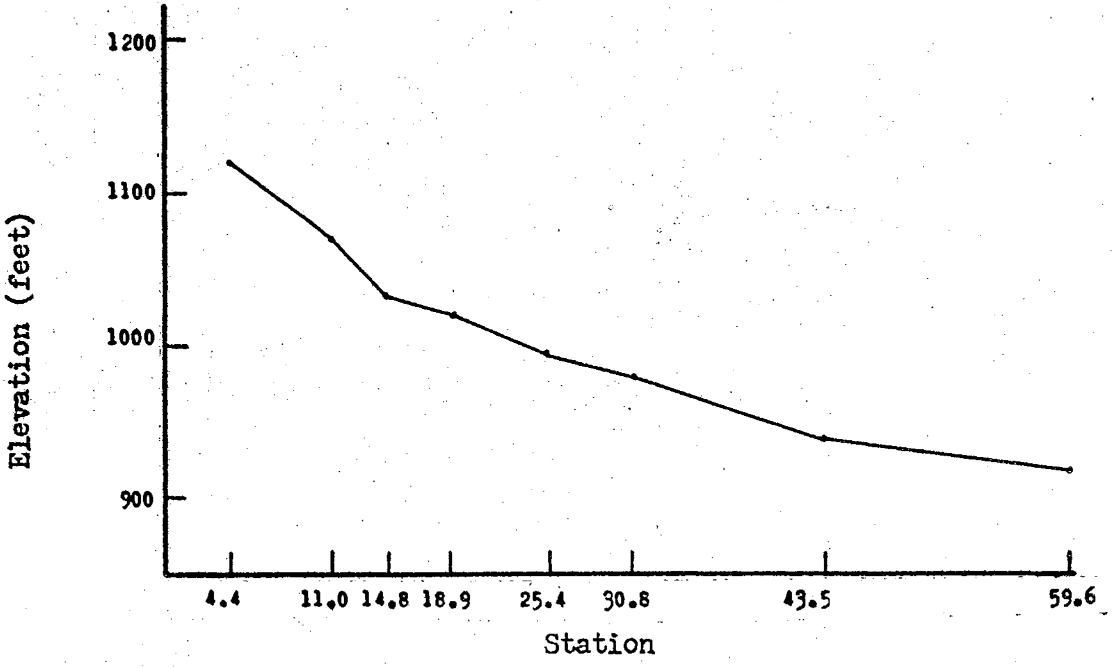

Fig. 2. Longitudinal stream profile.

According to a stream classification based on the degree of branching (Horton, 1945), Skeleton Creek passes from order 3 at the uppermost station to order 6 at the lowermost station. Stream depth varies from a few inches in riffles to more than 5 ft in a few pools. Width increases to more than 50 ft near the mouth, while mean annual flow reaches more than 50 cfs near the mouth. Riffles are abundant in the headwaters and pools are common in the lower reaches. Cover along the stream course varies from open, unshaded areas to dense, tree-lined banks.

Most of the exposed rocks in the drainage basin belong to the Enid group of the Permian Red Beds (Fitzpatrick, Boatright and Rose, 1939; Galloway, 1960). The Enid group consists of sandstones, shales and limestones, and in the upper part is characteristically crimson red. The surface formation is Quaternary in the vicinity of Enid and in narrow areas along the creek in Logan County. Most soils in the watershed belong to the Renfrow-Zaneis-Vernon association of the Red Prairie area (Gray and Galloway, 1959). Surface soils of this association are brown to reddish-brown clay, silt or sandy loams weathered from red shales, sandstones and clays. The predominant soil of the flood plains is the Port soil series, a brown to reddish-brown clay or silt loam. There is also much aeolian material along the stream.

Climate of the Skeleton Creek area is continental and characterized by wide fluctuations in temperature. Summers are warm and winters are fairly mild, with short cold periods. There is an average of 214 frost free days from 31 March to 31 October (Galloway, 1960). Rainfall averages about 32 in/year, but there are wide annual differences and severe droughts are common. Driest months are from December through February. The sun shines about 70% of the time and the average annual wind velocity is 11.6 mph.

Sources of Pollution

Both municipal and industrial wastes enter the headwaters of Skeleton Creek (Fig. 1). Domestic wastes from North Enid flow into two six-acre sewage lagoons and approximately 90,000 gal/day of effluent enter Skeleton Creek from the lagoons. Two miles below the sewage lagoons a dam across Skeleton Creek forms a small pond. Several hundred yards below the dam effluent from a hospital enters the creek. Treatment facilities at the hospital include an Imhoff tank, a trickling filter and a final settling basin. No estimate of discharge is available.

Boggy Creek, a small intermittent stream which carries little flow except effluent during most of the year, enters Skeleton Creek 4.8 miles below the hospital outfall. At least three effluent outfalls enter Boggy Creek. Domestic wastes from an air base enter Boggy Creek about

10 miles above its confluence with Skeleton Creek. The base has a biological treatment plant with a primary settling basin, a trickling filter, digestion buildings, sludge drying beds and a final settling tank. Average outfall in May, 1964, was 185,000 gal/day. Domestic wastes from Enid enter Boggy Creek 1.2 miles above its confluence with Skeleton Creek. The treatment plant contains bar screens, grit collectors, preaeration tanks, primary settling tanks, aeration tanks, digestion buildings and sludge beds. Over 4 million gal of sewage is treated per day. Approximately 1.5 million gal of this is pumped to an oil refinery for use in refining processes and the rest enters Boggy Creek.

Treatment facilities at the oil refinery include an API separator, a holding lake with a retention time of 27 days, lime settling pits, an Ozark Mahoning underwater burner and five holding ponds. Bacterial seed and aeration are introduced in the holding ponds. The effluent passes over several aeration dikes, flows 2 miles in a small ditch and enters Boggy Creek several hundred feet above the Enid sewage treatment plant outfall. Final effluent volume is 720,000 gal/day.

Description of Stations

Eight stations were selected for study and designated by numbers according to distance in miles from the confluence of Boggy Creek with Skeleton Creek. A number of differences existed among sampling stations and fluctuations in benthic composition reflected these differences as well as dilution and changes in the organic wastes.

Only a few trees occurred on banks at Stations 4.4 and 14.8, while banks at remaining stations were thickly lined with trees. Banks at all stations were moderately steep and eroded. The stream bottom was predominately fine sand and silt in marginal collecting areas and coarse sand and gravel in riffle areas. Substrate in pool sampling areas was coarse sand and rocks at Station 4.4; coarse sand at Stations 11.0, 14.8 and 18.9; and fine sand and silt at the remaining stations.

The stream in the vicinity of Stations 4.4 through 14.8 was shallow, riffles were abundant and current was relatively swift (Table I).

TABLE I

MEAN ANNUAL STATION CHARACTERISTICS

Station	Width (ft)	Depth (ft)	Current (fps)	Flow (cfs)
4.4	21.3	0.4	0.77	4.64
11.0	18.8	0.8	0.85	8.17
14.8	20.8	1.1	0.65	9.69
18.9	18.3	1.9	0.31	9.28
25.4	19.2	1.8	0.46	11.95
30.9	16.1	1.8	0.72	11.50
43.5	18.2	1.6	0.69	16.35
59.6	32.7	2.2	0.84	24.39

Stations 18.9 and 25.4 were situated in a sluggish area of the stream where current was low and riffles were scarce. Depth was similar at these two stations and greater than at upper stations. Pools predominated at the three lower stations, but current was greater than in the middle reaches. Depth was similar at Stations 30.9 and 43.5, while depth and width at Station 59.6 were greater than at any other station. A large increase in mean annual flow occurred in the stretch below Station 30.9.

III. PROCEDURES

Physico-chemical

Water samples were taken at monthly intervals from each station. Measurements were made of temperature, pH, conductivity, turbidity, phenolphthalein and methyl orange alkalinity and dissolved oxygen. Water samples were taken from approximately the same location as marginal bottom samples. Stream flow was measured at the same time.

Water temperature was measured at each station with a mercury Centigrade thermometer. Hydrogen-ion concentration was determined by use of a Hellige Comparator. Conductivity was measured with an Industrial Instruments Conductivity Bridge and turbidity with a Bausch and Lomb Spectronic 20 Colorimeter.

Phenolphthalein and methyl orange alkalinity were determined by titration with .02 N H_2SO_4 (A.P.H.A., 1960). Duplicate dissolved oxygen samples were taken at each station and fixed by the Alsterberg (Azide) modification of the Winkler method (A.P.H.A., 1960). Samples were collected in 126 ml bottles and the entire sample was titrated with .016 N sodium thiosulfate. Diurnal oxygen measurements were made at intervals during the investigation. Volume of stream flow was estimated according to the method proposed by Robins and Crawford (1954).

Water samples were collected each month from each station for determination of biochemical oxygen demand (BOD) and chemical oxygen demand (COD) (A.P.H.A., 1960). BOD tests through October, 1963, were seeded with water obtained from Station 59.6. This seed proved unsatisfactory and during the remainder of the study samples were seeded with settled sewage obtained from Stillwater Sewage Plant.

Chlorides were measured once during the study by titration with .0141 N silver nitrate (A.P.H.A., 1960). Bottom core samples were collected several times to determine physical nature of the bottom. Particle size was determined by passing samples through a vertical nest of five sieves with openings from 0.5 mm to 5.0 mm (Welch, 1948).

Sample Size

To determine adequacy of sample size (n), ten Ekman dredge samples were collected 23 March, 1964, from marginal areas at Stations 14.6, 30.9 and 59.6. A modification of a method described by Gaufin, Harris and Walter (1956) was used to determine sample size required to obtain an adequate sample of species present. It must be assumed that $n = 10$ collects 100% of the species, since ten samples were taken from each station. It is realized that this is not a correct assumption in a nonpolluted situation; however, the technique offers useful results. A coefficients table was constructed by the formula,

$$a_{jk} = \frac{\binom{n-k+1}{j} \times (j)}{\binom{n}{j} \times (n-k+1)}$$

where a_{jk} is the coefficient in kth column and jth row, $\binom{n-k+1}{j}$ and $\binom{n}{j}$ are combinations and $n = 10$. The value k is sample number and row j provides a distribution of probabilities that a species having appeared j out of n times will make its initial appearance in the kth sample. Since $n = 10$, the coefficients table has ten columns (k) and initially ten rows (j), with one row decrease for each increase of k. Decrease in rows occurs because certain probabilities do not exist. If a species appears in all ten samples its probability of making its initial appearance in the first sample is 1. Thus, $a_{10\ 1}$ is the only coefficient needed in row j = 10. Likewise, if a species appears in nine samples a probability exists that it will appear initially in either the first or second sample. Thus, a_{91} and a_{92} are the only coefficients needed in row j = 9. If a species appears in only one sample, a probability exists that it will make its initial appearance in any one of the ten samples. Hence, all ten coefficients are needed in row j = 1.

The probability table applies to any situation of n = 10. Actual data were applied to this table by the formula,

$$P_k = \sum_{j=1}^{n-k+1} a_{jk} \frac{m_j}{m}$$

where P_k is average probability of species making their initial appearances in the kth sample, m_j is number of different species appearing in j out of n samples and m is total number of species collected.

Biological

Bottom samples were taken at monthly intervals. Two samples each were taken from marginal areas, pools and riffles at each station. An Ekman dredge was used to collect samples from marginal areas and pools and a Surber square foot sampler was used in riffles. Marginal samples were collected approximately 3 ft from shore and pool samples were taken from the middle of the stream.

Samples collected with the Ekman dredge were washed in a sieve (#40 U. S. standard soil series with openings of 0.420 mm) and preserved in 80% alcohol. In large samples, organisms were removed by flotation in a sugar solution with a specific gravity of 1.12 (Anderson, 1959). Floating organisms were removed to clear water before final preservation.

Species Diversity

Total number of organisms (N), number of individuals per species (n_i) and number of species in a unit area (m) were used in calculating community diversity (H), diversity per individual ($\bar{H}$), maximum diversity (H_{max}), minimum diversity (H_{min}) and redundancy (R) in the following equations (Patten, 1962):

$$H = \sum_{i=1}^{m} n_i \log \frac{n_i}{N}$$

$$\bar{H} = \sum_{i=1}^{m} \frac{n_i}{N} \log \frac{n_i}{N}$$

$$H_{max} = \log N! - m \log (N/m)!$$

$$H_{min} = \log N! - \log [N - (m - 1)] !$$

$$R = \frac{H_{max} - H}{H_{max} - H_{min}}$$

An index of diversity derived from the linear relationship between number of species and logarithm of total individuals was calculated by the following equation (Margalef, 1951):

$$d = \frac{m - 1}{\ln N}$$

Computations were performed at Oklahoma State University on an IBM Type 1410 data-processing machine.

IV. RESULTS AND DISCUSSION

Physico-chemical Conditions

Mean monthly water temperature (C) at seven lower stations and months included in mean seasonal values presented in Figs. 3-10 and Table V are as follows:

Spring		Summer		Fall		Winter	
Mar	17.78	Jun	25.64	Sep	21.43	Dec	0.86
Apr	18.86	Jul	28.78	Oct	22.93	Jan	4.00
May	24.14	Aug	24.42	Nov	15.57	Feb	4.93

Precipitation was not evenly distributed throughout the year. Monthly rainfall exceeded 6 in. in June and July and was less than 1 in. in December, January and February. Precipitation varied from 0.11 in. in December to 9.46 in. in July. Rainfalls of 3.99 in. and 6.52 in. occurred between the June and July sampling dates. Longest periods without rain extended from 25 September to 16 October and from 22 December to 31 January.

Stream flow and turbidity exhibited seasonal and longitudinal variations (Figs. 3 and 4). In general, spring and summer months were periods of high flow and turbidity, whereas during fall and winter reverse conditions prevailed. Longitudinal variation in these conditions was slight in fall and winter and considerable in spring and summer, except in June when variation in flow between the two most distant stations was only 2.81 cfs. Stream flow consisted primarily of effluent at the time of the June collection. Flow increased

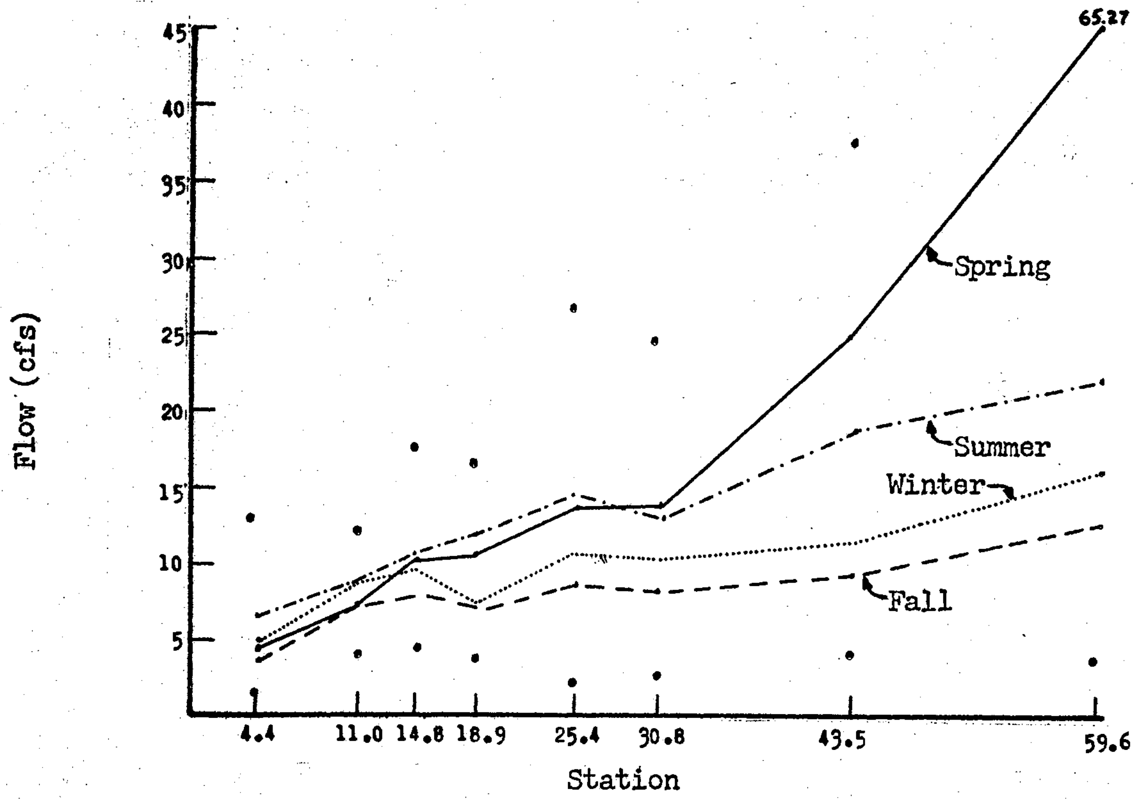

Fig. 3. Mean seasonal flow. Extremes are indicated by dots or by numbers when they exceed maximum ordinate value.

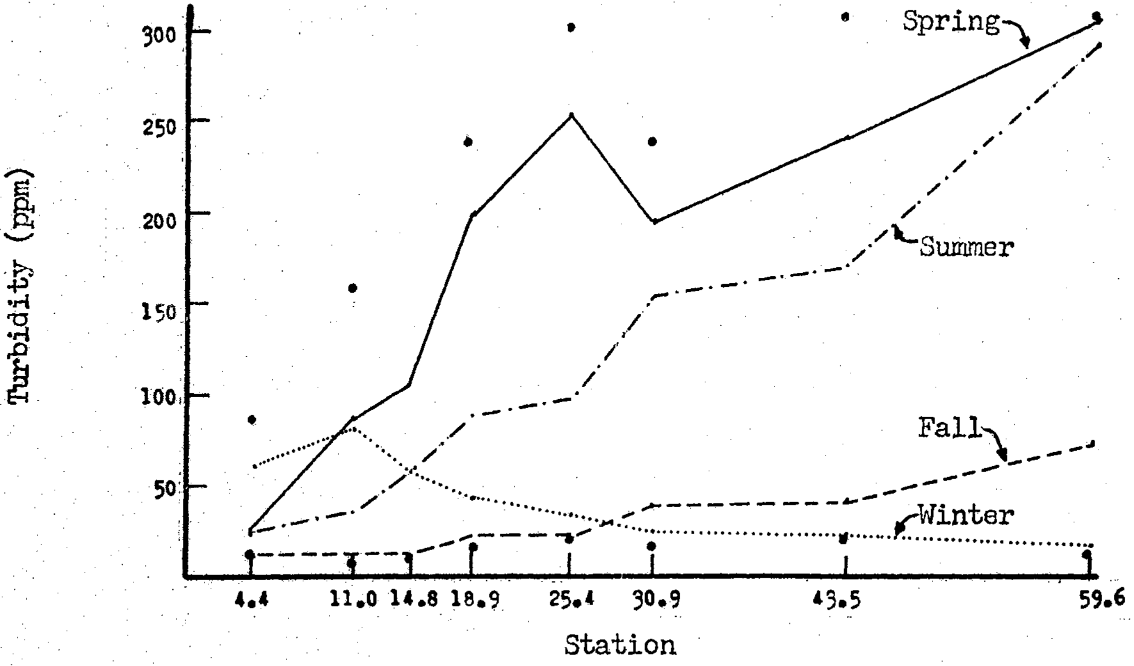

Fig. 4. Mean seasonal turbidity. Extremes are indicated by dots.

uniformly downstream, except in spring when increase was considerable in the stretch below Station 30.9. Low turbidity consistently measured at Station 4.4 was related to possible clearing effect of effluent, sand and gravel bottom and swift current (Table II). Higher turbidity at middle and lower stations in spring and summer was related to silt bottom, low current, inflow of tributaries and extensive use of adjacent land for cultivation.

TABLE II

MEAN ANNUAL PHYSICO-CHEMICAL CONDITIONS

Station	Turbidity ppm	Temperature C	Oxygen ppm	Alkalinity ppm	pH	Conductivity mhos/cm	COD ppm	BOD ppm
4.4	60	21.2	14.28	302	8.3	1724.83	113.34	14.18
11.0	52	18.3	7.84	320	8.1	1752.22	109.70	12.73
14.8	59	17.5	4.43	315	8.0	1775.15	81.18	11.06
18.9	117	17.8	4.39	311	7.9	1723.49	92.01	11.72
25.4	135	17.5	4.37	306	8.0	1618.05	95.73	11.72
30.9	104	16.9	4.81	299	8.0	1629.57	91.46	9.33
43.5	121	17.1	6.47	292	7.9	1748.41	83.72	10.28
59.6	176	16.9	9.17	271	7.9	1337.12	82.62	10.51

Reported flow and turbidity represents conditions only at the time of sampling and reflects preceding precipitation patterns as well as other environmental conditions such as soil moisture prior to precipitation, ground cover and frozen soil. Daily flow is recorded at a gauging station 2.8 miles below Station 43.5 (United States Department of Interior, Water Resourses Division, 1963). Mean daily flow was maximum in July and flow exceeded 1,000 cfs on several occasions in July. Daily flow records correlate well with precipitation records.

Variation in water temperature between stations was slight, except for consistently higher readings at Station 4.4. These higher values are attributed to station sampling order and the shallow, unshaded, clear-water condition at this station. Water temperature exhibited considerable seasonal fluctuation and varied from 0 C in December to 35 C in July. Measured temperature exceeded 23.5 C at all stations during summer and was less than 5 C at most stations in winter. Maximum water temperatures were recorded in July and minimum in December. Ice covered most stations in December.

Longitudinal variation in dissolved oxygen concentration was greater than seasonal fluctuation (Fig. 5). In general, oxygen concentration as determined by samples in daylight hours was high at the two upper and two lower stations and low at middle stations, except at

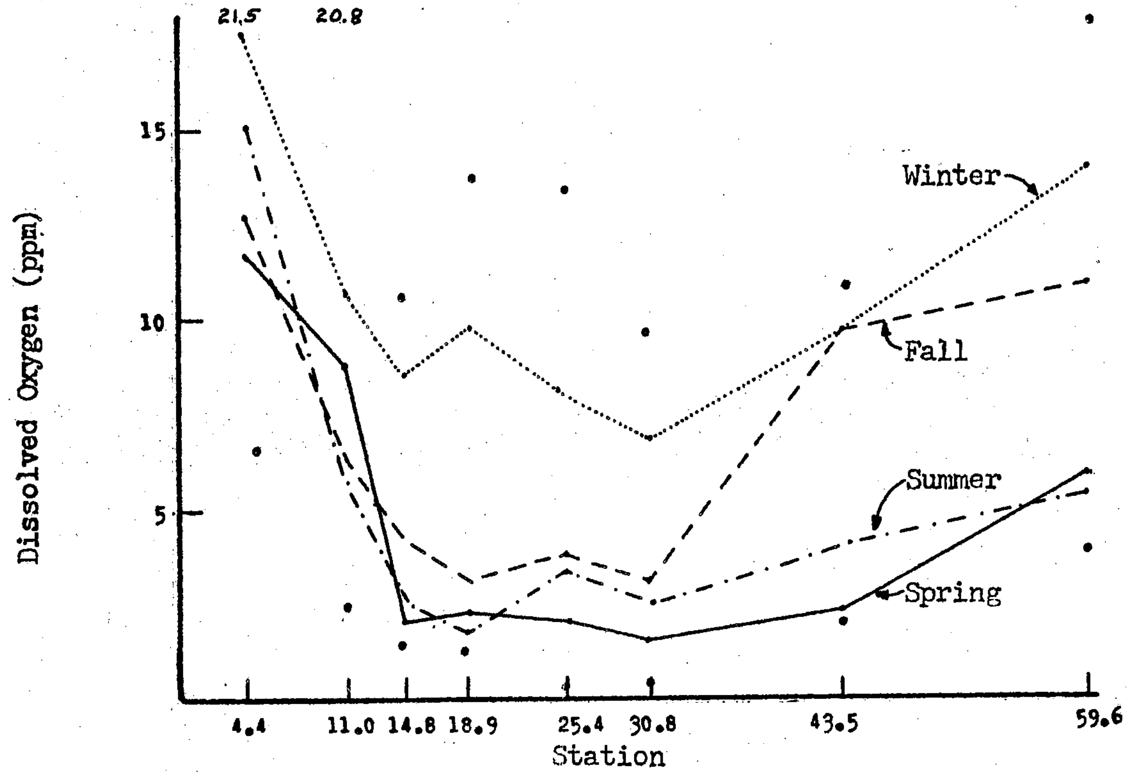

Fig. 5. Mean seasonal dissolved oxygen. Extremes are indicated by dots or by numbers when they exceed maximum ordinate value.

Station 43.5 in spring and summer when oxygen values were low. Greater flows during these seasons probably carried oxygen-reducing wastes farther downstream. Mean oxygen concentration in spring, summer and fall was similar, but winter concentration was higher. Oxygen concentration averaged 3.1 ppm in spring, 3.4 ppm in summer, 4.5 ppm in fall and 7.1 ppm in winter. Minimum oxygen content occurred at most stations in May and maximum in January. Oxygen varied from 0.2 ppm at Station 25.4 and 30.9 in May to 21.5 ppm at Station 4.4 in March.

Depression in oxygen concentration (Fig. 5) may be attributed to several factors. Dense algal growth in upper reaches of the stream may have resulted from organic enrichment (Hynes, 1963). Deoxygenation by bacterial breakdown of organic matter and respiration was undoubtedly high in upper reaches, but during daylight hours was less than oxygenation by green plants during photosynthesis. In middle reaches algae decreased in quantity because of reduction of nutrients, increased flow and dilution, increased turbidity and increase of herbivores (Hynes, 1963). Considerably less oxygen was contributed by photosynthesis and since bacterial decomposition was still high, dissolved oxygen content was low. Farther downstream bacterial decomposition decreased and photosynthesis increased; consequently, dissolved oxygen concentration was high.

Diurnal measurements of oxygen content present a more realistic picture (Table III). Diurnal variation was considerable at the upper stations because of dense algal growths and large oxygen demand of organic material. Diurnal fluctuation was large also at lower stations; however, the amount of organic material was less and minimum oxygen content not as low as in enriched areas. Saturation exceeded 100% at upper and lower stations. Diurnal variation and per cent saturation was considerably less in the middle reaches of the stream and minimum oxygen content was low.

TABLE III

DIURNAL OXYGEN EXTREMES

Station	Dissolved oxygen (ppm)		Variation (ppm)	Per cent saturation		Date
	Minimum	Maximum		Minimum	Maximum	
4.4	2.2	16.5	14.3	26	225	19-20 Jun
11.0	3.1	9.1	6.0	38	128	28-29 Jun
14.8	1.7	6.2	4.5	20	85	28-29 Jun
18.9	4.9	8.8	3.9	45	88	22-23 Mar
	1.0	2.4	1.4	12	30	19-20 Jun
	3.8	5.4	1.6	33	48	6-7 Dec
	7.0	10.0	3.0	54	81	2-3 Jan
25.4	1.5	6.2	4.7	19	85	5-6 Jul
	1.7	5.1	3.4	19	58	27-28 Sep
	8.0	9.8	1.8	61	81	2-3 Jan
	7.0	9.7	2.7	54	82	28-29 Feb
30.9	2.2	5.8	3.6	29	77	5-6 Jul
	2.4	6.7	4.3	27	80	27-28 Sep
43.5	6.5	18.6	12.1	85	254	12-13 Aug
59.6	5.1	15.6	10.5	66	208	12-13 Aug

Lytle Creek, Ohio, in August had considerable amounts of oxygen above the outfall, a sharp decrease at the outfall, a progressive increase downstream with maximum concentration 4.5 miles below the outfall followed by a second depression (Gaufin and Tarzwell, 1952). Initial measurement of dissolved oxygen on Skeleton Creek was made 5.6 miles below the Enid outfall. Measurements farther upstream might have resulted in an oxygen curve similar to that of Lytle Creek. It is probable that the depression in the Skeleton Creek mean summer oxygen concentration corresponds to the second depression on Lytle Creek. On

Lytle Creek maximum diurnal fluctuation in dissolved oxygen occurred 2.8 miles below the outfall and less fluctuation occurred downstream.

Several environmental factors appeared to have little influence on distribution of bottom organisms. Methyl orange alkalinity as $CaCO_3$ equivalent was greater than 120 ppm at all times at all stations, a condition which usually favors high productivity of bottom fauna. Mean annual pH ranged from 8.3 at Station 4.4 to 7.9 at Station 59.6. While both methyl orange alkalinity and pH decreased progressively downstream, variation was slight. Little seasonal fluctuation occurred in either factor, except for slightly lower spring values.

Mean annual conductivity was 1337 mhos/cm at Station 59.6 and varied from 1618 mhos/cm to 1775 mhos/cm at the other stations (Table II). Conductivity was lower in spring and summer, possibly as a result of greater flow and dilution. Chlorides were highest at the upper stations and decreased slightly downstream. Maximum chloride content recorded was 1.16 ppm at Station 11.0.

Mean annual chemical oxygen demand (COD) decreased from 113.34 ppm at Station 4.4 to 82.62 at Station 59.6 (Table II). Only 29% of the COD was satisfied in the 55.2 mile stretch between extreme stations. Mean annual biochemical oxygen demand (BOD) decreased from 14.18 ppm at Station 4.4 to 9.33 ppm at Station 30.9. Only 26% of the BOD was satisfied in the 26.5 mile stretch between these stations. These results indicate that only a small part of the organic materials were broken down in the stream.

Sample Size

Two Ekman dredge samples were collected from marginal areas at each station in the regular sampling program. Tests to determine adequacy of sample size indicated that two samples were sufficient to obtain most species from Stations 14.8 and 30.9 (Table IV). Two marginal samples at Station 59.6 could produce only about 50% of the species. However, results obtained by this procedure are indicative of conditions only during the period when samples were taken. As numbers of species vary, estimates obtained by this technique vary. It was assumed that sample size was adequate to evaluate stream conditions by methods used in this investigation.

TABLE IV

P_k BY STATION

Sample (k)	Station 14.8	Station 30.9	Station 59.6
1	1.00	.900	.355
2		.089	.140
3		.011	.113
4			.091
5			.074
6			.060
7			.050
8			.043
9			.038
10			.036

Community Structure

Distinct longitudinal and seasonal differences in numbers of species and faunal associations existed. Data from Station 18.9 are not included in Tables V and VI and Figs. 6-10 because riffle samples were not taken at this station and consequently a smaller area was sampled.

Large numbers of individuals occurred at the three upper stations, while fewer individuals were present at lower stations (Table V, Fig. 6). Populations were large at upper stations in summer and fall and comparatively smaller in winter and spring, while populations were smaller and more constant downstream. Lower numbers of individuals in spring at upper stations were accompanied by increased numbers of individuals in middle reaches. Maximum numbers of individuals were present at Station 14.8 during all seasons. A large decrease in numbers of individuals occurred in the stretch between Stations 14.8 and 25.4 in all seasons except spring.

A total of 42 species of benthic macroinvertebrates was collected and identified during the study. Numbers of species varied from six at upper stations to 27 at the lowermost station (Fig. 7). The large decrease in numbers of individuals which occurred in the stretch between Stations 14.8 and 25.4 was accompanied by a large increase in numbers of species. More species were present at lower stations in fall and winter than in spring and summer. Minimum numbers of species existed at most stations in July. At that time *Limnodrilus* sp. was the only species present at the five upper stations. *Tendipes attenuatus* was not present at upper stations in July, but was abundant in June and August. Reduction in numbers of species in July may have been influenced by heavy rainfall prior to the July collection date. At that time flow exceeded 6,000 cfs at the U. S. Department of the Interior gauging station.

Six species occurred at the three upper stations, but only *Limnodrilus* sp., *Tendipes attenuatus* and *Pelopia stellata* were abundant

TABLE V

RELATIVE SEASONAL ABUNDANCE OF BENTHIC MACROINVERTEBRATES

Stations	4.4				11.0				14.5				25.4				30.9				43.5				59.6			
Seasons	S	S	F	W	S	S	F	W	S	S	F	W	S	S	F	W	S	S	F	W	S	S	F	W	S	S	F	W
Limnodrilus sp.	1	4	2	2	2	4	4	4	4	4	4	4	3	3	2	2	3	3	2	2	2	2	1	2	2	2	2	1
Tendipes attenuatus (Walk.)	1	3	4	1	1	2	4	1	3	4	4	1	3	1	1	1	2	1	1	1	1	1	1	1	1	1	1	1
Pelopia stellata (Coq.)		2	2			2	2		1	2	1		2	1	1	1		1	1		1	1	1					
Culicoides variipennis (Coq.)		2	1	1		1	1	1	1	1	1	1	1	1	1	1	1	1	1					1	1	1		1
Physa anatina Lea		1		1					1	1	1		1	1	1	1	1		1	1	1	1		1	1	1	1	
Erepetogomphus sp.						1	1						1	1			1				1	1	1		1	1	1	1
Argia sp.											1						1		1		1	1	1	1	1	1		
Helisoma sp.				1									1								1							
Glossiphonia sp.						1	1							1	1	1		1	1	1			1	1				
Pentaneura sp.													1	1	1	1	1	1	1	2	1	1	1	2		1	1	1
Procladius sp.													1	1	1	2	1	1	1	1	1	1	1	1	1	1	1	
Cryptochironomus sp.															1	1		1	1	2			1	1			1	1
Glyptotendipes senilis (Joh.)														1				1	1	1		1	1	2		1	1	
Sphaerium transversum (Say)													1	1	1	1		1	1	1	1	1		1		1	1	1
Simulium vittatum Zett.															1	1			1	1			2	1	1		1	2
Dubiraphia sp.																1								1				1
Lymnaea bulimoides (Haldeman)														1							1	1					1	
Stenelmis sp.																	1					1	1	1	1	1	1	1
Caenis sp.																		1		1				1	1		1	1
Cheumatopsyche sp.																				1		1	1	1	1		1	1
Stenonema sp.																			1				1	1				
Tendipes riparius (Meig.)																			1								1	1
Polypedilium illinoense (Mall.)																												1
Tendipes sp.																							1		1	1	1	
Corydalus cornutus L.																								1		1		1
Tanypus sp. b.																									1			1
Branchiura sowerbyi Bedd.																									1	1	1	
Hexagenia sp.																									1	1	1	

1 = 1-5 individuals/ft^2; 2 = 5-25; 3 = 25-50; 4 = 50

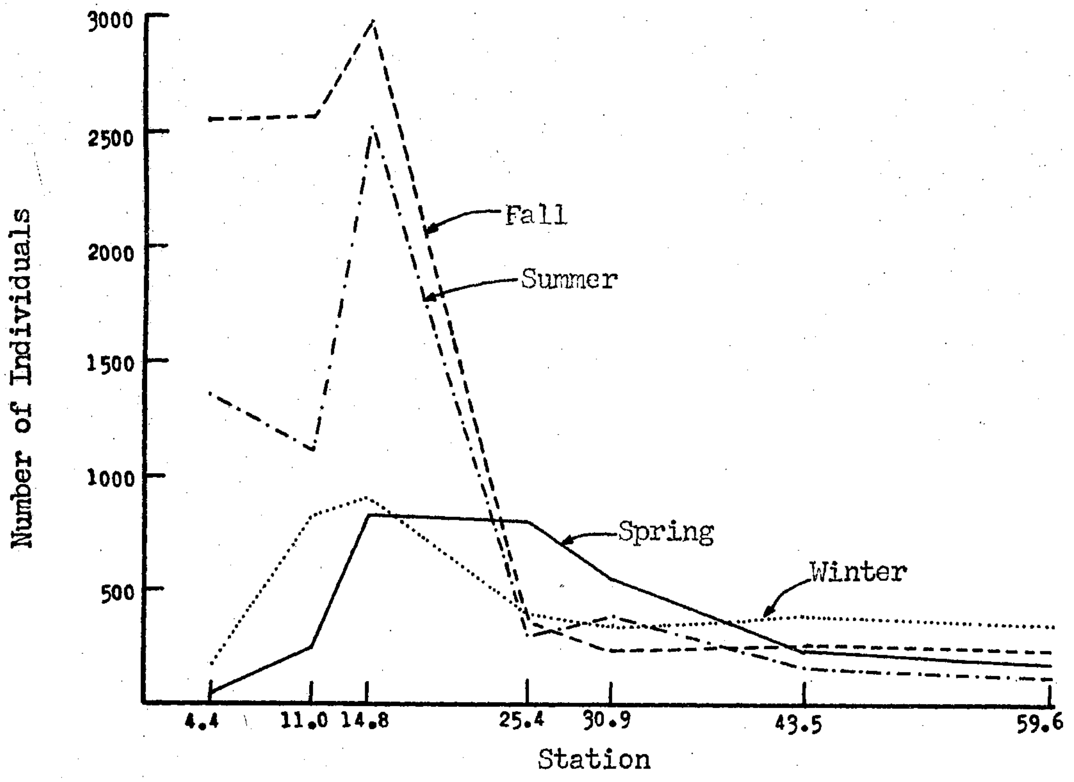

Fig. 6. Seasonal variation in numbers of individuals.

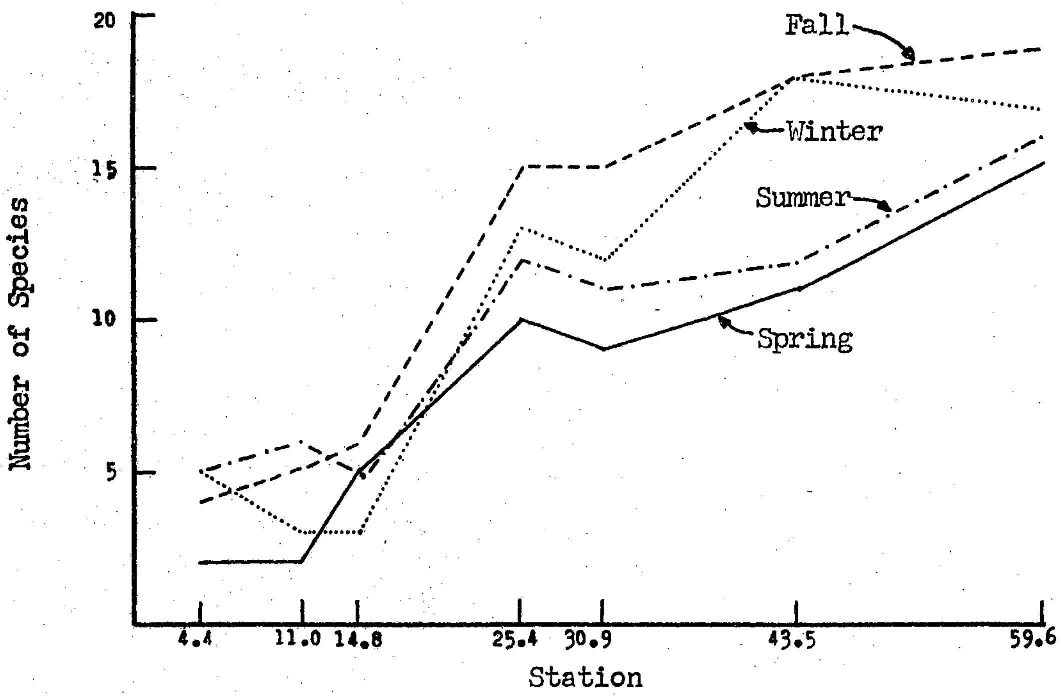

Fig. 7. Seasonal variation in numbers of species.

(Table V). Limnodrilus was most abundant in summer and fall, especially at Station 14.8. Largest numbers of Pelopia stellata occurred in summer, while Tendipes attenuatus was most abundant in fall, especially at Station 4.4. Populations in upper reaches were principally scavenger types which can endure low oxygen concentrations. Gaufin and Tarzwell (1956) reported that during summer and early fall, an abundant food supply and other favorable conditions for growth and reporduction enabled several species occurring in enriched zones to attain great abundance. Reduced early spring and winter populations at upper stations may have been influenced by low flow in those seasons. Mayflies, caddisflies and blackflies were absent from this area. No species was restricted to upper reaches of the stream; all species occurred downstream, but in smaller numbers. Upper sections experienced large diurnal fluctuations in dissolved oxygen with low nocturnal minima in summer, low flow and turbidity, high oxygen demand and frequent changes in substrate. Gaufin and Tarzwell (1956) found large populations of Culex pipiens and Eristalis bastardi in enriched areas of Lytle Creek, but these organisms were not collected during the present study. Possibly, effluent quality or rapid flow prevented their establishment in Skeleton Creek.

Twenty-five species were present in the middle stretch at Stations 24.6, 30.9 and 43.5. Faunal composition here was less constant than at upstream and downstream locations. During spring and summer benthic community structure resembled upstream reaches, while during fall and winter faunal composition was similar to downstream areas. This was especially apparent at Station 43.5. Limnodrilus accounted for greater numbers of individuals at these stations in spring and summer. Mayflies, caddisflies, blackflies and Elmidae were present in middle reaches in fall and winter, but in fewer numbers than at the lowermost station. Most of these insects were taken from fast riffles or shallow margins, where oxygen content was probably higher than in pool areas. Diurnal fluctuation in dissolved oxygen was less than at upper stations. Night-time minima were low in July, however, minima were considerably higher in January. Stream flow was greater than at upper stations.

Thirty species occurred at the lowermost station. No species occurred in large numbers and most species had less than five individuals per square foot. Greater numbers of individuals in fall and winter were principally due to larger numbers of _Simulium vittatum_, _Pentaneura_ sp., _Stenelmis_ sp. and _Cheumatopsyche_ sp. At the lowermost station, _Hexagenia_ sp., _Branchiura sowerbyi_ and _Corydalus cornutus_ were present. Station 59.6 had large diurnal fluctuations in dissolved oxygen, however, night-time minima were higher than at upstream stations. Stream flow was high at this station, especially in spring and summer. Chemical oxygen demand was less than in middle reaches and biochemical oxygen demand was similar to middle stations.

Gaufin and Tarzwell (1952, 1956) divided Lytle Creek into septic, recovery and clean water life zones. The septic zone had large populations of individuals. In May they found a low nocturnal oxygen concentration and an afternoon maximum of 19.2 ppm. As summer advanced, marginal vegetation shaded Lytle Creek, algae blooms were less marked and oxygen values were lower. Populations of mayflies, stoneflies, caddisflies and Elmidae were indicative of clean water conditions. Their absence denoted presence of pollution and/or low oxygen supply if physical nature of the habitat was otherwise suitable. Variety and numbers of organisms in the recovery zone of Lytle Creek were must less constant than in the septic or clean water zones.

In Skeleton Creek, an upper zone down to about mile 15 had large numbers of individuals and few species. Diurnal variation in dissolved oxygen was considerable, but summer afternoon minima were higher than in Lytle Creek. Most of the upper reaches of Skeleton Creek were not tree-shaded as in Lytle Creek. The upper zone in Skeleton Creek probably corresponded to the septic zone described by Gaufin and Tarzwell (1956). The lower part of Skeleton Creek below about mile 59 had populations of mayflies, caddisflies, blackflies and Elmidae. This reach of the stream possibly corresponded to the clean water zone described by Gaufin and Tarzwell; however, stoneflies were not found in Skeleton Creek and the fauna was not as rich and varied as the clean water zone in Lytle Creek. Faunal composition was less constant in middle reaches of Skeleton Creek than at upstream and downstream locations. This stream reach apparently corresponded to the recovery zone described by Gaufin and Tarzwell.

Lytle Creek not only had a different environment and a richer and more varied fauna than Skeleton Creek, but also considerably shorter life zones. A richer and more varied fauna and shorter life zones was also reported in Mad River by Gaufin (1958). Lytle Creek received only domestic sewage and Mad River received both domestic sewage and paper mill wastes. It is possible that oil refinery wastes are more deleterious to benthic macroinvertebrates than domestic wastes, however, regional differences in faunal richness undoubtedly exist. Because of differences in types of effluents and environmental differences, Hynes (1963) stated that there is neither need nor advantage in faunal classification into zones. He suggested that simple graphs and histograms clearly show the effects of pollution. However, graphs must present numerical data which most adequately summarize community structure and stream conditions. Changes which occurred in community structure in the Skeleton Creek recovery zone are not apparent in graphs of numbers of individuals or numbers of species.

Species Diversity

Diversity per individual ($\bar{H}$) and redundancy (R) were found to provide a better means of evaluating community structure. Mean annual R exceeded .50 at the five upper stations and was less at the two lower stations (Table VI). In general, mean annual $\bar{H}$ increased progressively downstream. Maximum $\bar{H}$ and minimum R occurred at the lowermost station during all seasons, reflecting the more varied fauna and improved stream conditions.

TABLE VI

MEAN ANNUAL DIVERSITY INDICES

Station	R	$\bar{H}$ (bits/ind)	d	H (bits/ft^2)	H_{max} (bits/ft^2)	H_{min} (bits/ft^2)	H_{max} - H_{min} (bits/ft^2)
4.4	.56	1.02	.98	86.31	239.26	3.03	236.23
11.0	.56	.84	.83	137.01	288.21	3.42	284.79
14.8	.63	.83	.82	171.59	466.39	4.47	461.92
25.4	.51	1.88	1.81	97.07	176.61	11.06	165.55
30.9	.58	1.59	1.51	59.22	138.60	10.00	128.60
43.5	.44	2.43	2.30	75.14	109.11	12.24	96.87
59.6	.20	3.44	2.99	79.75	97.96	13.45	84.51

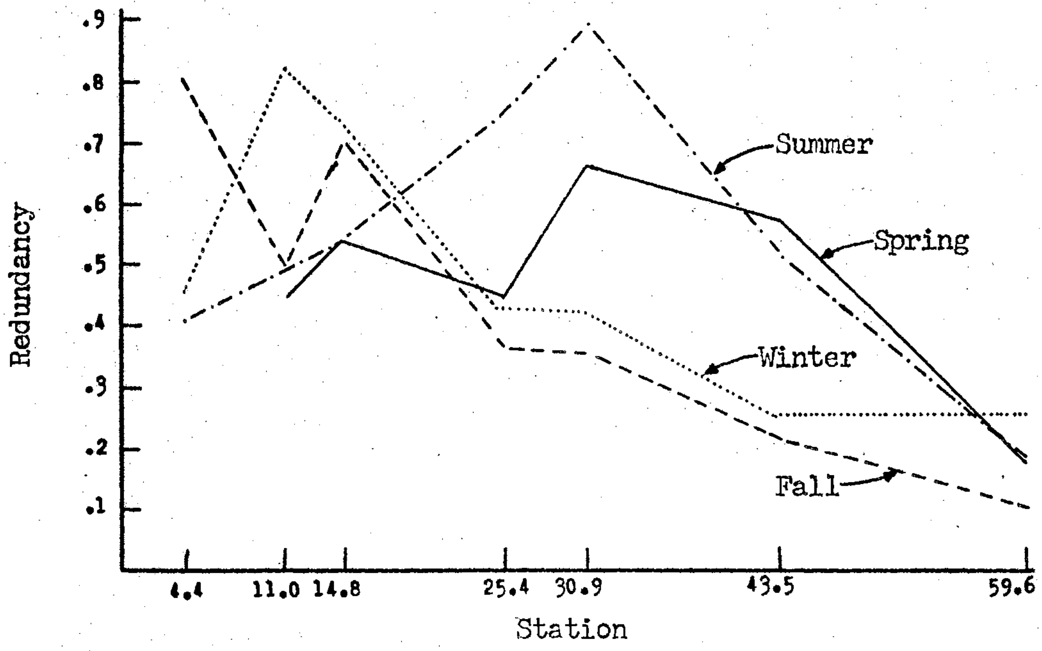

Fig. 8. Seasonal variation in redundancy (R).

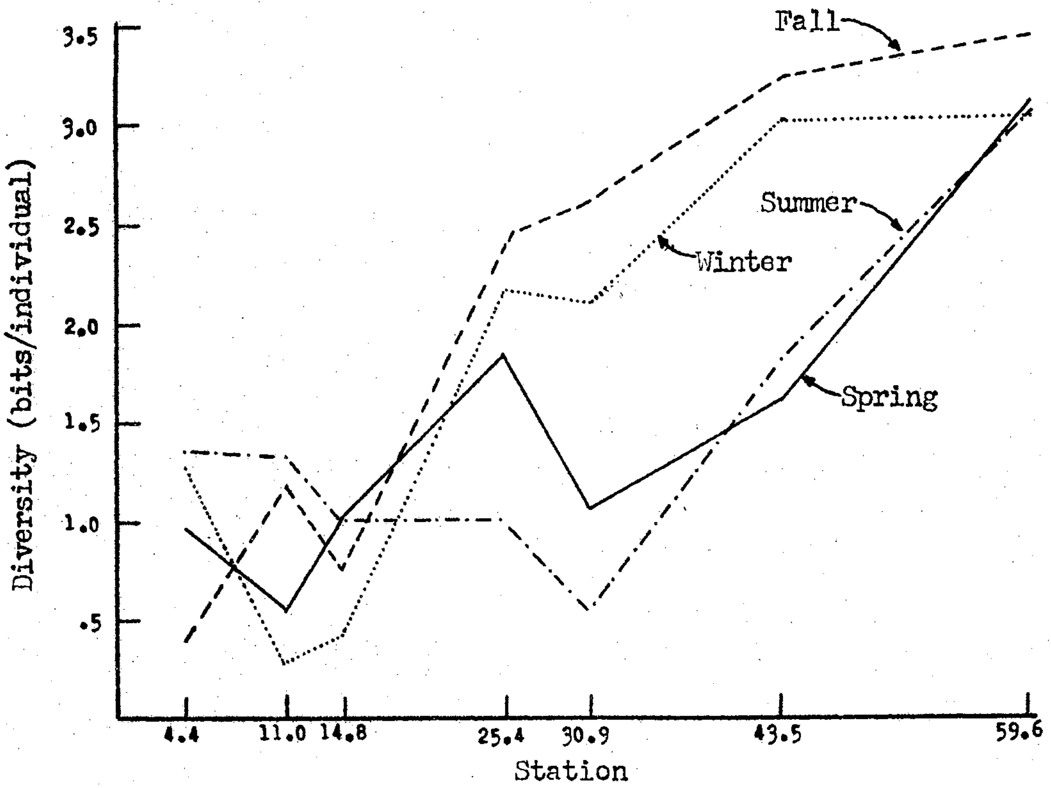

Fig. 9. Seasonal variation in diversity, bits/individual ($\bar{H}$).

In spring, minimum $\bar{\bar{H}}$ existed at Station 11.0, but maximum R occurred at Station 30.9 (Figs. 8 and 9). Only two species were present at Station 11.0 and one of them contained 88% of the individuals. Nine species occurred at Station 30.9, however, 73% of the individuals were *Limnodrilus* sp. and 22% were *Tendipes attenuatus*. Thus, considerable repetition of information existed and R was high. Community structure at Stations 4.4 and 14.8 was similar to that of Station 11.0. Station 25.4 resembled upper stations in R but not in $\bar{\bar{H}}$. Of ten species at Station 25.4 none included more than 50% of the individuals, so that R was relatively low and $\bar{\bar{H}}$ high. Eleven species were taken from Station 43.5 and $\bar{\bar{H}}$ was relatively high; however, Limnodrilus was abundant enough to produce high R.

In summer, peak in R and depression in $\bar{\bar{H}}$ occurred at Station 30.9. Eleven species were collected at this station; however, *Limnodrilus* composed over 90% of the individuals. This species was also abundant enough at Station 25.4 to produce high R. Three species were numerous at upper stations, consequently $\bar{\bar{H}}$ was higher and R lower than at Stations 25.4 and 30.9. Twelve species were present at Station 43.5 in summer, however, almost 70% were one species and consequently R was high.

In fall and winter, R decreased progressively downstream, while $\bar{\bar{H}}$ increased. High R at upper stations was due to abundance of two species. Maximum numbers of species were taken from the lower four stations in fall and winter and although several species were common, none were abundant. Thus, $\bar{\bar{H}}$ was high and R low. A considerable difference between fall and winter $\bar{\bar{H}}$ and R occurred at Stations 4.4 and 11.0. Over 93% of the individuals at Station 4.4 in fall were *Tendipes attenuatus* and 95% of the individuals at Station 11.0 in winter were *Limnodrilus* sp.

Thus, R was high on these occasions. Maximum $\bar{H}$ and minimum R occurred at most stations in fall.

Several other variables were examined for use as estimators in this situation (Table VI). Seasonal variation in species diversity in bits/ft^2 (H), which reflects the compositional richness of a mixed species aggregate of organisms (Patten, 1963), resembled seasonal variations in numbers of individuals in all seasons except winter (Fig. 10). In winter, numbers of individuals were high upstream and low downstream, while H progressively increased downstream. This parameter is dependent on the area sampled, while $\bar{H}$ and R are independent of the area sampled. Maximum diversity (H_{max}), considering

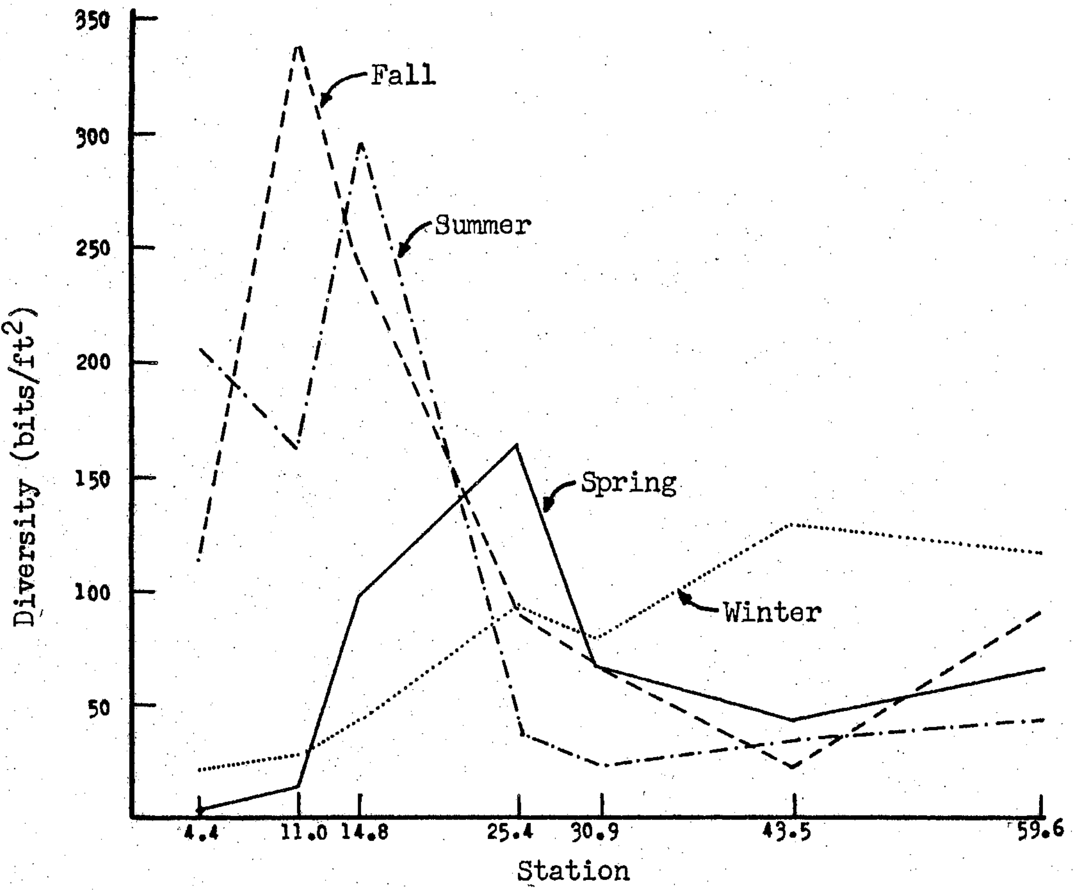

Fig. 10. Seasonal variation in diversity, bits/ft^2 (H).

each individual a different species, increased downstream to maximum values at Station 14.8 and then progressively decreased downstream. Minimum diversity (H_{min}), considering all individuals as one species, progressively decreased downstream and was highest at Station 59.6. The parameter d, derived from linear relationship between number of species and logarithm of total individuals, correlates well with $\bar{\bar{H}}$ (Margalef, 1957).

Use of Information Theory in Evaluating Water Pollution

Information theory methods offer distinct advantages over traditional methods of evaluating polluted stream conditions. This is particularly true when benthic macroinvertebrates are considered. The designation of particular organisms as "indicators" of pollution requires specific identification and many bottom organisms are difficult to identify to species. Information theory methods enable an investigator with limited background in taxonomy to evaluate stream conditions, since less precise taxonomic distinctions need be made. The only data required for community analysis by information theory methods are total number of recognizable species in a unit area. The assignment of a scientific name is not essential.

Two approaches typically are used in the study of natural and polluted communities. One approach emphasizes biomass and production and is concerned with assemblages of organisms in terms of matter and energy. The second approach emphasizes community structure and analyzes communities as complexes of individuals belonging to different species with particular ecological requirements. Few attempts have been made to bridge the structural and functional approaches to the ecosystem in a quantitative way. It is possible that information theory methods may someday make it possible to draw a quantitative expression of possible variations in energy flow as expressed by biomass estimates (Margalef, 1961).

A number of investigators have attempted to classify bottom organisms according to pollution tolerance. Frequently, there exists an appreciable lack of agreement as to the true status of many of these organisms because of regional differences in species and environment. Thus, it is often difficult to compare results of surveys of benthic macroinvertebrates. It becomes increasingly difficult to compare surveys involving different ecological groups, such as plankton and fish. Information theory methods may make such comparisons possible. Presently, it is difficult to compare results of this study of benthic macroinvertebrate community structure with concomitant studies of Skeleton Creek.

Associations or populations of benthic invertebrates provide more reliable criteria of organic enrichment than mere occurrence of a specific species. Gaufin (1956) states "... in order for any comprehensive list or grouping of indicator organisms to be applicable to all sections of the country, the pollutional status of typical aquatic organisms throughout the country must be studied and such information incorporated into the list." Use of associations or populations of benthic macroinvertebrates to describe polluted communities usually involves long, cumbersome lists and descriptions. Information theory methods summarize community structure clearly and briefly by providing numerical indices.

Benthic populations in Skeleton Creek were large at upper stations in summer and fall and comparatively smaller in winter and spring, while they were smaller and more constant downstream. Numbers of species progressively increased downstream. The patterns were reflected in graphs of numbers of individuals and numbers of species (Figs. 6 and 7), but these representations did not reflect changes in community composition. Changes in the distribution of individuals into species were reflected in graphs of redundancy (R) and diversity per individual ($\bar{H}$) (Figs. 8 and 9). For example, in the middle reaches of Skeleton Creek community composition in spring and summer was similar to that upstream, while in fall and winter it was similar to that downstream. This pattern was not apparent in graphs of numbers of individuals or numbers of species, but it was reflected in graphs of R and $\bar{H}$.

Physico-chemical conditions such as precipitation, flow, turbidity and conductivity, and to a lesser extent oxygen, showed more similarity between fall and winter and between spring and summer (Figs. 3, 4 and 5). In contrast, numbers of individuals were similar in summer and fall and in spring and winter. The seasonal similarities in physico-chemical conditions were reflected in graphs of R and $\bar{H}$. Thus, the parameters R and $\bar{H}$ are more closely related to extant physico-chemical conditions than numbers of individuals or numbers of species. It appears that R and $\bar{H}$ are more adequate in evaluating stream conditions using benthic macroinvertebrates than traditional methods.

V. SUMMARY

1. A study of physico-chemical conditions and community structure of benthic macroinvertebrates in a stream receiving domestic and oil refinery wastes was conducted between March, 1963, and February, 1964. An attempt was made to determine seasonal environmental changes, investigate effect of physico-chemical conditions on benthic macroinvertebrate populations, relate quantitative composition of benthic macroinvertebrates to various degrees of pollution and apply methods derived from information theory to benthic macroinvertebrate community structure. Benthic macroinvertebrates were collected from marginal areas, pools and riffles at eight stations.

2. Monthly precipitation exceeded 6 in. in June and July and was less than 1 in. in December, January and February. In general, spring and summer months were periods of high flow and turbidity; however, during fall and winter reverse conditions prevailed. Dissolved oxygen concentration, as determined by single daytime samples was high at upper and lower stations and low at middle stations. Longitudinal variation in dissolved oxygen content was more apparent than seasonal fluctuations. Diurnal variations of dissolved oxygen were considerable at upper and lower stations and less at middle stations. Only 29% of the chemical oxygen demand and 26% of the biochemical oxygen demand was satisfied in the sampled stream area.

3. Analysis of data to determine adequacy of sample size indicated

that two marginal samples were sufficient to obtain most species from Stations 14.8 and 30.9, while two samples could produce only 50% of the species at the lowermost station.

4. Forty-two species of benthic macroinvertebrates were collected and identified during the study. Populations were large at upper stations in summer and fall and comparatively smaller in winter and spring, while populations were smaller and more constant downstream. Numbers of species varied from six at upper stations to 27 at the lowermost stations.

5. *Limnodrilus* sp., *Tendipes attenuatus* and *Pelopia stellata* were abundant at the three upper stations and present in smaller numbers at lower stations. Benthic community structure in middle reaches in spring and summer resembled upstream areas and in fall and winter was similar to downstream areas. Downstream areas had populations of mayflies, caddisflies, blackflies and Elmidae.

6. Maximum diversity ($\bar{H}$) and minimum redundancy (R) occurred at the lowermost station during all seasons, reflecting the more varied fauna and improved stream conditions. In spring, minimum $\bar{H}$ existed at Station 11.0, but maximum R occurred at Station 30.9. In summer, peak in R and depression in $\bar{H}$ occurred at Station 30.9. In fall and winter, R decreased progressively downstream, while $\bar{H}$ increased.

7. Information theory methods of evaluating stream conditions using benthic macroinvertebrates offer several distinct advantages over traditional methods. Information theory methods enable an investigator with limited background in taxonomy to evaluate stream conditions. The Skeleton Creek data suggest that $\bar{H}$ and R are more closely related to

extant physico-chemical conditions than numbers of individuals or numbers of species. It was also observed that change in community composition in middle reaches was not especially reflected in graphs of numbers of individuals or species, but was apparent in graphs of $\bar{H}$ and R.

8. Several other variables were examined for use as estimators in this situation. The parameter d correlates well with H. Community diversity (H) and maximum diversity (H_{max}) increased downstream to maximum values at Station 14.8 and then progressively decreased downstream. Both of these measures were high in summer and fall and correlate with numbers of individuals. Minimum diversity (H_{min}) progressively decreased downstream. It appears that in this community situation, $\bar{H}$ and R are the most useful estimators of stream conditions.

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VITA

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Doctor of Philosophy

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