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FAUNAL CHANGES IN A SMALL EAST TENNESSEE RESERVOIR FOLLOWING REMEDIAL RECLAMATION OF COAL SURFACE MINE DRAINAGE

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ABSTRACT

Remedial reclamation of coal surface-mined land in the Ollis Creek watershed, Campbell County, Tennessee, was initiated on 125 ha (308 ac) of 162 ha (400 ac) total disturbed area in fall 1974 and was continued through spring 1978. In addition to a watershed monitoring program, studies in a reservoir receiving mine drainage ameliorated by an upstream reservoir were conducted to assess recovery of aquatic biota. Periodic water quality and biological measurements in this reservoir showed degradation following mining, and improvement following remedial reclamation. Three fish species were found in initial surveys during the reclamation phase; subsequent reintroductions during improving conditions increased this to six species. A limited analysis of metals in vertebrates revealed relatively low bioaccumulation. Aquatic macroinvertebrates were relatively diverse and abundant in shallow, vegetated overbank areas but depauperate in deeper areas, which probably reflected low habitat diversity as well as fish predation in this exposed area.

INTRODUCTION

The Coal Creek coal seam, one of the most important in Campbell County, Tennessee (Luther 1959), is the dominant recoverable seam in the Ollis Creek watershed. This seam ranges in thickness from 102 to 152 cm (Nelsen 1980). Frequently it has an associated "rider" or parting of the coal seam that is up to 10 cm thick and uneconomical to recover. Between spring 1970 and spring 1972, approximately 162 ha of the 2800 ha^2 Ollis Creek watershed were mined. During mining, coal from the rider, as well as pyritic material from the overburden, were mixed and left on or near the surface, creating an increasingly acidic environment upon oxidation. Even though conventional reclamation was continued through 1973-1974, a survey in spring of 1974 revealed only 24 percent ground cover on the minesite. In 1974 remedial reclamation plans were developed and in 1975 funds were obtained from the Environmental Protection Agency to monitor terrestrial and aquatic recovery for five years. Remedial reclamation was begun in 1975. Aquatic monitoring reported here is only one aspect of the total project (Zarger et al. 1979, 1981).

The literature is replete with the debilitating environmental effects of classic acid mine drainage. Acid and metals are toxic to aquatic life and frequently make water unfit for industrial and domestic consumption. Because of low pH, very high concentrations of sulfate, iron, aluminum, copper, zinc, lead, arsenic, and manganese are possible.

Studies of the biological aspects of mine-influenced lake ecosystems are mainly descriptive documentation of species occurrence. One of the first, Lackey (1938, 1939) found very few species tolerant of acidic conditions but some of those species were locally abundant. Lewis and Peters (1955) determined fisheries potential to be limited by low pH. Smith and Frey (1971) documented detrimental effects of low pH but found successional trends with increasing pH. That is, with improving conditions both diversity and homeostasis increased.

Concurrent with intensive remedial reclamation of mines and monitoring of streams in the Ollis Creek watershed, a study of the downstream-most reservoir in this drainage basin was initiated. In 1972 acid drainage reportedly killed the population of largemouth bass *(Micropterus salmoides)* and bluegill *(Lepomis macrochirus)* in this reservoir. This study was designed to document species occurrence and to determine potential residual metal contamination.

MATERIALS AND METHODS

Study Reservoir

The Ollis Creek watershed has several tributaries which ultimately drain into small reservoir (Reservoir 2, Figure 1). Releases from this reservoir, along with a few small, intermittent tributaries, comprise the entire drainage into the 0.6 ha study reservoir (Reservoir 1).

Reservoir 1 is approximately 5 m deep at the dam and is increasingly shallow in an upstream direction (Figure 2). The east bank descends steeply to the deepest parts of the

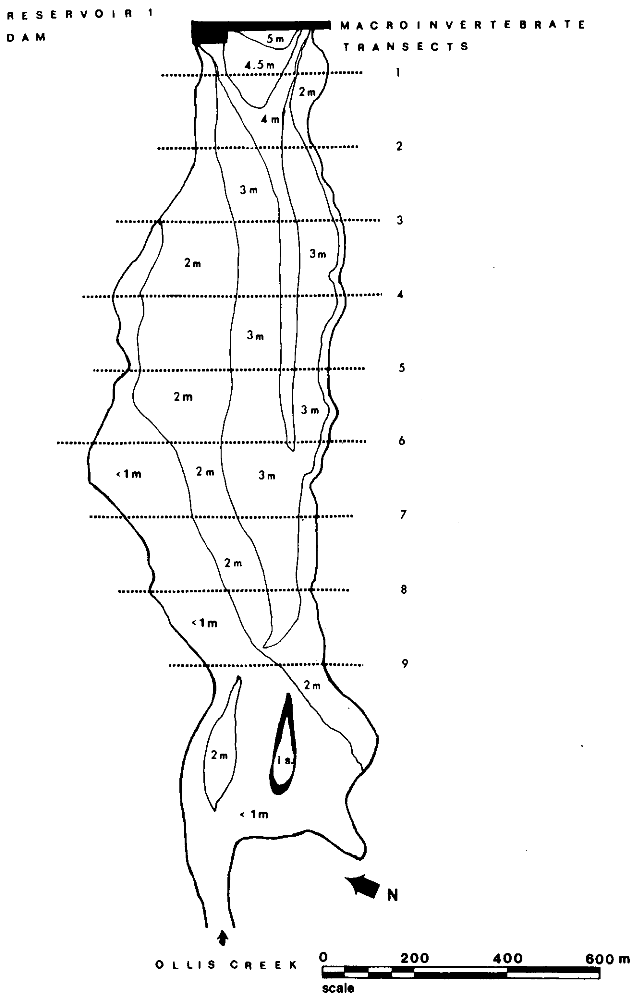

FIG. 1. *Ollis Creek watershed indicating locations of surface mines, Reservoirs 1 and 2 and the watershed location in East Tennessee.*

lake, whereas the west bank drops off slightly then forms a shallow shelf or littoral zone extending toward the middle of the reservoir. In all but extreme upstream portions of the reservoir, which are lotic during low reservoir levels, the substrate is soft mud and organic material. Ollis Creek deposits gravel and cobble in the uppermost sections of this reservoir.

Water Chemistry

At the reservoir dam, a water quality sample was collected at 2 m depth monthly from July 1970 to October 1972. Between August 1975 and December 1976, water quality samples were collected monthly at the surface and 1 m from the reservoir bottom and quarterly thereafter until July 1980. Acidity was determined within a few hours of collection by titration, while pH was measured onsite by portable meters. Metal and sulfate concentrations were determined in the laboratory using standard methods (EPA 1976).

Reservoir depth was determined in April 1977 utilizing a weighted, pre-measured tape. Measurements were taken every 6 m along transects spaced 9 m apart. The survey began at the dam and proceeded upstream.

Macroinvertebrates

The aquatic macroinvertebrate assemblage was sampled also in April 1977 by Eckman dredge every 6.2 m (20 ft) along nine transects in the reservoir (Figure 2). Subsequent samples utilized a Surber sampler (shallow) and Eckman dredge (deep). Vertebrates and macroinvertebrates present in these samples were identified to species where possible, enumerated, and weighed. Qualitative D-net, kick samples were also taken for 15-minutes at the end of the nine transects along the western shoreline.

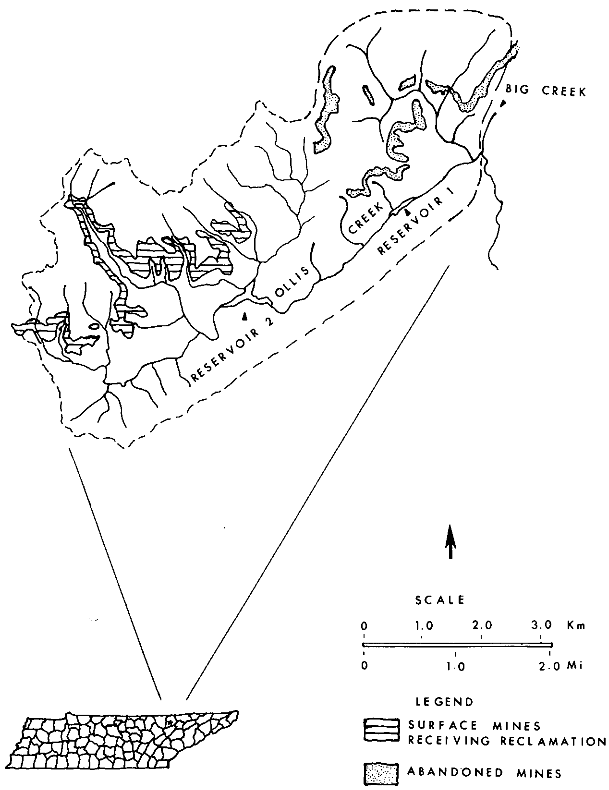

FIG. 2. *Reservoir 1 showing depth contours and macroinvertebrate transect locations.*

Vertebrates and Metals Bioaccumulation

Two fish surveys, April and September 1977, utilized five gill and five hoop nets to determine species present in Reservoir 1 and to obtain fish and snapping turtles for analysis of metal bioaccumulation. The largest (i.e., oldest) individuals were retained for heavy metal analysis and frozen on dry ice; 0.25 to 0.5 g of tissue was homogenzied and perchloric and nitric acids were added and the sample heated until the tissues were digested. Metal concentration was determined by aspiration into an atomic absorption spectrophotometer.

TABLE 1. *Chemical characteristics of individual surface water samples from Ollis Creek Reservoir 1, East Tennessee between July 1970 and October 1972.*

	Mining and Conventional Reclamation					
Date	pH	Fe mg/1	Mn mg/1	Sulfate mg/1	Total Hardness	Total Acidity
July 21, 1970	7.1	0.6	—	18	15	—
August 18, 1970	6.3	0.4	0.4	10	23	5
September 29, 1970	6.7	1.4	1.0	26	24	8
October 27, 1970	6.9	0.7	0.6	30	31	4
November 24, 1970	7.1	0.7	0.9	35	25	5
December 29, 1970	6.0	0.5	0.6	23	22	6
February 10, 1971	5.8	0.2	0.6	25	15	5
March 9, 1971	5.8	0.3	0.4	35	20	6
April 13, 1971	6.0	0.2	0.4	23	18	4
May 3, 1971	7.6	0.4	0.4	25	21	1
June 8, 1971	6.8	0.5	0.4	34	24	3
July 7, 1971	6.2	0.6	0.6	—	41	5
August 9, 1971	6.3	0.3	0.7	40	35	3
September 8, 1971	6.7	0.5	0.4	40	46	2
October 4, 1971	4.9	0.4	1.9	95	74	13
November 2, 1971	6.0	0.5	1.8	70	80	4
December 1, 1971	6.9	0.3	1.4	80	71	4
February 9, 1972	5.0	—	—	—	—	—
April 18, 1972	5.5	0.3	0.8	27	42	6
May 23, 1972	5.3	—	—	—	—	—
July 25, 1972	5.5	0.2	2.8	120	46	8
October 25, 1972	4.5	0.7	2.9	100	86	16

Results and Discussion

Chemical

Water quality parameters measured since 1970 reveal soft water (15-80 ppm $CaCO_3$) that is poorly buffered. During mining (spring 1970 to spring 1972), Reservoir 1 pH ranged from 5.8 to 7.6; however, shortly after mining ceased pH declined to a low of 4.5 in October 1972 (Table 1). Following reclamation-associated fertilizing and liming in 1974, recorded pH varied during the first 5 months of 1975 from an October low of 4.5 to a September high of the 6.6 in 1975 (table 2). The 3 samples taken in 1980 ranged from pH 5.4 (January) to pH 6.4 (July).

Iron and manganese levels differed significantly ($\alpha = 0.05$) most years between surface and bottom measurements. In 1975 mean iron concentrations were 0.26 mg/1 at surface but 0.97 mg/1 at bottom. Similarly mean manganese was 0.96 mg/1 at surface and 1.3 mg/1 at bottom. Mean iron concentrations peaked in 1976 and 1977 with highest manganese concentrations in 1977 and 1978. At the end of the study in 1980, mean iron was 0.4 mg/1 at surface and 0.5 mg/1 at bottom with mean manganese 0.8 mg/1 at surface 1.1 mg/1 at bottom. Iron concentrations were beginning to approach the EPA (1976) maximum recommended for domestic water supplies (0.3 mg/1) and were within criteria for protection of freshwater aquatic life (1.0 mg/1). Manganese surface and bottom concentrations exceed EPA maximum criteria for domestic water supply (0.5 mg/1).

Macroinvertebrates

In the May 13, 1977, quantitative sample, a total of 43 macroinvertebrate taxa were identified (Zarger et al. 1981). Subsequent annual samples had similar (42-46) total taxa except 1980 which had 31 total taxa. Dipterans and odonates usually dominated the macroinvertebrate community (Table 3). Most species collected were typical of ponds or reservoirs with soft, uniform substrate and abundant organic material. Larvae of stream-dwelling organisms (e.g., the trichopteran *Pycnopsyche*) that can tolerate reservoir conditions, were collected in Reservoir 1.

Summers (1981) studied 21 sediment ponds on the Ollis Creek mine. He found a total 130 taxa of aquatic macroinvertebrates dominated numerically by chironomids, ceratopogonids and oligochaetes. More than 50 percent of the species collected in Reservoir 1 were also present in one or more sediment ponds. This species commonality reflects the ability of adult insects to disperse and colonize other bodies of water as well as indicating similarity of water quality and habitat.

In the May dredge samples, taxa and densities varied significantly by depth ($P[H_0] \leq 0.05$). Rawson (1955) considered morphometry to be a dominant factor in the productivity of large lakes. Not surprisingly, macroinvertebrate occurrence was correlated with depth in Reservoir 1, depth influencing both abundance and community structure. In depths less than 1 m, hemipterans, trichopterans, and odonates were the most numerous taxa with an average of 6.8 taxa and 22.3 individuals per square foot. In areas of the same depth where aquatic vegetation was present, coleopterans replaced trichopterans; where a tributary

TABLE 2. *Chemical characteristics of individual surface water samples from Ollis Creek Reservoir 1, East Tennessee between August 1975 and July 1980*

	Remedial Reclamation						
Date	Temperature	pH	Fe mg/1	Mn mg/1	Sulfate mg/1	Total Hardness	Total Acidity
August 25, 1975	27.8	5.6	1.2	0.5	36	—	14
September 17, 1975	22.2	6.6	0.4	0.3	53	—	8
October 30, 1975	14.0	4.5	0.2	1.5	53	—	8
November 20, 1975	9.0	6.5	0.1	1.1	54	—	4
December 18, 1975	—	6.5	0.3	1.5	55	—	2
January 1, 1976	—	5.4	0.5	1.0	42	—	7
February 18, 1976	6.7	5.2	0.4	1.4	60	—	4
March 16, 1976	8.3	5.4	0.2	11.0	57	—	7
April 7, 1976	13.9	5.2	0.6	0.8	45	—	9
May 19, 1976	16.1	5.4	0.5	0.9	44	—	5
June 16, 1976	23.9	5.8	0.4	0.9	42	—	2
July 14, 1976	22.8	5.8	0.3	0.9	48	—	6
August 25, 1976	26.7	6.4	0.5	0.9	76	—	2
September 22, 1976	21.1	5.8	0.5	0.7	45	—	5
October 10, 1976	13.9	5.8	0.8	0.8	120	—	7
November 23, 1976	11.1	6.6	0.3	1.7	120	—	—
December 21, 1976	3.0	5.0	0.7	1.5	64	—	5
April 12, 1977	16.0	5.1	0.2	0.7	55	—	5
July 27, 1977	27.5	7.1	0.4	1.4	99	—	4
October 27, 1977	13.0	5.6	0.4	1.3	62	—	2
January 4, 1978	6.0	5.1	0.3	2.3	58	—	9
May 4, 1978	15.0	5.4	0.2	1.2	33	—	12
July 20, 1978	26.7	6.2	0.3	1.6	90	—	8
October 12, 1978	18.0	7.0	0.4	0.4	71	—	1
January 18, 1979	3.5	5.2	0.2	0.9	52	—	9
April 11, 1979	13.0	4.4	0.3	0.9	42	—	8
July 31, 1979	24.0	5.5	0.3	0.9	60	—	16
October 10, 1979	15.0	6.4	0.3	0.8	77	—	< 1
January 24, 1980	6.5	5.4	0.8	0.9	50	—	6
April 21, 1980	14.0	5.5	0.2	0.9	45	—	5
July 21, 1980	30.0	6.4	0.3	0.7	60	—	6

TABLE 3. *Macroinvertebrate community changes during annual September quantitative samples.*

	1977		1978		1979		1980	
Order	Percent Total Number	Biomass Percent	Percent Total Number	Biomass Percent	Percent Total Number	Biomass Percent	Percent Total Number	Biomass Percent
Ephemeroptera	6.2	3.2	3.1	1.5	1.8	0.7	4.5	2.7
Odonata	32.6	56.3	40.1	66.1	25.3	55.5	14.8	39.5
Trichoptera	1.8	2.4	2.0	0.8	0.9	0.4	2.5	1.5
Megaloptera	2.7	4.1	4.5	7.4	3.1	2.3	3.9	4.6
Diptera	54.6	33.3	48.9	23.7	66.8	40.2	72.5	50.1
Coleoptera	2.1	0.5	1.3	0.5	2.1	0.9	1.4	0.6
TOTAL TAXA	43		46		42		31	

was present, isopods, ephemeropterans, and megalopterans were dominant. In depths of 2 to 3 m, dipterans, coleopterans and *Hexagenia* (Ephemeroptera) were most abundant; the number of average taxa decreased to 4.1. However, average number of individuals increased to 37.8, primarily due to the abundance of dipterans. In depths greater than 3 m, oligochaetes and dipterans were essentially the only groups present; at these depths the average number of taxa was 2.3 with 6.4 individuals present per sample.

Samples in 1978, 1979, and 1980 contained similar macroinvertebrate communities but some groups differed in densities from those in 1977 (Table 3). Productivity remained highest in the weedy littoral zone. Decreases in Ephemeroptera during 1978 and 1979 were primarily due to reductions in *Hexagenia* populations; the increase in

1980 involved *Baetis* sp. In 1980, a drought reduced water levels and consequently overbank area but increased the density of aquatic vegetation where *Baetis* was abundant. Reductions in Odonata after 1979 coincided with fish reintroduction. It was not possible to determine whether the drought, or increased fish predation from recently restocked fish species, or other factors were responsible for changes in numbers of odonates and ephemeropterans. Increases in dipteran numbers probably reflect improved water quality and possibly increased productivity associated with liming and fertilizing to reclaim surface mines.

Vertebrates

Qualitative samples showed a minimum of two frogs *(Rana catesbeiana* and *Rana sphenocephala* one turtle *(Chelydra serpentina)* and two salamanders *(Notophthalmus viridescens,* and *Desmognathus* sp.*)* were present.

On April 24-25, 1977, a total of 38 white suckers *(Catostomus commersoni)* and one bluegill were collected. The suckers were present on deep shelves near shore and the single bluegill was near the dam in deep water. Most of these suckers were sexually mature. Numerous fish in the nets were mutilated, presumably by snapping turtles which were also collected in these nets.

On September 12-13, 1977, a total of 44 fish, including one white crappie *(Pomoxis annularis)*, seven white suckers, nine creek chubs *(Semotilus atromaculatus)* and 27 bluegill were collected. With the change of seasons and warmer water temperatures, fish were found in different habitats.

Adult bluegill were associated with cover such as fallen trees and were only collected along the steep east bank. Juvenile bluegill were found exclusively in the shallow, weedy overbank area. Adult white suckers and creek chubs were found either in the the original stream channel (greater than 2 m depth) or in the area of the reservoir headwaters. Juvenile creek chubs were found in deeper weedy portions of the shallow overbank area.

Although all fish in the 1977 sample appeared to be in good physiological condition, all white suckers collected had enlarged livers, about ten percent filled with cysts. Gills of these white suckers were also thickened and covered with brown slime that was probably sediment disturbed during feeding. In 1979, the TWRA began stocking Reservoir 1 with largemouth bass, bluegill, and channel catfish *(Ictalurus punctatus)*. Juvenile bass and bluegill were present in qualitative shoreline samples in 1980. Channel catfish were not present in these samples, possibly due to mortality or because of their tendency to inhabit deep areas not sampled by this technique.

Bioaccumulation of Heavy Metals

In February 1976, channel catfish were introduced into the reservoir for use in bioaccumulation studies. Despite attempts to acclimate these fish by gradually adding reservoir water to the transportation tank before their release, they immediately showed signs of stress and all died within hours of introduction.

Bluegill (a surface-feeding insectivore), white sucker (a bottom-feeding insectivore), and snapping turtle (an omnivore) were analyzed for copper, cadmium and lead concentrations (Table 4). In white sucker and bluegill, highest levels of copper were in body organs that have excretory functions (kidney and gills). These results correspond with those of Lucas et al., (1970) and Goodyear and Boyd (1972). In Great Lakes fishes, Lucas et al., (1970) found whole fish mean copper concentrations to be 1.3 ppm while mean liver concentrations were 9 ppm. Ollis Creek tissue concentrations are below these concentrations (Table 4). Similar to copper, cadmium highest levels would be expected in fish liver and kidneys (Brooks and Rumsey 1974). Lucas et al., (1970) found whole fish mean cadmium concentrations to be 94 ppb while mean liver concentrations

TABLE 4. *Concentrations of copper, cadmium, and lead in bluegill, white sucker, and snapping turtles in Ollis Creek Reservoir 1, Campbell County, Tennessee.*

	Copper (ppb)		Cadmium (ppb)		Lead (ppb)	
Tissue	Mean	Range	Mean	Range	Mean	Range
Bluegill (3 individuals)						
Muscle	0.42	0.2-0.6	*	*	*	*
Kidney	3.11	1.7-3.7	2.58	1.8- 3.2	*	*
Brain	0.58	0.5-0.6	*	*	*	*
Gill	4.33	1.8-5.0	4.00	4.0- 4.0	*	*
White Sucker (3 individuals)						
Muscle	0.66	0.2-1.1	*	*	0.2	0.0-0.6
Kidney	4.00	4.0-4.0	3.8	3.3- 4.0	0.2	0.1-0.3
Brain	1.40	0.2-2.0	1.3	0.4- 3.1	0.2	0.1-0.4
Gill	3.50	2.2-8.7	4.3	2.9- 6.7	0.3	0.2-0.5
Snapping Turtle (3 replicates of 1 individual)						
Liver	0.33	0.3-0.4	3.23	2.8- 3.7	0.11	0.1-0.2
Brain	0.29	0.1-0.5	5.89	3.4-10.1	0.94	0.7-1.2
Muscle	0.08	0.0-0.2	1.68	0.4- 4.0	0.31	0.0-2.0
Tendon	0.09	0.0-0.1	2.09	0.5- 4.0	0.14	0.1-0.2
Heart	0.12	0.1-0.2	1.24	0.8- 1.4	0.28	0.2-0.3

*Not analyzed

were 400 ppb. While cadmium levels in Ollis Creek fishes are relatively higher than copper or lead concentrations, they are considerably below levels in Great Lakes fishes. Lead levels in Ollis Creek white suckers and snapping turtles are relatively low and uniform for all tissues analyzed. Since lead will accumulate in bones, gills, fins (Brooks and Rumsey, 1974), brains, kidneys, liver, and heart (Badsha and Goldspink, 1982) in fish if lead is elevated in the environment, it can only be concluded that lead is relatively unavailable to organisms in Ollis Creek.

CONCLUSION

Reservoir 1 provided an opportunity to study the response of biota to acid mine drainage ameliorated by intensive remedial reclamation as well as passage through an upstream reservoir. At the initiation of aquatic biological monitoring, two years after remedial reclamation treatments were begun, aquatic macroinvertebrate communities were relatively diverse and abundant in shallow, weedy areas but were reduced in deeper portions of this reservoir. Only three fish species were present. Bioaccumulation tests revealed copper and cadmium concentrations primarily in excretory organs while lead concentrations were relatively low and uniform. At the termination of monitoring, remedial reclamation apparently improved water quality to within standards for aquatic life. Extent of this improvement was suggested by reproduction in 1980 of bass stocked in 1979 by the Tennessee Wildlife Resources Agency.

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VARIATION IN VEGETATIVE GROWTH AND TRICHOMES IN *CANNABIS SATIVA* L. (MARIHUANA) IN RESPONSE TO ENVIRONMENTAL POLLUTION

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ABSTRACT

Four populations of *Cannabis sativa* L. (marihuana) growing in their native habitat and exposed to different levels of environmental pollution were studied for several leaf morphology and leaf trichome features. Leaf length, petiole length, length and width of central leaflet, and the number of teeth on leaf margin decreased with increase in pollution. Trichome length and trichome density values were found to be higher in populations exposed to higher levels of environmental pollution.

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INTRODUCTION

Various plant taxa have been studied to investigate the effects of environmental pollution on different plant organs. Feder (1970) studied carnation and geranium under controlled conditions and found that branching and floral productivity were significantly reduced after the plants were exposed to oxidant-type pollutants for a given period of time. Further, increased internodal length, reduced leaf size, and leaf tissue damage were observed in geranium. Scheffer and Hedgcock (1955) studied the forest trees of the northwestern United States for the effects of sulfur dioxide on these plants and found that leaves exhibited characteristic markings in response to suflur dioxide damage. Studies by Solberg and Adams (1956) revealed that sulfur dioxide damaged spongy mesophyll first and then destroyed epidermal layers and chloroplasts in leaves. Pyatt (1970) is known for his work on the use of lichens as indicators of pollution, i.e. he found a decrease in lichen flora with increasing proximity to the source of pollution. Recent studies (Sharma and Butler, 1973; Sharma, Chandler and Salemi, 1980) indicate the cuticular response of several plant taxa to the stress of environmental pollution. In these studies involving woody and herbaceous dicotyledoneous taxa, cuticular features such as stomatal frequency, trichome frequency and type were studied and it was found that with an increase in the level of environmental pollution there was an increase in trichome density and length with a preponderance of multicellular "barbed" trichomes, while the stomatal frequency decreased with increasing pollution.

Cannabis sativa L. (marihuana), a dioecious dicotyledoneous taxon is probably a native of central Asia where wild or relatively wild populations of the plant are found in a wide variety of habitats. In addition, it is known to be a relatively pathogen free, THC (tetrahydrocannabinol) producing plant. In view of these considerations, it was thought appropriate to determine if this unique taxon responded to the stresses of environmental pollution caused by vehicular traffic in its natural habitat.

Materials and Methods

Cannabis sativa grows wild in the Himalayas. Plant populations were studied along a busy highway in Simla (31° 05′ N latitude and 77° 11′ E longitude), a small mountainous (elevation, 2500 meters) town in the northern Himalayas in northern India. There is no industry in the area and hence no major source of environmental pollution except the vehicular traffic along a busy highway passing through the area.

Four populations (A, B, C, D) representing various levels of environmental pollution were selected (Table 1). Each population consisted of 10 female plants. Populations A, B, and C were growing in the areas exhibiting obvious pollution from vehicular traffic along the main highway while population D was relatively protected from this pollution source since it was growing in a sheltered site behind huge residential dwellings. All the four populations came from fairly identical macrohabitats. They were collected along the same slope representing uniform microhabitat factors such as light, drainage, soil, and photoperiod etc. Morphological data collected included leaf length, petiole length, length and width of central leaflet, and number of teeth on the central leaflet. Leaves were sampled from the lower part of the plant in late summer to ensure consistency and maturity.

For trichome data, slides were made from the central portion of the central leaflets of mature leaves from the lower part of plants. Leaflets were washed with distilled water and mild detergent, air dried and Ducocement™ was applied to the upper surface of leaves (Williams, 1973). Upon drying, the film was removed and a small section from the central portion of film was used for making slides. Trichome density and trichome length were recorded from twenty fields at a magnification of 400x. Trichome data were collected in view of the role of these structures in the secretion of THC in *cannabis sativa*. Photomicrographs of the leaf suface features were taken. The data were analyzed (Table 3).

Results and Discussion

Populations A, B, and C represented varying degrees of environmental pollution along a busy highway where automobile traffic was the obvious source of environmental pollution. Population D was growing in the same general macrohabitat but was protected from the direct effects of automobile pollution because of its location in a semi-protected area behind a dwelling complex. No measurements of the levels of environmental pollution were made but great care was taken to record the obvious levels and sources of pollution around the plant populations under study.

Statistical analysis showing the mean values and the standard deviations of various vegetative and trichome features indicate a distinct pattern in the stress dynamics in this taxon (Tables 2 and 3). Population D, characterized by low level pollution and protected microhabitat had the highest mean value of 14.3 cm. for leaf length while populations A, B, and C representing polluted habitats had low values for this parameter. Mean values for petiole

TABLE 1. *Pollution Gradient of Cannabis Sativa Populations.*

Population	Number of samples	Relative pollution level*	Source of Pollution Vehicular traffic
A	10	+ + + +	
B	10	+ + +	" "
C	10	+ +	" "
D	10	+	" "

* +, lowest; + + + +, highest

TABLE 2. *Vegetative Growth Patterns* of Populations of Cannabis Sativa.*

Population	Leaf length (cm)	Petiole length (cm)	Central leaflet length (cm)	Central leaflet width (cm)	Number of teeth central leaflet
A	8.3 ± 1.7	2.2 ± 0.8	6.1 ± 0.9	1.2 ± 0.1	23.6 ± 2.8
B	6.5 ± 2.0	1.0 ± 0.7	5.4 ± 1.3	0.7 ± 0.1	18.5 ± 3.4
C	9.1 ± 2.8	1.9 ± 0.9	7.2 ± 2.0	0.7 ± 0.2	25.8 ± 6.1
D	14.3 + 5.3	4.2 + 2.6	10.5 ± 2.9	1.4 ± 0.4	32.4 ± 7.7

*The values represent means of 10 measurements ± standard deviation.

TABLE 3. *Trichome Features* on the Adaxial Leaf Surface of Cannabis Sativa Populations.*

Population	Trichome length (μ)	Trichome density (cm^2)
A	220.9 ± 76.5	4,366.6 ± 1355.8
B	179.6 ± 41.4	4,555.5 ± 982.7
C	195.2 ± 42.5	4,086.7 ± 827.0
D	153.7 ± 51.7	2,787.3 ± 909.3

*The values represent means of 20 measurements ± standard deviation.