## **Standard Operating Procedures**

## Laboratory Processing of Benthic Samples

Upon arrival at the laboratory, the Hester-Dendy (H-D), Surber, and qualitative samples were logged in and accounted for. Prior to sorting and identification, each sample was rinsed on a No. 60 (0.250 mm openings) U.S. Standard Testing Sieve to remove the preservative and the H-D plates were scraped to remove the organisms. Sorting of each H-D and Surber sample was conducted in a white enamel pan first under a magnifier lamp and finally under a compound dissecting scope. If necessary, a Folsum sample splitter was used to subsample the H-Ds and Surbers until a more manageable number of organisms was achieved. Prior to splitting, the sample was pre-picked to remove any large and/or rare taxa. In all a minimum of 250 organisms in representative proportions were removed from the fractionated samples. Organisms were sorted to higher taxonomic levels (generally Class or Order level) and preserved separately in labeled vials containing 70% ethyl alcohol. To assure a consistent level of quality and sorting efficiency, senior EA personnel checked all samples. The qualitative samples contained very little detrital matter and therefore were simply rinsed prior to identification.

Macroinvertebrate identifications were made to the lowest practical taxonomic level using the most current literature available (see attached list of taxonomic literature). Whenever possible, the level of identifications followed those recommended by Mr. Jeffery DeShon of the Ohio EPA (pers. comm., 1998). Chironomidae larvae were cleared in warm 10% potassium hydroxide and mounted in CMC-10 prior to identification. Generally, 100 chironomids from any single sample were mounted for identification. For all sample types, specimens were enumerated, coded and recorded on a standard laboratory bench sheet for data processing.

## Data Analyses

The Invertebrate Community Index (ICI) was used as the principal measure of overall macroinvertebrate community condition. Developed by the Ohio EPA, the ICI is a modification of the Index of Biotic Integrity for fish (Ohio EPA 1987). The ICI consists of ten individually scored structural community metrics:

- 1. Total number of taxa
- 2. Total number of mayfly taxa
- 3. Total number of caddisfly taxa
- 4. Total number of dipteran taxa
- 5. Percent mayflies

- 6. Percent caddisflies
- 7. Percent Tanytarsini midges
- 8. Percent other dipterans and non-insects
- 9. Percent tolerant organisms
  - 10. Total number of qualitative EPT taxa.

Scoring criteria for all ten metrics is dependent upon drainage area. The scoring of an individual sample was based on the relevant attributes of that sample compared to equivalent data from 232 reference sites throughout Ohio. Metric scores range from six points for values comparable to exceptional community structure to zero points for values that deviate strongly from the expected range of values based on scoring criteria established by Ohio EPA (1989a). The sum of the individual metric scores resulted in the ICI score for that particular location.

Calculation of the ICI was conducted using a computer program written for the software SAS<sup>®</sup> by EA in 1994. This program is continuously tested and updated to ensure its accuracy. Although the ICI is most often used to analyze the combination of H-D and qualitative data from a particular location, for this study, the ICI was also calculated for the combination of Surber and qualitative data when H-D data were not available.

The only other statistical comparison used was the relative abundance (or percent composition) of individual taxa from each site and sample type. Relative abundance was calculated for both sample types as:

Rel. Abund.(%)= 100 X <u># Individuals of a Taxa</u> Total # of Individuals in Sample

All sample processing and data analysis were completed by permanent and full-time EA Engineering, Science, & Technology, Inc. staff working our Deerfield, Illinois office and laboratory. Specific staff members that worked on this project and relevant experience are listed below by task:

		<b>Years of</b>
<u>Task</u>	EA Personnel	<b>Experience</b>
Login	Sarah Olson	1.5
Sorting	Nick Wood	1
Chironimid Mounting	Paul Hauser	2
	Sarah Olsen	1.5
Identification	Marty Sneen	17
Data Analysis	Matt Poore	1
	Joe Vondruska	22
	Marty Sneen	17

### **Selected Ohio EPA Reporting Requirements**

#### Item 12-Taxonomic literature

Although EA's taxonomic library contains substantially more references than are listed here, the following list only includes taxonomic literature used to identify the benthos in samples from Big Creek, Doan Brook, Euclid Creek, and Mill Creek.

- Bednarik, A.F. and W.P. McCafferty. 1979. Biosystematic revision of the genus <u>Stenonema</u> (Ephemeroptera: Heptageniidae). Canadian Bulletins of Fisheries and Aquatic Sciences 201:1-73.
- Bode, R.W. 1983. Larvae of North American <u>Eukiefferiella</u> and <u>Tvetenia</u> (Diptera: Chironomidae). New York State Museum Bulletin 452:1-40.
- Bolton, M.J. 1998. Guide to the identification of larval Chironomidae (Diptera) in the temperate eastern Nearctic north of Florida. Ohio EPA, Division of Surface Water,

Ecological Assessment Section, Columbus, Ohio.

Brown, H.P. 1976. Aquatic dryopoid beetles (Coleoptera) of the United States. Water Pollution Control Series 18050 ELDO4/72. 2nd edition. U.S. Environmental Protection Agency, Cincinnati, OH.

Burch, J.B. 1982. Freshwater snails (Mollusca: Gastropoda) of North America. EPA-600/3-82-026. U.S. Environmental Protection Agency, Environmental Monitoring and Support Laboratory, Cincinnati, OH.

Epler, J.H. 1987. Revision of the Nearctic <u>Dicrotendipes</u> Kieffer, 1913 (Diptera: Chironomidae). Evolutionary Monographs No. 9:1-102.

\_\_\_\_\_. 1995. Identification manual for the larval Chironomidae (Diptera) of Florida. Florida DEP, Division of Water Facilities, Tallahassee, FL.

\_\_\_\_\_\_. 2001. Identification manual for the larval Chironomidae (Diptera) of North and South Carolina. North Carolina DENR, Division of Water Quality, Raleigh, NC.

Grodhaus, G. 1987. <u>Endochironomus</u> Kieffer, <u>Tribelos</u> Townes, <u>Synendotendipes</u> new genus, and <u>Endotribelos</u> new genus (Diptera: Chironomidae) of the Nearctic region. Journal of the Kansas Entomological Society 60(2):167-247.

Jezerinac, R.F., G.W. Stocker, and D.C. Tarter. 1995. The crayfishes (Decapoda: Cambaridae) of West Virginia. Bulletin of the Ohio Biological Survey 10(1):1-193.

- Klemm, D.J. 1985. Guide to the freshwater Annelida (Polychaeta, naidid, and tubificid Oligochaeta, and Hirudinea) of North America. Kendall/Hunt Publishing Co., Dubuque, IA.
- Larson, D.J., Y. Alarie, and R.E. Roughley. 2000. Predaceous Diving Beetles (Coleoptera: Dytiscidae) of the Nearctic Region: with emphasis on the fauna of Canada and Alaska. NRC Research Press, Ottawa, Canada.
- Maschwitz, D.E. 1976. Revision of the Nearctic species of the subgenus <u>Polypedilum</u> (Chironomidae: Diptera). Doctoral Dissertation, University of Minnesota.
- McCafferty, W.P. and R.D. Waltz. 1990. Revisionary synopsis of the Baetidae (Ephemeroptera) of North and Middle America. Transactions of the American Entomological Society 116(4):769-799.

Merritt, R.W. and K.W. Cummins, eds. 1996. An introduction to the aquatic insects of North America. 3rd edition. Kendall/Hunt Publishing Co., Dubuque, IA.

Morihara, D.K. and W.P. McCafferty. 1979. The Baetis larvae of North America

(Ephemeroptera: Baetidae). Transactions of the American Entomological Society 105:139-221.

Needham, J.G. and M.J. Westfall, Jr. 1955. A manual of the dragonflies of North America (Anisoptera) including the Greater Antilles and the provinces of the Mexican border. University of California Press, Berkeley, California.

Pennak, R.W. 1989. Fresh-water invertebrates of the United States. 2nd edition. John Wiley & Sons, New York, NY.

- Roback, S.S. 1985. The immature chironomids of the eastern United States VI. Pentaneurini-genus <u>Ablabesmyia</u>. Proceedings of The Academy of Natural Sciences of Philadelphia 137(2):153-212.
- Saether, O.A. 1977. Taxonomic studies on Chironomidae: <u>Nanocladius</u>, <u>Pseudochironomus</u>, and the <u>Harnischia</u> complex. Bulletin of the Fisheries Research Board of Canada 196:1-143.
- Simpson, K.W. and R.W. Bode. 1980. Common larvae of the Chironomidae (Diptera) from New York State streams and rivers with particular reference to the fauna of artificial substrates. New York State Museum Bulletin 439:1-105.
- Wiederholm, T., ed. 1983. Chironomidae of the Holarctic region. Keys and diagnoses. Part 1. Larvae. Entomologica Scandinavica Supplement 19:1-457.
- Wiggins, G.B. 1996. Larvae of the North American caddisfly genera (Trichoptera). 2nd edition. University of Toronto Press, Toronto, Canada.

### Item 13-Reference Collection

A reference collection was not necessary for identification of these specimens. However, if a reference collection had been needed to verify any specimens, EA maintains a sizable macroinvertebrate voucher collection with over 1800 specimens representing over 700 taxa. If this taxonomic library proved to be insufficient, every reasonable attempt would be made to have the specimen(s) identified or verified by a noted authority.

### Item 16-Chironomidae Identification

Chironomidae larvae were cleared in warm 10% potassium hydroxide and mounted in CMC-10 prior to identification. Generally, 100 chironomids from any single sample are mounted for identification. Species level identifications generally follow those suggested by Ohio EPA. Item 17-Copies of Raw Data

Copies of the laboratory bench sheets are appended to the hard copy of this document.

### Item 18-ICI Calculation

The Invertebrate Community Index (ICI) was used as the principal measure of overall macroinvertebrate community condition. Developed by the Ohio EPA, the ICI is a modification of the Index of Biotic Integrity for fish (Ohio EPA 1987). The ICI consists of ten individually scored structural community metrics:

1. Total number of taxa

5. Percent mayflies

6. Percent caddisflies

- 2. Total number of mayfly taxa
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- 4. Total number of dipteran taxa
- 8. Percent other dipterans and non-insects
- 9. Percent tolerant organisms
- 7. Percent Tanytarsini midges
- 10. Total number of qualitative EPT taxa.

Scoring criteria for all ten metrics is dependent upon drainage area. The scoring of an individual sample was based on the relevant attributes of that sample compared to equivalent data from at least 232 reference sites throughout Ohio. Metric scores range from six points for values comparable to exceptional community structure to zero points for values that deviate strongly from the expected range of values based on scoring criteria established by Ohio EPA (1989a). The sum of the individual metric scores resulted in the ICI score for that particular location.

Calculation of the ICI was conducted using a computer program written for the software SAS® by EA in 1994. This program is continuously tested and updated to ensure its accuracy.

## Item 20-Statistical Analyses

The only other statistical comparison used was the relative abundance (or percent composition) of individual taxa per site and sample type. Relative abundance was calculated for both sample types as:

Rel. Abund.= <u># Individuals of a Taxa</u> Total # of Individuals in Sample

# Item 21-Results

Complete results are appended to the hard copy of this document.

## Item 25-Electronically Formatted Data

For convenience, the data and text are provided in electronic format as Word 2003<sup>®</sup> files via email and on the enclosed CD-RW.