

# Effects of Oil Field Brines on Biological Integrity of Two Tributaries of the Little Muskingum River, Southeastern Ohio<sup>1</sup>

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**Abstract.** Two headwater tributaries of the Little Muskingum River were compared to assess possible effects of oil field brines on biological integrity of the streams. Diatom, macroinvertebrate, and fish communities were analyzed during the summer, 1990, for possible changes in community-structure caused by brines. Dissolved chemicals associated with brines and natural surface waters were quantified monthly between May and October, 1991. Cranenest Fork, with a lower density of wells producing brines than Straight Fork, had a slightly more integrated benthic macroinvertebrate community and lower proportions of salt-tolerant diatoms. Twelve of fifteen measures of benthic macroinvertebrate community structure were consistent with expected effects of greater brine enrichment in Straight Fork. Both streams, however, conformed to benthic macroinvertebrate criteria for protection of aquatic life established in Ohio Water Quality Standards. The major difference between streams was the larger (13:1) percentage of salt-tolerant diatoms such as *Navicula salinarum*, *Navicula tripunctata*, and *Navicula viridula v. avenacea* in Straight Fork than in Cranenest Fork. Fish communities were similar between streams.

Cranenest Fork and Straight Fork shared relatively similar geomorphological characteristics and land-use patterns. Chemical-physical quality was similar except for conductivity and levels of sodium and chlorides, all of which were significantly greater in Straight Fork, as would be expected with the probability of greater brine enrichment of this stream. The maximum concentration of total dissolved solids (<300 mg/L) was very low compared to most surface water in Ohio. No chemical-physical water quality violations of Ohio Water Quality Standards were noted in either stream.

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## INTRODUCTION

The Little Muskingum River is a small tributary of the Ohio River located on the unglaciated plateau of the Appalachian Highlands of southeastern Ohio. The river has been assigned special status as a State of Ohio Resource Waterway and designated as a warmwater aquatic habitat, suitable for agricultural and industrial uses, and safe for primary body-contact recreation (Ohio Environmental Protection Agency 1987). The watershed is one of the most heavily forested and sparsely populated areas in Ohio. There are no large municipalities. Only a few scattered villages, rural homes, and small farms occur in the area. The watershed is relatively free of major water pollutants associated with mining activities, heavy industry, and municipal sewage, common in other sections of Ohio (U.S. Forest Service 1990).

The primary water quality problems affecting aquatic life in the river are siltation from eroding soils, organic wastes from households and farm animals, and brines from the production of oil and gas within the watershed. The effects of the latter pollutants were the special focus of the present investigation. Accidental and deliberate discharge of brines were known to occur and were suspected of having an impact on aquatic life. Of special interest was the possible effects of brines on aquatic life from so-called Exempt Mississippian (oil/gas) Wells (EMWs).

An Exempt Mississippian Well (EMW) as defined by the State of Ohio, Division of Oil and Gas, must have been

drilled and completed before 1 January 1980; located in the unglaciated part of the state; drilled no deeper than the Mississippian Big Injun Sandstone in the entire area with Pennsylvanian age bedrock or drilled no deeper than the Mississippian Berea Sandstone in the area with Permian age bedrock; and the well must be permanently connected from the wellhead to a residence for domestic fuel supply.

Owners of EMWs must comply with all Ohio laws and rules regulating oil and gas production, but are exempt from paying a fee if brines are not disposed into an injection well, filing a certificate of liability insurance, and filing a plan for storage and disposal of brine with the Ohio Division of Oil and Gas. Otherwise brine disposal practices must follow methods approved by the Division of Oil and Gas of the Ohio Department of Natural Resources. Actual brine disposal practices were a matter of controversy between oil/gas producers and government regulators. In some areas brines are released from EMWs into riparian zones or directly into the river or its tributaries.

The objective of the present study was to determine whether these brine releases significantly affected the biotic integrity of the river. We report here the results of chemical-physical ambient water analyses and of assessments of biological integrity for two headwater streams in the Little Muskingum River system. One stream, Cranenest Fork, with a low density of EMWs within the watershed, is compared to Straight Fork, a stream with a relatively high density of EMWs. The streams are otherwise quite similar with respect to geomorphology and land-use patterns. No reference streams completely free of EMWs could be located.

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## MATERIALS AND METHODS

### Study Area

The Little Muskingum River originates from several tributaries at elevations of 250-300 m in Monroe County and flows generally southwest for approximately 100 km to the Ohio River near Marietta, OH. Two headwater tributaries of the Little Muskingum River, Cranenest Fork and Straight Fork, were selected for comparative analysis (Fig. 1). These streams share relatively similar geomorphometric characteristics and land-use patterns, except for numbers of EMWs per unit area of watershed (Table 1). Straight Fork has more than twice the density of EMWs as Cranenest Fork. Otherwise the streams are similar in watershed area, stream length and gradient, and bedrock composition. Upper Pennsylvanian and Permian Dunkard Systems form the bedrock (Bownocker 1920, Stauffer and Schroyer 1920, Thornbury 1965). Bedrock exposures include Green and Washington Formations of the Permian System and the Monongahela Formation of the Pennsylvanian System. Though shales are the dominant bedrock, sandstones and limestones affect steepness of valley sides, local stream gradients, and ground water supplies. Springs are more numerous in areas underlain by limestone bedrock (Hayhurst et al. 1974). Riparian vegetation is similar for both streams, consisting mostly of aggrading hardwood forests. Land-use, except for intensity of oil/gas production, is limited primarily to widely dispersed rural homes and small farms. No municipal sewage systems discharge to either stream.

Brines from EMWs in the study area have been analyzed by the Ohio Division of the Geological Survey. Based on 11 samples, mean chloride levels of approximately 61,000 mg/L were noted. A typical analysis of a filtered sample was (in mg/L): Na 26,500, potassium 176, magnesium 1,440, calcium 9,000, strontium 138, lithium 2, iron 156, iodine 10, bromine 622, chloride 61,200, total dissolved solids 100,600, pH 5.7 (Ohio Geological Survey 1991).

### Chemical/Physical Water Analyses

Water samples for selected chemical-physical analyses were collected monthly between 10 May and 11 September 1990 at single sites on each stream (as shown in Fig. 1). Sampling events included an over bank flood (17 May) and low flow of one-tenth mean annual discharge (2 August). Sample collection, initial processing, and preservation methods were in accordance with *Standard Methods* (American Public Health Association 1989). Water samples were analyzed in the laboratory using United States Environmental Protection Agency (1984) protocols and *Standard Methods* (American Public Health Association 1989). Analyses were completed for chloride, bromide, sulfate, bicarbonate, nitrate, sodium, potassium, calcium, magnesium, iron, manganese, zinc, chromium, lithium, strontium, lead, barium, and arsenic. These elements and ions were selected for analysis because of the possibility that a "chemical fingerprint" of EMW brines could be detected (Breen et al. 1985). Quality control of chemical analyses was assured by using chemical standards provided by the United States Environmental Protection Agency and by arranging for duplicate analyses of split water samples with the Ohio Environmental Protection

Agency water analytical laboratory. Results between laboratories were nearly identical. Water temperatures, dissolved oxygen, pH, and conductivity were determined for ambient water at the time of each sample collection. Water temperature was measured with a thermistor; dissolved oxygen with a YSI Model 57 meter; pH with a Corning Autocal Model 108 pH meter; and conductivity with a YSI Model 33 S-C-T meter.

### Benthic Macroinvertebrates

Benthic macroinvertebrates were sampled from Hester-Dendy artificial substrate samplers (Hester and Dendy 1962) and from natural substrates. Hester-Dendy samplers were exposed for approximately six weeks (Ohio Environmental Protection Agency 1988). Natural substrates were sampled with a modified shovel sampler, a device designed to retain all organisms within the top 10-20 cm of sediments, so that accurate quantitative results could be obtained (Prater et al. 1977). Samples of natural substrates with associated invertebrates were placed into a 10 L bucket half-filled with stream water. The organisms were dislodged from the substrates with a stiff bristled brush and by agitation. Dislodged organisms were screened from the water with a U.S. Standard No. 30 soil sieve and placed into 500 ml plastic jars. The jars of organisms were retained temporarily on ice while transported to the laboratory. Within 24 hours the living organisms were hand-picked from the collection, sorted into major taxonomic groups, and preserved in 80% ethanol.

All organisms were identified to the lowest practical level using taxonomic keys primarily from Merritt and Cummins (1984) and from Peckarsky et al. (1990). Individuals of each species (or lowest taxon) were counted. From these data a variety of measures of community composition were determined. These included the total number of taxa; the number of taxa and the proportion of selected groups including ephemeropterans, plecopterans, trichopterans, and dipterans; and the proportion of organic pollution-tolerant organisms. These measures of benthic community composition have been widely used as indexes of water quality (Hellawell 1986, Plafkin et al. 1989). The Ohio Environmental Protection Agency recently has incorporated these measures into a composite index known as an Invertebrate Community Index (ICI). Values for ICI have been calibrated for various regions of Ohio and incorporated into Ohio Water Quality Standards (Ohio Environmental Protection Agency 1987). In addition to these measures, the density of benthic macroinvertebrates was determined. This measure of benthic community composition is not widely used as an index of water quality because of difficulties in making accurate estimates. In the present study, estimates of density, even though scaled over a wide range, were considered important because decreased densities of benthic invertebrates were an expected major effect of brine enrichment. Three composite indexes of water quality, Shannon-Weiner diversity (Wilhm and Dorris 1968), Belgian Biotic (Depauw and Vanhooren 1983), and Trent Biotic (Woodiwiss 1964) were calculated from the inventory of taxa. These indexes are well-known and have been used widely to assess ambient water quality in

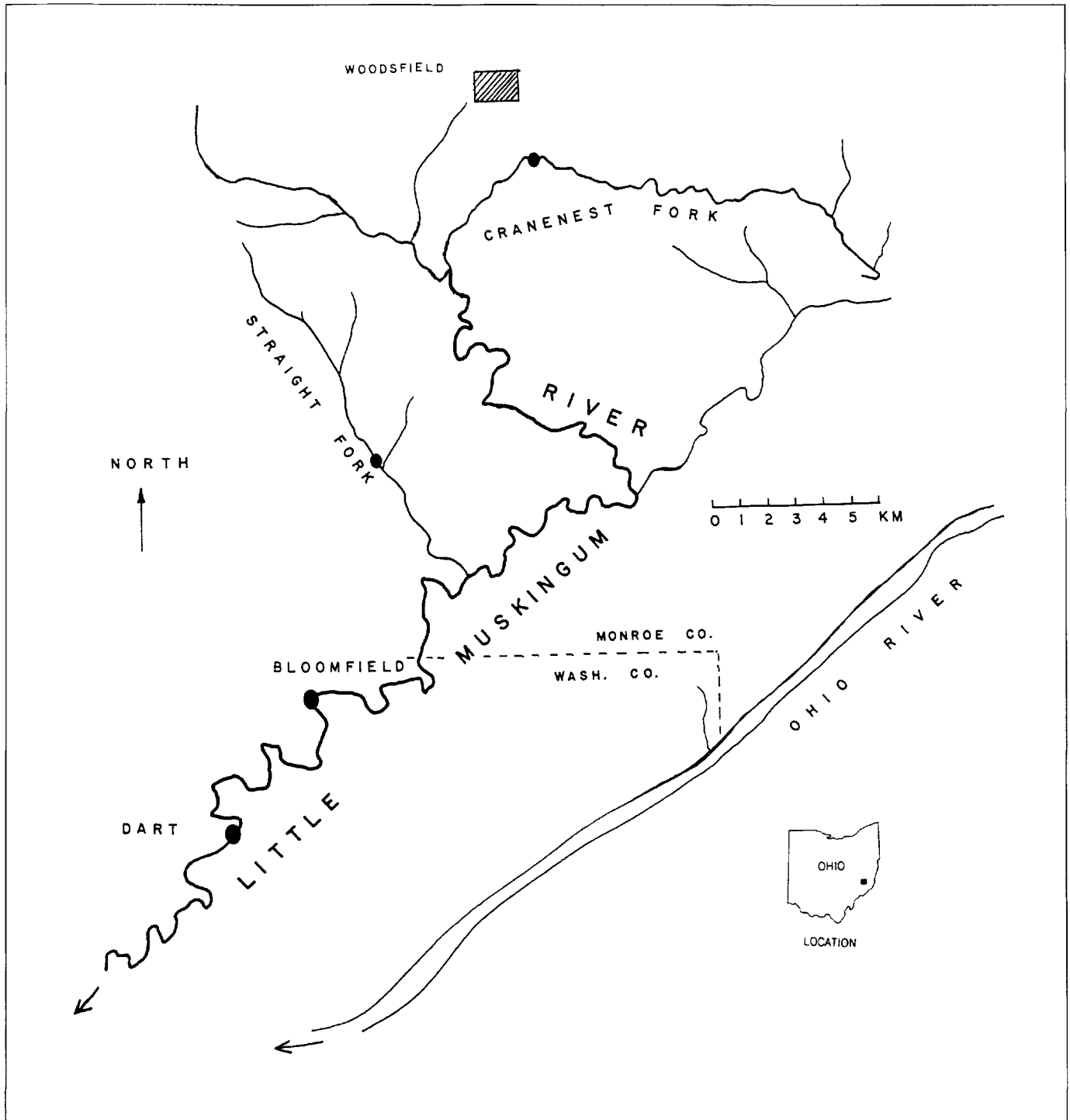


FIGURE 1. Map of Little Muskingum River and vicinity showing locations of sampling stations for chemical-physical and biological assessments.

streams and rivers (Hellowell 1986). Although each index incorporates many of the same measures of benthic community composition, each provides a sufficiently different perspective of benthic composition so that the best possible assessment of benthic community integrity can be made.

### Epilithic Diatoms

Epilithic diatoms were collected from selected sample locations on 6 June 1990. Rocks not larger than 100 cm<sup>2</sup> in surface area that appeared to have well-developed diatom

communities were selected, placed into plastic bags, and transported on ice to the laboratory. Diatoms were scraped from the rocks and cleaned of organic matter with a strong oxidizing solution of nitric acid and potassium dichromate (Patrick and Reimer 1966). Cleaned diatoms were mounted in Hyrax™ on microscope slides. Five hundred diatoms were selected at random for identification under bright-field illumination at magnifications up to 1000X. Most diatoms were identified from taxonomic keys in Patrick and Reimer (1966, 1975).

An inventory of taxa was developed for the diatom

TABLE 1

*Comparison of selected geophysical characteristics and density of exempt Mississippian Oil/Gas Wells for Cranenest Fork and Straight Fork, Little Muskingum River, southeastern Ohio.*

Characteristic	Cranenest Fork	Straight Fork
Length (km)	22	15.8
Elevation at source (m)	366	326
Gradient at mouth (m)	231	204
Gradient (m/km)	5.7	7.7
Watershed area (km <sup>2</sup> )	50	19
Total oil/gas wells <sup>a</sup>	334	213
Total oil/gas wells <sup>b</sup>	328	215
Possible producing EMWs <sup>a</sup>	148	149
Mean possible producing EMWs/km <sup>2</sup> of watershed <sup>a</sup>	3.0	7.8

<sup>a</sup>Numbers based on Monroe and Washington county tax maps. Some error, probably  $\leq 5\%$ , may be due to problems of scale, lack of clarity of map symbols, and probable lack of updating of well data.

<sup>b</sup>Numbers based on data from Ohio Department of Natural Resources, Division of Oil and Gas.

community from each sample location and the percent composition for each taxon in the community was calculated. Interpretation of these data was based on relative numbers of diatoms and on known environmental requirements for each taxon of diatoms (Lowe 1974).

### Fish Communities

Standardized collections of fish were made at each location on 26 June 1990 with a 6 m bag seine and 2.5 m minnow seine. Because the streams were small and water clarity was good, electrofishing techniques were not required. All potential habitats within a 300 m stream segment were sampled. Most fish were identified and counted in the field, checked for obvious abnormalities and external parasites, and returned to the stream. Voucher specimens, very small specimens, and species difficult to identify were preserved in ethanol and transported to the laboratory. Taxonomic keys in *The Fishes of Ohio* (Trautman 1981) were used to identify each specimen. Each specimen was examined for deformities, eroded fins, lesions, and tumors, so-called DELT anomalies (Karr 1981, Ohio Environmental Protection Agency 1988).

An inventory of fish species and numbers of individuals per species were obtained for each sample location. From these data, Indexes of Biotic Integrity (IBI) were calculated (Ohio Environmental Protection Agency 1988). The IBI is a composite index based on 12 measures (or metrics) derived from species composition, trophic composition, and condition of the fish in the community. This index was originally proposed by Karr (1981) and has been modified for application in Ohio streams (Ohio Environmental Protection Agency 1988). Biological criteria in Ohio Water Quality Standards (Ohio Environmental Protection Agency 1987) establish an IBI value of at least 44 for warmwater habitats in this ecoregion of Ohio.

## RESULTS

### Chemical/Physical Assessment

Concentrations of selected inorganic ions and conductivity values were low compared to most surface water in Ohio (Table 2), reflecting high quality water in both streams and indicating that the water should be suitable for warmwater aquatic life and acceptable for most domestic, agricultural, and industrial uses. No deviations from acceptable water quality criteria for the designated uses of these waterways were noted (U.S. Environmental Protection Agency 1986). No violations of Ohio Water Quality Standards (Ohio Environmental Protection Agency 1987) were noted. Chloride concentrations and conductivity values, indicators of possible brine pollution, were relatively low in both streams. The maximum chloride level of 74 mg/L in Straight Fork was well below the 250 mg/L chloride permitted in Ohio Water Quality Standards (Ohio Environmental Protection Agency 1987). Also, in Straight Fork the maximum conductivity value of 505  $\mu\text{mhos/cm}$ , indicating approximately 303 mg/L total dissolved solids (TDS), was substantially below the 1,500 mg/L TDS allowed under Ohio Water Quality Standards. Water temperatures, ranging from 15°C in May to 27°C in July, were normal and did not deviate from temperatures suitable for the designated uses.

Concentrations of sodium, potassium, calcium, magnesium, and iron were relatively low compared to most surface water in Ohio (U.S. Geological Survey 1989). Based on six analyses of each element, concentrations never exceeded 32 mg/L for sodium; 3 mg/L for potassium, 67 mg/L for calcium, 14 mg/L for magnesium, and 0.17 mg/L for iron. Concentrations of manganese, zinc, chromium, lithium, strontium, lead, barium, and arsenic also were very low, usually below detection limits. Because of the very low levels of these elements, it was not possible to distinguish a "chemical fingerprint" for EMW brines.

Although all chemical-physical water quality data conformed to criteria set forth in Ohio Water Quality Standards, significant differences between streams were noted in conductivity and in sodium and chloride concentrations (Table 2). Mean conductivity values and sodium and chloride levels were approximately 50% greater in Straight Fork than in Cranenest Fork. This is consistent with the greater density of EMWs in the Straight Fork watershed and the probably greater release of brines from these wells. It was beyond the scope and means of this study to determine the method of brine disposal from each well. It is assumed that brine disposal methods were similar in each watershed. One EMW believed to be representative of wells in the area and known to produce brines was sampled during this study. This well produced brine, when pumped, with a concentration of 60,000 mg/L chloride at a rate of approximately 10 barrels per month. This concentration of chloride is comparable to chloride levels noted for brine from other EMWs in the region (Ohio Geological Survey 1991).

No differences were noted in the additional chemical water quality data collected. It is especially noteworthy that nitrate levels and dissolved oxygen concentrations,

TABLE 2

Chemical-physical water quality data for Cranenest Fork and Straight Fork, May-October 1990.

	Cranenest Fork	Straight Fork
Water Temp. (°C)		
N	7	6
min	15.5	15.5
max	23.6	27.0
Conductivity (µmhos/cm)		
N	7	6
$\bar{x}$	224	385
min	100	230
max	345	505
pH (units)		
N	6	6
$\bar{x}$	7.6	7.8
min	7.0	7.3
max	8.3	8.4
Dissolved Oxygen (mg/L)		
N	6	5
$\bar{x}$	8.9	10.1
min	7.2	8.1
max	10.3	11.6
Chloride (mg/L)		
N	6	6
$\bar{x}$	18.6	34.3
min	4.6	13.0
max	39.0	74.0
Sulfate (mg/L)		
N	6	6
$\bar{x}$	31.3	28.5
min	27	19
max	36	34
Bicarbonate (mg/L)		
N	6	6
$\bar{x}$	101	171
min	44	84
max	130	204
Nitrate (mg/L)		
N	6	6
$\bar{x}$	.88	.62
min	0	0
max	2.6	2.9
Sodium (mg/L)		
N	6	6
$\bar{x}$	9.4	15.4
min	4.1	9.5
max	12.0	32.0

N = number of analyses;  $\bar{x}$  = mean; min = minimum value; max = maximum value.

which can reveal organic enrichment, were not significantly different between streams. Differential organic enrichment between streams would complicate interpretation of biological differences.

### Biological Integrity

Fifteen measures of benthic macroinvertebrate community composition and two measures each of the fish and diatom communities were made for Cranenest Fork and Straight Fork (Table 3). The major difference between biological communities of the two streams was in species composition of epilithic diatoms. The proportion of salt-tolerant diatom species and varieties was considerably greater in Straight Fork than Cranenest Fork (Table 3). Salt-tolerant forms found in Straight Fork, but not found in Cranenest Fork included *Navicula salinarum*, *N. tripunctata*, and *N. viridula* v. *avenacea*. The latter variety accounted for 11% of the diatom community, among the largest percentage composition for any taxon, in Straight Fork.

Small differences in benthic macroinvertebrate communities occurred between streams. The most notable differences were the lower number of taxa, fewer species and lower proportion of trichopterans, and larger proportion of salt-tolerant chironomids in Straight Fork. The density of insects was somewhat lower in Straight Fork and all composite index scores, Shannon-Weiner diversity, Belgian Biotic, Trent Biotic, and Invertebrate Community Index, were lower for Straight Fork. It should be emphasized that differences between streams were marginal.

Sorensen's coefficient of community similarity *C* (Sorensen 1948) between streams was 0.67, indicating a moderate difference in species composition. Complete similarity for this coefficient is one, but replicate samples from the same community seldom yield *C* values greater than 0.85 (Cox 1985).

The fish community in each stream consisted of the same five species: southern redbelly dace (*Ploxinus erythrogaster*), central striped shiner (*Notropis chrysocephalus chrysocephalus*), bluntnose minnow (*Pimephales notatus*), stoneroller minnow (*Campostoma anomalum anomalum*), barred fantail darter (*Etheostoma flabellare flabellare*). Although the number of species was small, diversity of functional types including feeding guilds and breeding guilds was relatively large. Most species collected are intolerant of organic pollutants and are sensitive to habitat disturbances such as siltation and redistribution of sediments. No deformities, eroded fins, lesions, or tumors were noted on fish from either tributary.

The most abundant species at each site was the bluntnose minnow, a hardy omnivorous species that is among the most tolerant fish of adverse water quality and habitat conditions in streams (Trautman 1981). Central striped shiners, an insectivorous species, were more numerous at Cranenest Fork. This is noteworthy because a major effect expected of brine pollution would be eliminating or reducing the numbers of insectivorous fish.

### DISCUSSION

The results of this study indicate that release of EMW brines into headwater tributaries, while not increasing the levels of dissolved chemicals above generally accepted water quality standards, nevertheless causes differences in aquatic communities. These differences were most apparent in the species composition of epilithic diatom communities. Moderate differences were noted in benthic

TABLE 3

*Measures of benthic macroinvertebrate, fish, and diatom community-structure in two headwater tributaries of the Little Muskingum River, southeastern Ohio.*

	Cranenest Fork	Straight Fork
<b>BENTHIC MACROINVERTEBRATES</b>		
1. Number of taxa	22	17
2. Number of ephemeropteran taxa	4	5
3. Number of trichopteran taxa	5	3
4. Number of dipteran taxa	8	5
5. Percent Ephemeroptera	26	29
6. Percent Trichoptera	45	25
7. Percent Tanytarsini (Chironomidae)	1	0
8. Percent Diptera and non-insects	1	21
9. Percent tolerant of organic enrichment	1	2
10. Number of EPT <sup>a</sup> taxa	11	10
11. Invertebrate Community Index (ICI) <sup>b</sup>	44	42
12. Density (number/m <sup>2</sup> )	1360	896
13. Shannon-Weiner Diversity <sup>c</sup>	3.3	3.1
14. Belgian Biotic Index <sup>d</sup>	9	7
15. Trent Biotic Index <sup>e</sup>	10	9
<b>FISH</b>		
16. Number of species	5	5
17. Index of Biotic Integrity (IBI) <sup>b</sup>	24	18
<b>DIATOMS</b>		
18. Number of taxa	25	22
19. Percent salt-tolerant forms	<1	13

<sup>a</sup>Plafkin et al. 1989; <sup>b</sup>Ohio Environmental Protection Agency 1988; <sup>c</sup>Wilhm and Dorris 1968; <sup>d</sup>DePauw and Vanhooren 1983; <sup>e</sup>Woodiwiss 1964.

macroinvertebrate communities, but no differences were detected in fish communities.

### Epilithic Diatoms

Increased proportions of salt tolerant diatoms, especially of *Navicula viridula* v. *avenacea*, in Straight Fork were the most important finding. *Navicula viridula* v. *avenacea* is a freshwater species, but is stimulated by small amounts of salt (Lowe 1974). *Navicula salinarum* is a well-known, brackish-water species (Patrick 1977). *Navicula tripunctata* has been reported from a wide range of salt concentrations (Lowe 1974).

The number of species of diatoms was not significantly different between the two sites, indicating no major impact of brines. A major impact would be expected to cause large changes in species composition and greatly reduced numbers of species (Patrick 1977). It is not known whether the replacement of oligohalobous species (freshwater forms occurring in salt concentration of less than 500 mg/L) with mesohalobous or euhalobous species (brackish-water and marine forms) significantly affects higher trophic levels in aquatic communities. Scraper/grazer organisms such as some beetle larvae, mayfly nymphs, and caddisfly larvae presumably would be affected by changes in diatom species composition, but no studies of this type have been reported. Likewise, no studies have been reported of the possible effects on herbivorous fish species. Biocriteria for diatom communities comparable to criteria based on ICI values for invertebrate

communities have not been incorporated into Ohio Water Quality Standards.

### Benthic Macroinvertebrates

Brine enrichment was expected to decrease the number of taxa and proportionally increase salt-tolerant species of benthic macroinvertebrates. Brines also were expected to lower the density of animals, especially insects, and to reduce composite index scores for Shannon-Weiner diversity, Belgian Biotic Indexes, Trent Biotic Indexes, and for Invertebrate Community Indexes. Composite index scores decrease with brine enrichment because of declining number of taxa, uneven distribution of individuals per species, and of reduction or elimination of salt-sensitive species (Hellawell 1986). Differences between streams for 12 of 15 measures of the benthic macroinvertebrate communities were consistent with expected effects of brine enrichment in Straight Fork (Table 3). Differences, however, were small, but most notable for the lower number of taxa and lower density of organisms in Straight Fork.

Shannon-Weiner diversity indexes exceeded 3.0 in both streams indicating a relatively even distribution of numbers of individuals per species and a moderate number of species, both characteristic of undisturbed communities. Shannon-Weiner indexes greater than 3.0 have been associated with relatively unpolluted waterways (Wilhm and Dorris 1968, Olive and Smith 1975).

According to the authors of the Belgian Biotic Index, the value of 9 for Cranenest Fork indicates unpolluted

water, but the value of 7 for Straight Fork indicates slightly polluted water (Depauw and Vanhooren 1983). Trent Biotic Indexes of 9 and 10 for Straight Fork and Cranenest Fork respectively, indicated "fair to good" water quality where a maximum score of 15 for excellent water quality is possible (Woodiwiss 1964). The Belgian and Trent Indexes are most sensitive to the presence of organic-pollution tolerant plecopterans and ephemeropterans, and to numbers of species. Ohio Environmental Protection Agency ICI scores of 42 and 44 for Straight Fork and Cranenest Fork exceeded the minimum score of 36 designated in Ohio Water Quality Standards for this region of Ohio as indicative of satisfactory biological integrity (Ohio Environmental Protection Agency 1989). Further perspective on the significance of ICI scores for Cranenest Fork and Straight Fork can be seen from an ICI score of 50 for a nearby tributary of Sunfish Creek, a stream with even fewer EMWs per unit of watershed (0.65) than Cranenest Fork.

### Fish Communities

Fish communities were nearly identical in the two tributaries, indicating no major effect from brine enrichment. Index of Biotic Integrity (Ohio Environmental Protection Agency 1988) values of 24 for Cranenest Fork and 18 for Straight Fork were considerably below the IBI value of 44 established in Ohio Water Quality Standards for streams in this region of Ohio (Ohio Environmental Protection Agency 1989). Although these results are consistent with expected effects of brine enrichment, the susceptibility of these headwater streams to intermittent flow regimes cast doubt on the suitability of IBI for assessing fish communities in these areas. Intermittent flow regimes may dislocate fish during floods and reduce available habitats during droughts.

The biological integrity of fish communities in Cranenest Fork and Straight Fork is consistent with reported effects of brine discharges in other rivers. The State of Kentucky, for example, has evaluated streams receiving oil well brines in northeast Kentucky (Kentucky Dept. Environmental Protection 1990). Nineteen species of fish were collected from a clean-water area of the South Fork Red River (conductivity = 300  $\mu\text{mhos/cm}$ ), but only 14 species were noted from a nearby brine-impacted site (conductivity = 5,600  $\mu\text{mhos/cm}$ ). In Ross Creek, a tributary of the Kentucky River, 11 species of fish, mostly cyprinids and darters, were collected from a clean-water area (conductivity = 350  $\mu\text{mhos/cm}$ ). No fish were observed or collected from the most heavily brine-impacted area (conductivity >20,000  $\mu\text{mhos/cm}$ ), but 11 species were noted at a moderately brine-impacted site with conductivity values of approximately 9,500  $\mu\text{mhos/cm}$  (Kentucky Division of Environmental Services 1982).

### CONCLUSIONS

The cause(s) of altered aquatic communities in Straight Fork, e.g., lower density of benthic insects and increased proportions of salt-tolerant diatoms, could not be unambiguously determined. Intermittent brine releases, as expected in this region, probably are the major cause. The larger number of EMWs per  $\text{km}^2$  of watershed than in Cranenest Fork, a lower density of benthic insects, and

increased proportions of salt-tolerant diatoms in Straight Fork are the major pieces of evidence supporting this hypothesis. Although flooding could have reduced the numbers of insects and fish, it could not have increased the proportion of salt-tolerant diatoms.

If brines are responsible for biological alterations in Straight Fork, it cannot be conclusively demonstrated that brines originated from Exempt Mississippian Wells. Stream segments without EMWs could not be located and used as reference sites. As noted earlier the concentrations of inorganic chemicals were so low that a distinctive "fingerprint" of EMW brines could not be detected in the ambient water.

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### LITERATURE CITED

- American Public Health Association 1989 Standard Methods for the Examination of Water and Wastewater. 16th ed. Am. Public Health Assn., Am. Waterworks Assn., and Water Pollution Control Fed. Washington, DC.
- Bownocker, J. A. 1920 Geologic Map of Ohio. Ohio Div. Geol. Survey. Columbus, OH.
- Breen, K. J., C. G. Angelo, R. W. Masters, and A. C. Sedam 1985 Chemical and isotopic characteristics of brines from three oil- and gas-producing sandstones in eastern Ohio, with applications to the geochemical tracing of brine sources. Water-Resources Investigation Report 84-4314. U.S. Geological Survey, Columbus, OH. 53 pp.
- Collins, H. R. and B. E. Smith 1977 Geology and mineral resources of Washington County, Ohio. Ohio Div. Geol. Survey, Bull. 66. 134 pp.
- Cox, G. W. 1985 Laboratory Manual of General Ecology. Wm. C. Brown Publ., Dubuque, IA. 248 pp.
- DePauw, N. and G. Vanhooren 1983 Method for biological quality assessment of watercourses in Belgium. *Hydrobiologia* 100: 153-168.
- Hayhurst, E. N., T. N. Rubel, G. E. Kelley, and P. Beining 1974 Soil survey of Monroe County, Ohio. U.S. Dept. of Agriculture, Soil Conservation Service. 123 pp.
- Hellawell, J. M. 1986 Biological Indicators of Freshwater Pollution and Environmental Management. Elsevier Applied Science Publishers, Barking, England. 546 pp.
- Hester, F. E. and J. S. Dendy 1962 A multiple-plate sampler for aquatic macroinvertebrates. *Trans. Am. Fish. Soc.* 91: 420-421.
- Karr, J. R. 1981 Assessment of biotic integrity using fish communities. *Fisheries* 6: 21-27.
- Kentucky Division of Environmental Services 1982 Ross Creek drainage biological and water quality investigation. KY Dept. Nat. Res., Div. of Water, Biol. Branch. Tech. Rept. No. 1. 48 pp.
- Kentucky Department of Environmental Protection 1990 South Fork Red River drainage biological and water quality investigation for stream use designation. KY Dept. Nat. Res., Div. of Water, Ecological Support Section. Tech Rept. No. 26. 55 pp.
- Lowe, R. L. 1974 Environmental requirements and pollution tolerance of freshwater diatoms. EPA-670/4-74-005. National Env. Research Center, U.S. Env. Prot. Agency, Cincinnati, OH. 334 pp.
- Merritt, R. W. and K. W. Cummins (eds.) 1984 An Introduction to the Aquatic Insects of North America. 2nd ed. Kendall/Hunt Publ. Co., Dubuque, IA. 722 pp.
- Ohio Environmental Protection Agency 1987 State of Ohio Water Quality Standards. Chapter 3745-1 of the Adm. Code. Ohio Env. Prot. Agency, Div. Water Quality Monitoring and Assessment, Columbus, OH.
- \_\_\_\_\_ 1988 Biological criteria for the protection of aquatic life. Vol. II. Users manual for biological field assessment of Ohio surface waters. Doc. 0046e/0013e. Ohio Env. Prot. Agency, Columbus, OH.
- \_\_\_\_\_ 1989 Addendum to: Biological criteria for the protection of aquatic life. Vol. II. Users manual for biological field assessment of

- Ohio surface waters. Ohio Env. Prot. Agency, Columbus, OH. 21 pp.
- Ohio Geological Survey 1991 Open-file analyses of brines from exempt Mississippian oil and gas wells in Ohio. Ohio Division of Geological Survey, Mineral Resources and Geochemistry Section, Columbus, OH.
- Olive, J. H. and K. R. Smith 1975 Benthic macroinvertebrates as indexes of water quality in the Scioto River basin, Ohio. Bulletin of the Ohio Biological Survey, Vol. 5. The Ohio State Univ. Press, Columbus, OH. 124 pp.
- Patrick, R. 1977 Ecology of freshwater diatoms and diatom communities. *In*: D. Werner (ed.), The Biology of Diatoms. Univ. of California Press, Berkeley, CA. pp. 284-332.
- \_\_\_\_\_ and C. W. Reimer 1966 The Diatoms of the United States. Vol. 1. Acad. Nat. Sci. Philadelphia, Monogr. 13. 688 pp.
- \_\_\_\_\_ and C. W. Reimer 1975 The Diatoms of the United States. Vol. 2, part I. Acad. Nat. Sci. Philadelphia, Monogr. 13. 213 pp.
- Peckarsky, B. L., P. R. Fraissinet, M. A. Penton, and D. J. Conklin, Jr. 1990 Freshwater Macroinvertebrates of Northeastern North America. Cornell Univ. Press, Ithaca, NY. 442 pp.
- Plafkin, J. L., M. T. Barbour, K. D. Porter, S. K. Gross, and R. M. Hughes 1989 Rapid bioassessment protocols for use in streams and rivers. Environmental Protection Agency/444/4-89-001. U.S. Env. Prot. Agency, Washington, DC.
- Prater, B. L., D. R. Barton, and J. H. Olive 1977 New sampler for shallow-water benthic macroinvertebrates. The Progressive Fish Culturist 39: 58.
- Sorensen, T. 1948 A method of establishing groups of equal amplitude in plant sociobiology based on similarity of species content and its application to analyses of the vegetation of Danish commons. K. danske vidensk. Selsk. 5: 1-34.
- Stauffer, C. R. and C. R. Schroyer 1920 The Dunkard Series of Ohio. Bull. 22. Ohio Geological Survey. 167 pp.
- Thornbury, W. D. 1965 Regional geomorphology of the United States. John Wiley and Sons, New York, NY. 609 pp.
- Trautman, M. B. 1981 The Fishes of Ohio. Ohio State Univ. Press, Columbus, OH. 782 pp.
- United States Environmental Protection Agency 1984 Test method: The determination of inorganic anions in water by ion chromatography method 300.0. EPA 600/4/84-017. U.S. Env. Prot. Agency, Washington, DC.
- \_\_\_\_\_ 1986 Quality criteria for water. EPA 440/5-86-001. U.S. Env. Prot. Agency, Washington, DC. 256 pp.
- U.S. Forest Service 1990 Little Muskingum River: Eligibility determination for the Wild and Scenic River Systems. Spec. Report. Wayne National Forest, Athens, OH.
- U.S. Geological Survey 1989 Water resources data, Ohio. Water Year 1988. Vol. 1, Ohio River Basin. U.S. Geological Survey, Columbus, OH. 298 pp.
- Wilhm, J. L. and T. C. Dorris 1968 Biological parameters for water quality criteria. Bioscience 18: 477-481.
- Woodiwiss, F. A. 1964 A biological system of stream classification used by the Trent River Board. Chem. Ind. Lond. 11: 443-447.