

# *AQUATIC RESOURCES CENTER, INC*

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08 April 2014

Mr. Josh Upham  
Murfreesboro Water and Sewer  
Storm Water Program  
220 NW Broad Street  
Murfreesboro, TN 37130

**RE:** Identification and Enumeration of Benthic Invertebrates  
Murfreesboro Water and Sewer Purchase Order No. 21937  
Aquatic Resources Center, Inc Project No. 672

Dear Mr. Upham,

Attached please find the results for the above referenced project. Habitat assessments show no impairment at all stations except Garrison Creek which showed impairment to the habitat. The total number of individuals removed ranged from 182 to 224 individuals per sample. A total of 70 taxa was identified with a range of 13 to 38 taxa per sample. Tennessee Macroinvertebrate scores (TMI) ranged from 14 to 34 with four scores not meeting the biocriteria guideline of 32 or higher.

If you have any questions or comments regarding these data, please contact me at your earliest convenience.

Thank you very much.

Sincerely yours,



Todd W. Askegaard  
President

Aquatic Resources Center, Inc. collected five benthic invertebrate samples on 24 March 2014 for Murfreesboro Water and Sewer Department for benthic invertebrate analysis. The samples were collected from West Fork Stones River (two sites), Sinking Creek, Garrison Creek and Lytle Creek. These streams are located in Ecoregion 71i. The drainage areas of the streams are greater than 2 square miles except for Garrison Creek (1.50 square miles). Below is a description of the methods and presentation of the data.

Samples were collected according to Tennessee Department of Environment and Conservation (TDEC rev. 2011) and USEPA (Barbour *et al.* 1999) protocols, with two different methods used due to the stream widths of the various sites. In larger streams, a two-person square meter kick net with a 500 micron mesh was used to sample in the riffle area and the run area. In smaller streams, the modified method using an 18 inch kicknet (with a 500- $\mu$ m mesh collecting net) was used to collect approximately 1 m<sup>2</sup> of material from the riffle area, and one 1 m<sup>2</sup> from the run area, for a total coverage area of approximately 2 m<sup>2</sup>. The sediment samples from the riffle and run at a site were combined, washed in a 500- $\mu$ m mesh sieve to remove excess sediment, emptied into a labeled heavy-duty plastic jar, and fixed with 95 percent ethanol. The samples were placed into a cooler for transport to the Aquatic Resources laboratory in Nashville.

In conjunction with the benthic invertebrate sampling, habitat assessments were performed at each site, following the procedures of Barbour *et al.* (1999) and TDEC (rev. 2011) (Table 1). Parameters such as substratum, embeddedness, velocity, depth, bank characteristics and land use were assessed and rated to determine if and to what extent the habitat is capable of supporting a diverse benthic community. In addition, other site characteristics such as sketches of the site, important features, weather, sampling personnel, methods, and other aquatic life were noted in ink in a dedicated, paginated logbook. Photographs of each site were taken. Water quality measurements were taken for dissolved oxygen, pH, conductivity and temperature using an Orion 5-Star multi-parameter portable meter. Water velocity and depth at each specific sampling location were measured using a Global Water Model FP101 flow probe (all meters were calibrated prior to use). Water quality measurements, velocity and depth, and substratum are presented in Table 2.

Each sample was washed using a US Series No. 35 (500  $\mu$ m mesh) sieve to remove ethanol and excess detritus. Because each sample contained a large amount of material (detritus and organisms), it was subsampled using the Caton (1991) method, which is recommended by TDEC and USEPA. This procedure consists of dividing the sample into 30 equal portions (termed grids) using a specified subsampling device, then sorting at least four of these grids (which have been randomly selected) to obtain 200  $\pm$  20 percent (160-240) organisms. If the sorting of a grid had been started, it was finished in its entirety. The benthic organisms removed from the sample were placed by major groupings (e.g., mayflies, worms, snails) into glass vials containing 70 percent EtOH (ethyl alcohol). Each vial was labeled with information such as date of collection, location, specific sample identification, name of taxonomic group and number of organisms. The residue from the sorted portion of the sample was preserved separately from the portion that was not sorted.

All information (site, number sorted, number mounted, identifications, and notes) was entered onto laboratory bench sheets.

Organisms were identified using either a dissecting or compound microscope. The latter microscope was used for identifying chironomids (midgefly larvae and pupae) and oligochaetes (aquatic segmented worms) after these organisms were mounted on microscope slides using CMCP mounting medium. Most organisms were identified to the generic level, unless the specimens were too small or damaged to allow identification to this level.

The sample from West Fork Stones River 2 was checked for Quality Assurance/Quality Control (QA/QC) on sorting efficiency. The sample passed the check with 95 percent efficiency (11 qc organisms/224 total organisms). The sample from West Fork Stones River 2 was selected for the QA/QC check on taxonomic accuracy. The comparison of identifications between two taxonomists showed no significant difference (total chi-sq. = 20.0) using a chi-square test ( $\alpha = 0.05$ , d.f. = 38, critical chi-sq. = 25.2).

A phylogenetic list of taxa shows that a total of 70 taxa was identified from the five samples (Table 3). The number of taxa per sample ranged from 13 to 38. Total number of individuals removed from a sample ranged from 182 to 224. Tolerance values ranged from 1.20 (*Dipheter*) to 9.5 (*Limnodrilus*, *Varichaetadrilus*, Tubificinae: bifid chaetae). The genus, *Varichaetadrilus*, does not appear in the Tennessee Taxa List 2011 (TDEC 2011). The tolerance value assigned to this taxon was an average of values assigned other Tubificinae with bifid chaetae. A total of 18 taxa were designated as having Clinger habit and 10 taxa as Nutrient Tolerant. These data were used in the calculations of the biological measures required by TDEC (2011) and the values for these measures are shown in the table below.

Values for Biological measures.

<b>Biological Measure</b>	<b>W.F. Stones River 1</b>	<b>W.F. Stones River 2</b>	<b>Sinking Creek</b>	<b>Garrison Creek</b>	<b>Lytle Creek</b>
Taxa Richness	38	32	34	13	20
EPT Richness	7	9	7	2	4
Percent EPT – Cheumatopsyche	8.2	14.7	10.5	1.1	2.1
Percent Oligochaeta + Chironomidae	47.8	25.4	42.5	15.5	88.5
Percent Clinger	34.1	64.3	19.0	10.2	46.9
Percent Nutrient Tolerant	66.5	67.4	50.5	90.9	90.6
NCBI	5.59	4.96	5.98	7.48	6.58

Scores for Biological measures.

<b>Biological Measure</b>	<b>W.F. Stones River 1</b>	<b>W.F. Stones River 2</b>	<b>Sinking Creek</b>	<b>Garrison Creek</b>	<b>Lytle Creek</b>
Taxa Richness	6	6	6	4	4
EPT Richness	4	6	4	2	2
Percent EPT – Cheumatopsyche	0	2	0	0	0
Percent Oligochaeta + Chironomidae	4	6	4	6	0
Percent Clinger	4	6	2	0	6
Percent Nutrient Tolerant	2	2	4	0	0
NCBI	6	6	4	2	4
<b>TMI Score</b>	<b>26</b>	<b>34</b>	<b>24</b>	<b>14</b>	<b>16</b>

**TABLE 1**

**HABITAT ASSESSMENTS FOR FIVE SAMPLING SITES,  
RUTHERFORD COUNTY, TENNESSEE, 24 MARCH 2014**

Habitat Parameter (High Gradient Stream)	Values				
	W.F. Stones River 1	W.F. Stones River 2	Sinking Creek	Garrison Creek	Lytle Creek
Epifaunal substrate/available cover	17	17	15	10	15
Embeddedness	15	16	16	11	15
Velocity/depth regime	15	15	15	8	16
Sediment deposition	16	16	17	15	16
Channel flow status	17	17	18	15	18
Channel alteration	17	15	15	11	15
Frequency of riffles (or bends)	17	16	14	8	14
Bank stability (Left bank/Right bank)	6/6	8/8	7/7	6/6	7/7
Vegetative protection (Left bank/Right bank)	6/6	7/7	5/5	5/5	5/5
Riparian vegetative zone width (Left bank/Right bank)	9/7	4/4	3/3	3/3	5/3
<b>Total Score</b>	154	150	140	106	141
<b>Assessment</b> <sup>3</sup>	Not impaired	Not impaired	Not impaired	Impaired	Not impaired

The following scores indicate the assessment for each habitat parameter:  
20 - 16 optimal      15-11 suboptimal      10 - 6 marginal      5-0 poor

(These individual parameter scores vary for the last three parameters where left and right banks are assessed.)

Using EPA values, the following total scores indicate these same assessment categories:  
200 - 166 optimal      153-113 suboptimal      100 - 60 marginal      47-0 poor

<sup>3</sup> Using TDEC (2011) guidelines for Ecoregion 71i (high gradient), the following total score indicates habitat assessment:  $\geq 121$  not impaired (streams  $> 2$  sq. miles),  $\geq 119$  not impaired (streams  $\leq 2$  sq. miles).

**TABLE 2**

**WATER QUALITY MEASUREMENTS, CURRENT VELOCITY, WIDTH, DEPTH AND SUBSTRATUM, RUTHERFORD COUNTY, TENNESSEE, 24 MARCH 2014**

Parameter	W.F. Stones River 1	W.F. Stones River 2	Sinking Creek	Garrison Creek u/s	Garrison Creek d/s	Lytle Creek
Temperature (°C)	12.0	12.2	10.1	15.6	15.5	14.0
pH (standard units)	7.97	8.93	8.41	8.22	8.19	8.02
Conductivity (µS/cm)	374	368	465	493	491	361
Dissolved oxygen (mg/L)	10.01	9.30	9.45	12.85	12.84	12.54

**WIDTH, WATER VELOCITY, AND WATER DEPTH**

Parameter	W.F. Stones River 1		W.F. Stones River 2		Sinking Creek		Garrison Creek		Lytle Creek	
Approximate width of creek (m)	18.6		11.6		7.3		rifle 6.1	run 3.0	14.3	
Water velocity (m/sec) <sup>2</sup>	rifle 0.11	run 0.11	rifle 0.34	run 0.19	rifle 0.14	run 0.07	0.08	0.02	rifle 0.12	run 0.04
Water depth (m)	0.43	0.33	0.41	0.43	0.23	0.20	0.10	0.20	0.20	0.41

**SUBSTRATUM (percent composition)**

	W.F. Stones River 1	W.F. Stones River 2	Sinking Creek	Garrison Creek	Lytle Creek
Bedrock	0	10	0	0	10
Boulders	5	10	20	0	35
Cobble	30	40	35	50	25
Pebbles	30	10	20	20	10
Granules	20	5	10	20	10
Sand	10	15	10	5	5
Silt/Clay	5	10	5	5	5

TABLE 3

## PHYLOGENETIC LIST OF TAXA AND ABUNDANCE OF BENTHIC INVERTEBRATES, MRFREESBORO, TN, 24 March 2014

Phylum	Class	Order	Family	Taxa	WF1	WF2	Sinking	Garrison	Lytle	Tolerance value	Clinger	Nutrient tolerant
Nematoda				Nematoda		2				6.00		
Platyhelminthes	Turbellaria	Tricladida	Dugesiidae	Dugesiidae	1		4			6.48		
Annelida	Oligochaeta	Tubificida	Naididae	<i>Branchiura</i>	2					8.28		
Annelida	Oligochaeta	Tubificida	Naididae	<i>Limnodrilus</i>	1	1			1	9.50		
Annelida	Oligochaeta	Tubificida	Naididae	<i>Nais</i>	20	11	17	2	70	8.88		x
Annelida	Oligochaeta	Tubificida	Naididae	<i>Ophidonais</i>	2					2.00		
Annelida	Oligochaeta	Tubificida	Naididae	<i>Slavina</i>	1				1	7.06		
Annelida	Oligochaeta	Tubificida	Naididae	<i>Varichaetadrilus</i>		3				9.50		
Annelida	Oligochaeta	Tubificida	Naididae	Tubificinae: bifid chaetae	1	2			11	9.50		x
Arthropoda	Crustacea	Amphipoda	Crangonyctidae	<i>Crangonyx</i>		1	1	2		7.87		
Arthropoda	Crustacea	Amphipoda	Crangonyctidae	Crangonyctidae				1		7.87		
Arthropoda	Crustacea	Amphipoda	Hyalellidae	<i>Hyalella</i>	1					7.75		
Arthropoda	Crustacea	Isopoda	Asellidae	<i>Lirceus</i>	18		58	109	1	7.85		x
Arthropoda	Crustacea	Isopoda	Asellidae	Asellidae				42		7.85		x
Arthropoda	Crustacea	Decapoda	Cambaridae	Cambarinae	1					7.50		
Arthropoda	Arachnida	Acariformes		Acariformes			4			5.53		
Arthropoda	Insecta	Ephemeroptera	Baetidae	<i>Acerpenna</i>	3	1	4	1	1	3.70		
Arthropoda	Insecta	Ephemeroptera	Baetidae	<i>Baetis</i>			1			4.51		
Arthropoda	Insecta	Ephemeroptera	Baetidae	<i>Dipheter</i>			4			1.20		
Arthropoda	Insecta	Ephemeroptera	Caenidae	<i>Caenis</i>	3					7.41		x
Arthropoda	Insecta	Ephemeroptera	Ephemerellidae	<i>Teloganopsis</i>		2				1.57	x	
Arthropoda	Insecta	Ephemeroptera	Heptageniidae	Heptageniidae		9	1		1	4.00	x	
Arthropoda	Insecta	Ephemeroptera	Heptageniidae	<i>Maccaffertium</i>	3	15	7			3.15	x	
Arthropoda	Insecta	Ephemeroptera	Heptageniidae	<i>Stenacron</i>	3	1	3		1	3.58	x	
Arthropoda	Insecta	Ephemeroptera	Isonychiidae	<i>Isonychia</i>	1	1	1			3.45		
Arthropoda	Insecta	Odonata	Coenagrionidae	<i>Argia</i>	3	2	2			8.17		
Arthropoda	Insecta	Plecoptera	Perlodidae	<i>Isoperla</i>				1		1.50	x	
Arthropoda	Insecta	Trichoptera	Hydropsychidae	<i>Cheumatopsyche</i>	1		9		8	3.22	x	x
Arthropoda	Insecta	Trichoptera	Hydropsychidae	<i>Macrostemum</i>		1				3.52	x	
Arthropoda	Insecta	Trichoptera	Hydroptilidae	<i>Hydroptila</i>	1					6.22	x	
Arthropoda	Insecta	Trichoptera	Leptoceridae	<i>Nectopsyche</i>		1				2.94		
Arthropoda	Insecta	Lepidoptera	Pyralidae	<i>Petrophila</i>	1					2.09	x	
Arthropoda	Insecta	Trichoptera	Philopotamidae	<i>Chimarra</i>		2			1	2.76	x	
Arthropoda	Insecta	Coleoptera	Psephenidae	<i>Psephenus</i>		2	1			2.35	x	
Arthropoda	Insecta	Coleoptera	Elmidae	<i>Dubiraphia</i>		3				5.93	x	
Arthropoda	Insecta	Coleoptera	Elmidae	<i>Microcylloepus</i>		1				2.11	x	
Arthropoda	Insecta	Coleoptera	Elmidae	<i>Stenelmis</i>	23	89	10		3	5.10	x	x
Arthropoda	Insecta	Coleoptera	Dytiscidae	Hydroporinae				1		5.50		
Arthropoda	Insecta	Diptera	Chironomidae	<i>Ablabesmyia</i>	2					7.20		

TABLE 3

PHYLOGENETIC LIST OF TAXA AND ABUNDANCE OF BENTHIC INVERTEBRATES, MRFREESBORO, TN, 24 March 2014

Phylum	Class	Order	Family	Taxa	WF1	WF2	Sinking	Garrison	Lytle	Tolerance value	Clinger	Nutrient tolerant
Arthropoda	Insecta	Diptera	Chironomidae	<i>Cardiocladius</i>		2				5.87		
Arthropoda	Insecta	Diptera	Chironomidae	<i>Cladotanytarsus</i>	4					4.09		
Arthropoda	Insecta	Diptera	Chironomidae	<i>Conchapelopia</i>	4		2		4	4.50		
Arthropoda	Insecta	Diptera	Chironomidae	<i>Corynoneura</i>			2	2	2	6.01		
Arthropoda	Insecta	Diptera	Chironomidae	<i>Cricotopus</i>			1	4	1	5.78	x	x
Arthropoda	Insecta	Diptera	Chironomidae	<i>Cricotopus/Orthocladius</i>	29	15	1	13	73	4.86	x	x
Arthropoda	Insecta	Diptera	Chironomidae	<i>Cryptochironomus</i>	1					6.40		
Arthropoda	Insecta	Diptera	Chironomidae	<i>Dicrotendipes</i>	1			4		8.10		
Arthropoda	Insecta	Diptera	Chironomidae	<i>Eukiefferiella</i>	2	4	22			3.43		
Arthropoda	Insecta	Diptera	Chironomidae	<i>Microtendipes</i>			2			5.53		
Arthropoda	Insecta	Diptera	Chironomidae	<i>Nilotanypus</i>			2		1	3.90		
Arthropoda	Insecta	Diptera	Chironomidae	<i>Parakiefferiella</i>			2			5.40		
Arthropoda	Insecta	Diptera	Chironomidae	<i>Parametriocnemus</i>			6			3.65		
Arthropoda	Insecta	Diptera	Chironomidae	<i>Phaenopsectra</i>	1					6.50	x	
Arthropoda	Insecta	Diptera	Chironomidae	<i>Polypedilum</i>	2	16	4		5	5.69		x
Arthropoda	Insecta	Diptera	Chironomidae	<i>Pseudochironomus</i>	3	1				5.36		
Arthropoda	Insecta	Diptera	Chironomidae	<i>Rheocricotopus</i>			15			7.30		
Arthropoda	Insecta	Diptera	Chironomidae	<i>Rheotanytarsus</i>			4	1	1	5.89	x	
Arthropoda	Insecta	Diptera	Chironomidae	<i>Stempellinella</i>	5	1				4.62		
Arthropoda	Insecta	Diptera	Chironomidae	<i>Tanytarsus</i>	5		1	2		6.76		
Arthropoda	Insecta	Diptera	Chironomidae	<i>Thienemanniella</i>			3			5.86		
Arthropoda	Insecta	Diptera	Chironomidae	<i>Thienemannimyia grp</i>		1				6.20		
Arthropoda	Insecta	Diptera	Chironomidae	<i>Tvetenia</i>	1		1	1		3.65		
Arthropoda	Insecta	Diptera	Empididae	<i>Hemerodromia</i>			1		1	7.57		
Arthropoda	Insecta	Diptera	Simuliidae	<i>Prosimulium</i>		4			1	4.01	x	
Arthropoda	Insecta	Diptera	Simuliidae	<i>Simulium</i>			1			4.00	x	
Arthropoda	Insecta	Diptera	Tipulidae	<i>Tipula</i>			1			7.33		
Mollusca	Bivalvia	Veneroidea	Corbiculidae	<i>Corbicula</i>	2	2	1		1	6.12		
Mollusca	Bivalvia	Veneroidea	Sphaeriidae	<i>Pisidium</i>	2	3			1	6.60		
Mollusca	Bivalvia	Veneroidea	Sphaeriidae	<i>Sphaerium</i>	1	3				7.58		
Mollusca	Bivalvia	Veneroidea	Sphaeriidae	Sphaeriidae	1					6.60		
Mollusca	Gastropoda	Limnophila	Ancylidae	<i>Ferrissia</i>	1					6.55		
Mollusca	Gastropoda	Limnophila	Lymnaeidae	<i>Fossaria</i>				1		6.90		
Mollusca	Gastropoda	Limnophila	Planorbidae	<i>Menetus</i>	1					8.23		
Mollusca	Gastropoda	Mesogastropoda	Hydrobiidae	Hydrobiidae		1				5.78		
Mollusca	Gastropoda	Mesogastropoda	Pleuroceridae	<i>Leptoxis</i>		3				1.79		
Mollusca	Gastropoda	Mesogastropoda	Pleuroceridae	<i>Pleurocera (Elimia)</i>	24	18	1		2	2.46		x
				Total number of individuals	182	224	200	187	192			
				Total number of taxa	38	32	34	13	20			