ABI ENVIRONMENTAL SERVICES

Report of Benthic Macroinvertebrates

LSWC Benthic Program 2024

Taxonomy, Methods, and Quality Control

For
Lesser Slave Watershed Council
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LAND ACKNOWLEDGEMENT

ABI Environmental Services is in the traditional territories of the Niitsitapi (Blackfoot) and the people of the Treaty 7 region in Southern Alberta, which includes the Siksika, the Piikuni, the Kainai, the Tsuut'ina, and the Iyarhe Nakoda. We are situated on land where the Bow River meets the Elbow River, and the traditional Blackfoot name of this place is "Mohkinstsis" which we now call the City of Calgary. The City of Calgary is also home to Metis Nation of Alberta, Region Ill.

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Sample Reception

Samples of benthic macroinvertebrates were collected from two sites using a kicknet (400um) and following the CABIN method for the collection of benthic macroinvertebrates by the staff of the Lesser Slave Watershed Council. The samples were received by ABI Environmental Services on October 9, 2024. All sample containers were found to be free from damage. The benthic invertebrates from each sampling site were collected in 500 ml labelled containers (jars) (Table 1). The preservative in each container was replaced with 70% ethanol on October 10, 2024.

Sample Processing

Some samples contained significant amounts of macrophytes (twigs, leaves), silt/mud and sand. Large plant material like twigs and leaves were rinsed and then removed from the samples and discarded. The amount of smaller macrophytes and the silt/mud, gravel and sand were reduced in the samples by washing and sieving to separate the invertebrates from this debris. Samples were emptied on to stacked sieves, in order from the top: 4000 um, 2360 um and 400 um, and then gently washed with water. A pan or basin was placed under the bottom sieve (400 um). The large vegetation and substrate resting on the 4000 and 2360 um sieves were gently washed with water, inspected for invertebrates, and discarded. Any large invertebrates captured on the coarse sieves were transferred to a temporary holding jar. The contents resting on the sieves were inverted into a basin and gently washed into a shallow white pan. These invertebrates from the coarse and fine fractions were combined. Samples with course and fine (sand) particulates were also inspected for invertebrates and if any found were removed and added to a temporary holding jar. The fraction that passed through the 400-um sieve was inspected for invertebrates and none were found. This procedure was repeated for both samples.

Subsampling and Sample Sorting

The invertebrates in the white pan were inspected and roughly counted to determine if subsampling was necessary. Subsampling was carried out as the invertebrates were found to be abundant in samples from all sites (Table 2). The method of subsampling was accomplished following the CABIN procedure (Martens et al., 2021). Briefly, the samples were mixed with water and transferred to a Marchant box. The Marchant box was sealed, inverted, swirled, and quickly righted. Using a random number generator from Excel, the first five cells of the Marchant Box were selected, and the inverts removed to watch glasses using a transfer pipette. The inverts in each watch glass were counted and rough sorted into major taxa groups. If at least 300 organisms of the taxa of interest (listed in Table 4) were not reached in these five cells, additional cells were randomly chosen until the 300 -organism criterion was reached. If the count was met partway through the cell the entire cell contents was counted as per the CABIN protocol. At the end of sorting these invertebrates were placed in labeled glass vials with 70% ethanol. Excluded taxa were identified and noted (Table 5).

For each of the samples, the sorted fractions (invertebrates removed) were bulked together, labeled as "sorted" and retained for auditing. For each of the samples, the unsorted fractions were labelled as "unsorted" and transferred back to their original containers and retained. These fractions will be retained for 90 days.

Sorting Audit Protocol

Both samples were re-sorted by another team member. Sorting precision was calculated as percent sorting efficiency (% SE) using the CABIN method.

$$\%SE = (1 - \frac{\#\ of\ Organisms\ Missed}{Total\ \#\ of\ Organisms\ Found}) * 100$$

The sorting efficiency is in Table 3, the samples met or exceeded the CABIN protocol of 95% with an average sorting efficiency of 98.3% (Table 3).

Identification and Taxonomy

The rough sorted samples were further examined to identify organisms to the lowest taxonomic level possible. During identification the taxa were entered on paper data sheets and then transferred to an Excel spreadsheet. The CABIN protocols for effort and identification level of respective taxa were followed as closely as possible. In the case of Chironomidae, temporary glycerin slide mounts of dissected specimens were made to confirm identifications to the genus level. Where there were disarticulated specimens only those with heads were counted to avoid double counting specimens. There were also exuviae from larval moults in some samples that were not counted as this may have been double counting specimens present or counting specimens that were not in the sampled portion of the stream bed as exuviae tend to float downstream after a moult. This is especially true for Ephemeroptera. Where possible pupal keys were used to identify fly pupae to family/genus.

Counts per taxa and sample were summarized using a Pivot Table in Excel. Taxa counts for the subsampled (all 2 sites) samples were calculated as Count and Total Count. Total Count was calculated as the number per subsample scaled up to 100% and is the number of inverts present in an entire sample. The Count and Total Count data are presented in Tables 2 and 7, respectively. The number of organisms identified (Count) in the included taxa from the sub-samples were 788 (Table 2). To facilitate comparisons among the samples, the subsampled collections were scaled up to a full sample (Total Count), with the total number of organisms being 9,604 (Tables 2 and 7). These organisms were distributed among 21 families and 37 genera (Table 7).

Auditing Protocol

The auditing protocol was performed on the same samples as the sorting efficiency. We followed the CABIN protocol for determining the Identification Error Rate and tabulated the incorrect

identifications and missed organisms (Table 6A, 6B and 6C). The Identification Error Rate for samples OTR01A and SWR01 were 0% and 0%, respectively. These error rates were within the tolerances for CABIN (%IE ≤ 5%).

$$\%\ Identification\ Error = \left(\frac{\#\ Incorrect\ Identifications}{Total\ \#\ of\ Organisms\ in\ Audit}\right) * 100$$

Taxonomic Keys and References

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Equipment List

- Tyler stainless steel and brass sieves: 13.3 mm, 4000 um, 2000, 1000 um and 400 um
- Catchment pan and basins
- Marchant box for subsampling
- Tools: Transfer pipettes, forceps, acid free paper, squeeze bottles
- Watch glasses – several diameter sizes: 6.5, 8.5, 10.5 cm
- Mason Jars: 125, 250, 500 and 1000 ml
- 70% Ethanol
- Glass vials with screw tops
- Glycerin for temporary slides, slides and cover slips
- Dissecting microscopes: Leica MZ6 and Leica MS5 (4 – 80X)
- Compound microscope Olympus CX41 (40-1000X)
- Light sources: two MI-150 Fiber-lights

Tables

Table 1. Sample reception – field site/sample identification and description, number and size of collection jars and date of collection from the Lesser Slave Watershed Council Benthic Program 2024.

No.	Sample ID	Field Site Description	Jar # & Size	Date Collected	Date Received[A]
1	OTR01A	Otauwau River upstream of Cardinal km 12 bridge.	1 -500 ml	2024/Sep/24	2024/Oct/09
2	SWR01A	Upper Swan River near House Mountain bridge	1 - 500 ml	2024/Oct/03	2024/Oct/09

A. Alcohol changed in on October 10, 2024.

Table 2. Subsampling effort and measured and calculated number of invertebrates per sample, Lesser Slave Watershed Council Benthic Program 2024.

Sample ID	Number of cells sampled from Marchant box[1]	No. of organisms in subsamples[2]	Total number of organisms in sample
OTR01A	5	421	8420
SWR01A	31	367	1184
Total		788	9604

1. A Marchant box has 100 cells, this is the number/percentage sampled to reach at least 300 organisms. 2. Taxonomy performed on these organisms.

Table 3. Sorting efficiency (SE) for three randomly selected samples Lesser Slave Watershed Council Benthic Program 2024.

Sample ID	Original Count	QA Audit Count	Comments	% SE
OTR01A	410	421	*11 Missed: 7 Cricotopus, 4 Polypedilum,*	97.4%
SWR01A	364	367	3 Missed: 1 *Ephemerella, 1 Tanytarsus, 2 Cricotopus,*	99.1
			Average % Sorting Efficiency	**98.3%**

Table 4. Standard taxonomic effort for practical Identification.

Group	Taxa	Attained Level of Identification
Insects	Coleoptera	Family/Genus
	Diptera	Order/ Family/Genus
	Ephemeroptera	Family/Genus
	Plecoptera	Family/Genus
	Trichoptera	Family/Genus
Non-insects	Annelida: Enchytraeidae, Tubificida,	Family/Genus/Species
	Trombidiformes (Mite)	Genus

Table 5. Excluded Taxa -Total Count per site, Lesser Slave Watershed Council Benthic Program 2024.

	Taxa
Aquatic	
Non-aquatic	
	Diptera: Chironomidae – Adult terrestrial drop in

Table 6A. Identification error rate for sample OTR01A, Lesser Slave Watershed Council Benthic Program, 2024.

Order	Family	Genus	Raw Count	Audit Count	Audit Flag	IE Error	Comments
Total							
Total organisms in audit			421	421			
Average % Identification Error Rate						0%	Pass

Table 6B. Identification error rate for sample SWR01A Lesser Slave Watershed Council Benthic Program, 2024.

Order	Family	Genus	Raw Count	Audit Count	Audit Flag	IE Error	Comments
Total							
Total organisms in audit			367	367			
Average % Identification Error Rate						0%	Pass

Table 7. Total count of benthic macroinvertebrates from field sites in Lesser Slave Watershed Council Benthic Program, 2024.

Taxonomic Group	OTR01A	SWR01A	Total
Order: Coleoptera			
Family: Elmidae			
Optioservus	20		20
Order: Diptera			
Family: Ceratopogonidae			
Mallochohelea	20		20
Family: Chironomidae	80	3	83
Ablabesmyia	120		120
Brillia		10	10
Cricotopus	1500	65	1565
Cryptochironomus	300	10	310
Eukiefferiella	140		140
Meropelopia		13	13
Orthocladius		13	13
Pagastia	60	6	66
Parakiefferiella	60		60
Parametriocnemus	80		80
Polypedilum	2960	142	3102
Tanytarsus	2460	274	2734
Family: Empididae			
Neoplasta		10	10
Order: Ephemeroptera			
Family: Ameletidae			
Ameletus	80	116	196
Family: Baetidae			
Baetis		29	29
Family: Ephemerellidae			
Ephemerella	120	155	275
Family: Ephemeridae			
Ephemera	20		20
Family: Heptageniidae		10	10
Epeorus		3	3
Heptagenia		10	10
Maccaffertium		29	29
Rhithrogena		135	135
Family: Leptophlebiidae		10	10
Leptophlebia	20	16	36

Table 7. Total count of benthic macroinvertebrates from field sites in Lesser Slave Watershed Council Benthic Program, 2024.

Taxonomic Group	OTR01A	SWR01A	Total
Order: Plecoptera			
Family: Chloroperlidae			
Plumiperla		19	19
Suwallia		39	39
Family: Nemouridae	20		20
Zapada	20		20
Family: Perlidae			
Hesperoperla		3	3
Family: Perlodidae		6	6
Diura		3	3
Isoperla		13	13
Family: Taeniopterygidae			
Taeniopteryx	20		20
Order: Trichoptera			
Family: Brachycentridae			
Brachycentrus	40	3	43
Family: Hydropsychidae			
Cheumatopsyche	60	3	63
Hydropsyche	40	16	56
Family: Rhyacophilidae			
Rhyacophila		3	3
Order: Trombidiformes			
Family: Sperchontidae			
Sperchon		16	16
Order: Tubificida			
Family: Naididae	20		20
Nais	120		120
Family: Enchytraeidae	20		20
Total	8400	1184	9584