Arcellaceans (Testate Lobose Amoeba) as Proxies for Arsenic and Heavy Metal Contamination in the Baker Creek Watershed Region, Northwest Territories, Canada.

by

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Abstract

Arcellaceans (testate lobose amoebae) were examined for 61 sediment surface samples from lakes in the vicinity of the Giant Mine near Yellowknife, Northwest Territories to; (1) quantify the impact of the mine on the Baker Creek Watershed region, (2) determine the utility of arcellaceans as indicators of arsenic and heavy metal contamination and gauge the success of remediation efforts. Several statistical methods, including cluster analysis, Deterended Correspondence Analysis (DCA), and Redundancy Analysis (RDA), were used to quantify the impact of mining activity on the arcellacean assemblages. Cluster analysis revealed five arcellacean assemblages associated with a range of environmental conditions (e.g. polluted, transitional and remediated). Partial RDA results confirm that arsenic has the greatest influence on the arcellacean distribution, explaining 10.7% of the total variance. Stress-indicating species (e.g. Centropyxids) correlate with high arsenic concentrations, while species characteristic of more healthy lake conditions (e.g. Difflugids) dominate sites with significantly lower arsenic concentrations.

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1. Introduction

1.1 Giant Mine

In 1933, the discovery of significant gold mineralization in the Yellowknife Supergroup of the Slave Geological province prompted the establishment of three major gold mines in the vicinity of the City of Yellowknife, Northwest Territories, Canada; Discovery Mine, Con Mine, and Giant Mine. Of the three mines, Giant Mine went on to become one of the most productive and most continuous gold mining operations in Canadian mining history. The mine produced gold from 1948 until 1999, after which the ownership of the mine transferred to the Government of Canada. While all ore processing activities shifted to the neighboring Con Mine, mining activities at the Giant Mine continued until mid-2004 before officially halting in July 2004 (Golder, 2013). Between 1948 and 2004, the mine's production of 7.6 million ounces of gold resulted in a post-World War II economic boom for the territory and mining industry. Unfortunately, the Giant Mine operation was also responsible, particularly in the early days, for the daily release of significant quantities of arsenic trioxide (As_2O_3) particles into the atmosphere and surrounding environment. Arsenic is a metalloid that is known to be toxic to both plants and animals, due to its affinity for protein, lipids and other cellular components (Ferguson and Gavis, 1972; Rosemond et al., 2008). Moreover, chronic exposure to arsenic can lead to severe health effects in humans, such as skin lesions, anemia, liver damage and cancer (Caussy and Priest, 2008).

At the Giant Mine, gold was recovered from sulfides, predominantly arsenopyrite and pyrite. In order to extract gold, the high quantities of arsenic and sulfur contained in these sulfides had to be removed. The extraction of gold from the host ore was normally achieved by using a high-temperature roasting method. A byproduct of this method was the release of arsenic trioxide (As_2O_3) particles into the atmosphere. In the early 1950s, inefficient extraction practices and lax emission control policies in place at the mine resulted in 2.6 million kg/year of air fall derived deposition of As_2O_3 to the surrounding environment and the release of arsenic contaminated effluent (25,000 kg/year) to the adjacent Baker Creek (MacDonald, 1997; SRK Consulting, 2002). Aerial emissions and release of contaminated effluent to Baker Creek decreased substantially in mid-1980s (5,700 kg/year and 500 kg/year, respectively), due to much more environmentally sustainable mining practices, which resulted in the storage of approximately 237,176 tons of As_2O_3 in the depths of the Giant Mine (MacDonald, 1997; SRK Consulting, 2002). In total, it was estimated that the Giant Mine alone was responsible for releasing approximately 19 million kg of As_2O_3 as aerial emissions since 1949 (Galloway et al., 2012).

Despite the closure of the mine, arsenic contamination remains a concern to the residents of Yellowknife as a legacy of the enormous quantities of As_2O_3 that were deposited across the landscape through the years (MacDonald, 1997; SRK Consulting, 2002). In response to this major environmental concern, the federal and territorial governments cooperated to produce the Giant Mine remediation project, which was finalized by the government of the Northwest Territories in 2007 (Golder, 2013)

1.2 Baker Creek

Baker Creek Watershed (BCW) is a small creek that originates at Duckfish Lake, approximately 25 km northeast of Yellowknife, and flows south and southeast until it discharges into Yellowknife Bay (Golder, 2013). The drainage area of the BCW is ~121 km^2. Peak daily discharge volumes, reported between 1983 and 2010, ranged from 0.27 to 8.35 m^3/s, with peak discharge in May during spring freshet (Environment Canada, 2011). Water depths within the creek are quite shallow, varying from few centimeters to ~2.3 meters deep. The substrate in Yellowknife Bay, where the creek discharges, is dominated by silt and sand deposits, as expected in a depositional area.

The sediment and water of the BCW are known to have been contaminated by arsenic since the beginning of the Giant Mine operations in the early 1940s. This is not a surprising observation since the creek passes through the mine site. Numerous studies have reported elevated arsenic concentration in the creek's water and sediment. For example, Moore et al. (1978) reported very high arsenic concentrations, in the creek's surface water, ranging from 1500 to 20,400 µg/L. High arsenic contaminations, ranging from 1764 -3821 mg/kg, have also been reported from the sediments near the mouth of the BCW (e.g. Jackson et al. 1996; Mace, 1998). However, most of these studies were dedicated for measuring arsenic concentrations in the contaminated BCW without considering the environmental impact of Giant Mine on the ecology of the BCW and surrounding region. Several studies have focused on investigating arsenic contamination patterns and impact in sites beyond the lease area of Giant Mine though (Galloway et al., 2012; Golder., 2013).

1.3 Arcellaceans

Arcellaceans (informally known as thecamoebians or testate lobose amoebae) are an artificial group of unicellular protozoans that occur abundantly in Quaternary lacustrine sediments (Loeblich and Tappan, 1964; Medioli and Scott, 1983; Scott and Medioli, 1983). They are found throughout the world, from tropical to polar regions, in a wide variety of freshwater habitats, such as lakes, streams, rivers, ponds, mosses, soil and in tree barks (Medioli and Scott, 1983; Ogden and Hedley, 1980; Patterson et al., 1985; 1996; Medioli et al., 1990ab) with a few species able to tolerate marginally brackish habitats (Charman et al., 2000; Patterson and Kumar, 2002). The soft amoeboid cell is protected by a beret-, or sac-like test (shell) that range in size from 5 to 300 µm. The test is either autogenous, made of secreted siliceous, proteinaceous or calcareous materials, or xenogenous, made from agglutinated foreign materials, such as sand grains and diatoms frustules (Patterson and Kumar, 2000)

Over the past three decades, research on arcellaceans shed light on their value as proxies for variable environmental and climatic parameters, including paleoclimatic reconstruction (McCarthy et al., 1995), water table fluctuations, (Charman et al., 1998), lake acidity (Kumar and Patterson, 2000), land-use change (Patterson et al., 2002), ecosystem health and seasonal environmental changes (Neville et al., 2011), pH variability (Patterson et al., 2013), nutrient loading (Patterson et al., 2012) and water quality (Roe et al., 2010). The value of arcellaceans in paleontological and paleolimnological studies is attributed to: (1) their abundance in organic-rich surface sediments (between 500-3,000 specimens per ml), (2) the resistance of their tests to dissolution, and (3) their sensitivity to a wide variety of environmental variables such as temperature, pH, Total Organic Carbon, Dissolved Oxygen and heavy metal contamination (Medioli et al. 1990a; Warner, 1991; Warner and Charman, 1994; Warner and Bunting, 1996; Patterson et al., 1996; Charman et al., 2000; Patterson and Kumar, 2002; Patterson et al., 2002).

A number of recent studies in Canada and Europe have demonstrated that distinct arcellacean assemblages, as well as individual species and strains may be significantly impacted by industrial pollutants (Asioli et al., 1996; Patterson et al., 1996; Reinhardt et al., 1998; Patterson and Kumar, 2000; Neville et al., 2011; Kihlman and Kauppila, 2009;2012). In addition, some of these studies have identified a positive correlation between arcellacean infrasubspecific strains and heavy metal contamination (Asioli et al., 1996; Patterson et al., 1996; Reinhardt et al., 1998). Arcellaceans are characterized by a rapid reproduction rate of days to weeks, which makes them particularly useful for monitoring the ecosystem health of contaminated lakes and for assessing the progress of remediation efforts (Patterson et al., 1996; Reinhardt et al., 1998; Neville et al., 2011; Patterson et al., 2012; 2013).

1.4 Study objectives

This study aims to: (1) quantify the impact of Giant Mine on the Yellowknife area, including the BCW; and (2) elucidate the potential of arcellaceans as an efficient and inexpensive tool to identify and characterize arsenic and other heavy metal contamination, as well as to monitor the progress of remediation efforts in the study area.

2. Regional setting

Lakes investigated in this study are located in the central NT near the city of Yellowknife (Figure 2.1 and Table 2.1). The bedrock underlying the Yellowknife area is part of the southern Slave structural province of the Canadian Shield. The bedrock is generally comprised of Archean meta-volcanic and meta-sedimentary rocks of the Yellowknife Supergroup including mafic to felsic volcanic rocks (e.g. basalt, andesite and pillowed flows). These deposits trend north-south of Yellowknife greenstone belt found in the central part of the region (Jolliffe, 1942; Henderson, 1985). These deposits have in turn been intruded by widespread granitoid rocks consisting of granite, granodiorite, and tonalite to the west and southeast, as well as isolated intrusive bodies of metasedimentary rocks to the north and east (Stubley, 1997). The region is also crosscut by a variety of Proterozoic diabase and gabbro dykes, which trend in a northeasterly direction. Several major fault lines divide the volcanic rocks from the younger granitoid rocks, including the Kam Lake Fault and the West Bay Fault, which run through Yellowknife (Kerr and Wislon 2000).

Topographic elevations in the area range from 157 m above sea level at Great Slave Lake, rising gradually to 350-400 m north of Thisletwaite Lake. The Yellowknife area is dominated by a low-relief terrain, mainly consisting of rocky outcrops associated with glacial and glaciolacustrine sediments in topographic lows. The Yellowknife River is the principle component of the drainage system in the area, with the outlet flowing south into Yellowknife Bay, Great Slave Lake. Most streams and rivers are shallow, and few have cut into the underlying bedrock or surficial sediments. The drainage system is influenced by bedrock structure. As a result, numerous small elongated lakes have formed along fault lines and joints in the bedrock.

The climate of the region is continental, characterized by short dry cool summers. The mean annual temperature is -5.2º C, with the hottest month being July (average temperature = 16.5º C) and the coolest being January (average temperature = -27.9º C). Mean annual precipitation in the Yellowknife area is low (267.4 mm), with an average monthly precipitation of 22.3 mm. Probability of precipitation in the Yellowknife region varies throughout the year, with it being most likely in November (occurring on 84% of the days) and least likely in May (occurring on 35% of the days). The driest weather is in April (average rainfall of 10.3 mm), while the wettest month is August (average precipitation of 41.7 mm). Prevailing wind direction varies around the year, with the dominant wind direction is often out of east.

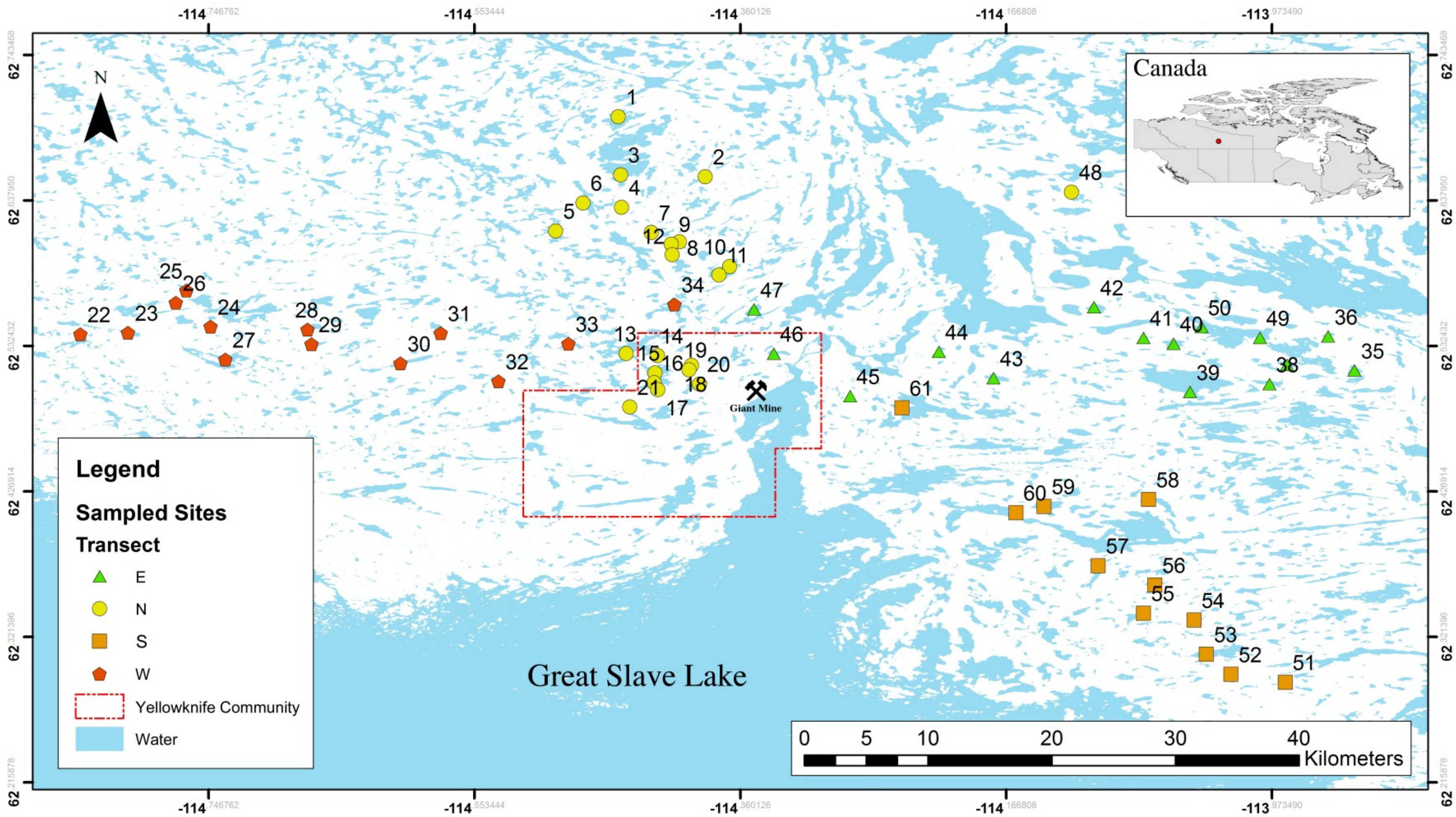

Figure 2.1 Map of the Northwest Territories showing the locations of the sixty-one sediment-water-interface samples collected from 59 lakes in the region surrounding the Giant Mine, and the inset shows the location of the study area within Canada as a red dot.

Table 2.1 Coordinates of the fifty-nine sampled lakes

No.	Sample code	Site	Transect	Co-ordinates Lat – Long
1	BC-1	N Duckfish	North	62.6987, -114.4489
2	BC-2	Icing Lake	North	62.6552, -114.3856
3	BC-3	Duckfish Lake	North	62.6566, -114.4471
4	BC-4	Rock Lake	North	62.6330, -114.4465
5	BC-5	N690	North	62.6158, -114.4944
6	BC-6	North Lake	North	62.6362, -114.4744
7	BC-7	Trail Lake	North	62.6147, -114.4248
8	BC-8	East Vital (TP2)	North	62.6080, -114.4045
9	BC-9	East Vital (TP1)	North	62.6063, -114.4103
10	BC-10	Ryan Lake North	North	62.5900, -114.3678
11	BC-11	Ryan Lake South	North	62.5840, -114.3756
12	BC-12	North of TP3	North	62.5987, -114.4096
13	BC-13	Martin	North	62.5269, -114.443
14	BC-14	KL1	North	62.5254, -114.4203
15	BC-15	Lower Martin	North	62.5129, -114.4221
16	BC-16	L. Martin SW	North	62.5058, -114.4227
17	BC-17	L3	North	62.5007, -114.4201
18	BC-18	L1	North	62.5184, -114.3956
19	BC-19	KL9	North	62.5152, -114.3974
20	BC-20	L2	North	62.5048, -114.3897
21	BC-21	KL8	North	62.4882, -114.4405
22	BC-22	*West 1*	West	62.5411, -114.8400
23	BC-23	*West 3*	West	62.5422, -114.8054
24	BC-24	NA	West	62.5466, -114.7452
25	BC-25	NA	West	62.5727, -114.7628
26	BC-26	*West 2*	West	62.5638, -114.7707
27	BC-27	*West 6*	West	62.5226, -114.7345
28	BC-28	*West 7*	West	62.5444, -114.6748
29	BC-29	*West 8*	West	62.5338, -114.6719
30	BC-30	*West 9*	West	62.5199, -114.6072
31	BC-31	*West 10*	West	62.5418, -114.5780
32	BC-32	*West 11*	West	62.5069, -114.5361
33	BC-33	*West 12*	West	62.5342, -114.4850
34	BC-34	*West 13* - Landing Lake	West	62.5626, -114.4080
35	BC-35	*East 1*	East	62.5148, -113.9134
36	BC-36	*East 2*	East	62.5395, -113.9326
37	BC-37	*East 3*	East	62.5189, -113.9626

Table 2.1 Continued

No.	Sample code	Site	Transect	Co-ordinates Lat – Long
38	BC-38	*East 4*	East	62.5049, -113.9754
39	BC-39	*East 5* - Bighill Lake	East	62.4992, -114.0331
40	BC-40	*East 6*	East	62.5342, -114.0450
41	BC-41	*East 7*	East	62.5385, -114.0669
42	BC-42	NA	East	62.5606, -114.1029
43	BC-43	*East 9*	East	62.5092, -114.1759
44	BC-44	*East 11*	East	62.5284, -114.2156
45	BC-45	NA	East	62.4962, -114.2803
46	BC-46	*East 13* - Shot Lake	East	62.5270, -114.3357
47	BC-47	*East 14* - Vee Lake	East	62.5591, -114.3501
48	BC-48	Pluton Lake	North	62.6440, -114.1193
49	BC-49	Pontoon Lake SE	East	62.5385, -113.9822
50	BC-50	Pontoon Lake NW	East	62.5462, -114.0246
51	BC-51	*South 1*	South	62.2884, -113.9639
52	BC-52	*South 2*	South	62.2941, -114.0033
53	BC-53	*South 3*	South	62.3087, -114.0213
54	BC-54	*South 4*	South	62.3335, -114.0302
55	BC-55	*South 5*	South	62.3385, -114.0669
56	BC-56	*South 6*	South	62.3589, -114.0587
57	BC-57	*South 7*	South	62.3729, -114.0998
58	BC-58	*South 12*	South	62.4211, -114.0632
59	BC-59	*South 9*	South	62.4161, -114.1392
60	BC-60	Mason Lake (sheltered bay)	South	62.4115, -114.1596
61	BC-61	*South 11* - Hay Lake	South	62.4876, -114.2426

3. Methods

3.1 Field work

Sixty-one sediment-water interface samples were collected from fifty-nine lakes across the region of the Giant Mine in August of 2012. The samples were collected using an Eckman grab sampler that was suspended from a helicopter. Sampling sites were broadly distributed to cover the maximum area surrounding the Giant Mine. The samples were carefully collected along four transects (north, south, east and west) to ensure coverage of the region surrounding the lease area of the mine (Figure 2.1). Water samples were also collected for the purpose of determining the concentration of nutrients present in each lake.

A Trimble Scout Global Positioning System (GPS) was used to determine the geographical position at each station (Table 2.1). Sample site selection was determined using a commercial "fish finder" sonar equipped with a bottom hardness indicator. The bottom hardness indicator facilitated faster sampling time as it readily distinguished rocky, sandy and muddy substrate. Samples were collected from muddy substrates as winnowed sandy substrates usually yield small allochthonous arcellacean populations whereas rocky substrates are usually barren.

A YSI Professional Plus handheld multi-paprameter instrument equipped with quarto cables was used to record pH, temperature (in °C), conductivity (in microsiemens), and dissolved oxygen (in mg/l) at 0.1 m depth intervals through the water column and at the sediment-water interface (Table 3.1).

Table 3.1 Environmental parameters measured for each of the sixty-one sediment-water-interface samples.

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Variable Sample	Water Depth	DO (T)	DO (B)	Cond. (T)	Cond. (B)	pH (T)	pH (B)	Temp. (T)	Temp. (B)
BC-1	4.6	11.66	9.65	53.1	58.0	6.87	7.29	9.5	9.6
BC-2	7.0	9.55	7.95	40.2	42.2	7.32	7.07	11.8	11.6
BC-3	2.4	11.71	10.35	93.2	93.2	8.12	8.15	9.5	9.4
BC-4	2.9	10.77	8.72	33.8	40.5	7.98	7.41	9.9	9.8
BC-5	3.0	11.71	10.25	55.8	55.6	7.86	7.75	8.8	8.7
BC-6	11.5	10.09	0.05	95.4	115.6	7.94	6.88	11.6	4.6
BC-7	1.3	12.35	12.12	31.3	31.3	7.98	7.75	8.1	8.1
BC-8	1.1	10.98	10.53	44.3	44.2	7.65	7.46	8.4	8.3
BC-9	1.0	11.88	11.17	45.1	45.1	7.63	7.48	8	8.1
BC-10	13.3	11.33	1.81	89.9	104.0	8.18	6.97	12.3	10.3
BC-11	5.2	10.37	3.44	90.2	102.1	8.20	8.65	12.5	12.5
BC-12	1.0	11.27	10.66	43.1	43.0	7.31	7.22	8.4	8.3
BC-13	4.5	11.08	10.31	65.4	65.3	7.59	7.73	11	10.8
BC-14	1.0	10.65	9.87	110.3	112.6	7.57	7.46	8.5	8.5
BC-15	2.0	11.59	10.58	65.3	65.3	7.88	7.71	9.1	9.2
BC-16	1.3	12.16	11.74	65.2	65.1	7.94	7.88	9.2	9.2
BC-17	0.8	11.53	11.6	93.2	93.4	7.78	7.79	9.4	9.5
BC-18	1.0	12.40	2.18	182.5	182.2	8.45	8.42	9.2	9.2
BC-19	0.7	11.49	11.47	162.6	162.6	8.05	8.05	10	10
BC-20	1.4	11.58	10.54	326.6	326.4	8.48	8.32	9	9
BC-21	0.6	11.34	11.34	118.7	118.7	7.91	7.65	9.8	9.8
BC-22	2.5	7.23	4.31	88.6	96.1	7.56	7.02	9.1	9
BC-23	5.1	9.48	9.01	70.8	70.8	7.42	7.21	11.1	11.1
BC-24	3.1	10.66	10.17	34.4	34.6	7.26	7.05	9.6	9.6
BC-25	1.0	12.50	12.51	51.8	51.8	7.26	7.38	8.7	8.7
BC-26	1.0	12.02	11.89	175	175.1	7.69	7.75	8.9	8.8
BC-27	3.6	11.81	11.37	99.4	99.1	7.81	7.65	10.4	10.4
BC-28	1.3	13.75	13.79	84.3	84.3	7.99	7.97	8.5	8.4
BC-29	1.5	14.12	13.87	97.1	97.0	8.29	8.27	8.8	8.8
BC-30	3.5	11.12	10.23	63.5	64.8	7.60	7.44	8.7	8.7
BC-31	0.8	13.68	13.51	36.6	36.6	7.97	7.77	11.5	11.5
BC-32	1.2	13.61	11.82	55.9	56.7	7.65	7.62	8.8	8.8
BC-33	3.3	11.44	8.73	66.6	67.7	7.52	7.39	10.5	10.5

Table 3.1 Continued

Variable Samples	Water Depth	DO (T)	DO (B)	Cond. (T)	Cond. (B)	pH (T)	pH (B)	Temp. (T)	Temp. (B)
BC-34	2.8	12.80	12.24	60.4	60.3	7.60	7.68	9.7	9.7
BC-35	5.9	10.93	10.21	72	71.9	7.79	7.48	12.1	12
BC-36	1.5	14.70	13.55	55.2	55.4	8.11	7.99	9.4	9.4
BC-37	7.3	9.83	4.29	202.1	208.5	7.91	7.49	11.7	11.1
BC-38	12.0	11.18	0.07	99.1	100.5	8.13	6.78	11.7	5.1
BC-39	8.7	11.62	11.64	133.9	133.0	8.27	8.3	10.3	10.2
BC-40	4.4	11.71	10.16	112.9	119.2	8.09	7.66	9.9	9.7
BC-41	1.4	12.12	11.8	89.2	88.6	7.59	7.47	9.7	9.8
BC-42	2.9	11.97	11.76	63.5	63.6	7.59	7.47	11	10.9
BC-43	5.3	12.04	9.88	264.3	276.4	8.44	8.31	10.2	10.1
BC-44	2.0	12.64	12.35	278.7	278.4	8.25	8.17	10.6	10.2
BC-45	2.9	11.83	11.11	185.8	183.2	8.14	8.01	10.8	10.7
BC-46	4.2	10.16	8.68	125.2	126.1	7.80	7.49	10.1	10
BC-47	3.4	12.53	12.49	182.4	182.0	7.91	8.15	10.1	10
BC-48	3.3	11.83	11.25	626	626.0	9.01	9.08	11.6	11.6
BC-49	5.7	10.69	10.35	284.5	284.8	8.45	8.39	11.6	11.7
BC-50	4.9	10.68	10.4	284.9	284.8	8.45	8.43	10.1	10.1
BC-51	1.0	11.53	11.23	94.3	94.0	8.20	7.87	10.3	10.1
BC-52	1.6	11.07	10.68	112.8	112.2	7.91	7.64	9.8	9.8
BC-53	1.5	11.08	10.4	72.4	73.6	7.69	7.52	10	10
BC-54	1.5	10.65	10.4	162	163.4	7.71	7.7	10.3	10.4
BC-55	3.8	11.09	8.98	83	84.6	7.77	7.49	10	10
BC-56	2.0	11.75	10.32	92.2	93.2	8.00	7.68	12.1	11.4
BC-57	9.0	10.07	1.38	184.6	193.9	8.18	7.66	11.3	11.3
BC-58	2.0	10.82	10.64	93.5	93.4	8.01	7.89	12.5	12.2
BC-59	6.7	9.00	NA	755.4	NA	8.40	NA	11.1	11
BC-60	4.5	11.23	10.84	166.2	165.3	8.53	8.39	NA	NA
BC-61	6.5	10.46	9.23	219.7	219.5	8.16	8.17	NA	NA

3.2 Laboratory work

The upper 5 mm of sediment from each Eckman grab was removed for arcellacean, geochemical and sedimentological analyses. Sediment samples for particle size analysis (% clay, slit and sand) were digested in a heated bath (50°C) with 10 % H_2O_2 to remove organics (Murray 2002; van Hengstum et al. 2007; Donato 2009). Digested samples were then analyzed using a Beckman Coulter LS 13 320 laser diffraction analyzer fitted with a universal liquid medium (ULM) sample chamber over a measurement range of between 0.4 and 2,000 lm. The samples were loaded into the instrument until an obscuration level of 10 ± 3 % was attained. GRADISTAT (Version 8; Blott and Pye, 2001) was used to compile the results (Table 3.2).

Samples were analyzed for metals of interest in environmental research at ACME Analytical Laboratories (Vancouver) Ltd. using the geochemical Ultratrace Inductively Coupled Plasma Mass Spectrometry (ICP-MS) after Aqua Regia digestion for low and ultra-low determinations in the sediment samples (Table 3.3). Total organic carbon (TOC) was determined using Rock-Eval® Analysis 6 Analysis (S1, S2, and S3 sediment fractions; Vinci Technologies, Rueil- Malmaison, France) at the Geological Survey of Canada, Calgary (Table 3.4). The standard reference materials used for this method included IFP 1600000, Institute Francais du Petrole and an internal 9107 Shale standard. The analytical reproductiviy based on measurements of series duplicates was generally better than 5%.

Water samples were transported to Caduceon Environmental Laboratories (Ottawa) to analyze the following parameters: nitrate (in mg/l), nitrate (in mg/l), ammonia (mg/l), total Kjeldahl nitrogen (in mg/l) and total phosphorous (in mg/l) (Table 3.5).

Table 3.2 Particle size analysis (% clay, silt and sand) results

Sample	% SAND	% SILT	% CLAY
BC1	0.09	0.74	0.17
BC2	0.04	0.79	0.17
BC3	0.04	0.84	0.12
BC4	0.04	0.75	0.20
BC5	0.20	0.67	0.13
BC6	0.06	0.83	0.11
BC7	0.07	0.78	0.15
BC8	0.25	0.62	0.13
BC9	0.13	0.68	0.19
BC10	0.30	0.63	0.07
BC11	0.79	0.17	0.03
BC12	0.13	0.72	0.14
BC13	0.18	0.66	0.16
BC14	0.10	0.76	0.15
BC15	0.07	0.75	0.18
BC16	0.12	0.74	0.14
BC17	0.08	0.81	0.10
BC18	0.10	0.77	0.13
BC19	0.13	0.81	0.06
BC20	0.06	0.80	0.15
BC21	0.07	0.84	0.09
BC22	0.05	0.68	0.27
BC23	0.02	0.89	0.09
BC24	0.25	0.66	0.09
BC25	0.08	0.73	0.19

Sample	% SAND	% SILT	% CLAY
BC26	0.04	0.84	0.12
BC27	0.16	0.61	0.23
BC28	0.07	0.75	0.18
BC29	0.07	0.75	0.18
BC30	0.21	0.67	0.12
BC31	0.06	0.73	0.21
BC32	0.05	0.73	0.22
BC33	0.07	0.77	0.15
BC34	0.05	0.81	0.14
BC35	0.04	0.90	0.06
BC36	0.13	0.74	0.12
BC37	0.28	0.52	0.20
BC38	0.03	0.86	0.10
BC39	0.09	0.80	0.11
BC40	0.06	0.75	0.19
BC41	0.08	0.71	0.20
BC42	0.15	0.70	0.15
BC43	0.04	0.83	0.13
BC44	0.31	0.64	0.05
BC45	0.09	0.80	0.11
BC46	0.32	0.57	0.11
BC47	0.02	0.81	0.17
BC48	0.12	0.73	0.16
BC49	0.13	0.71	0.16
BC50	0.08	0.69	0.23

Sample	% SAND	% SILT	% CLAY
BC51	0.11	0.68	0.21
BC52	0.18	0.61	0.21
BC53	0.11	0.61	0.27
BC54	0.24	0.60	0.17
BC55	0.10	0.78	0.12
BC56	0.12	0.71	0.17
BC57	0.05	0.87	0.07
BC58	0.01	0.63	0.36
BC59	0.10	0.69	0.21
BC60	0.53	0.44	0.04
BC61	0.00	0.69	0.31

Table 3.3 ICP-MS results (PPM)

Sample	Mo	Cu	Pb	Zn	Ag	Ni	Co	Mn	Fe	As
BC 1	2.58	31.32	10.13	91.2	0.12	21.3	8.9	226	16500	107.9
BC 2	13.37	24.77	9.16	240.2	0.119	28.6	39.5	10000	87300	905.2
BC 3	1.99	28.06	9.72	108.7	0.12	20.8	9.1	604	19600	126.3
BC 4	2.47	34.65	17.39	128.8	0.15	24.8	9.7	226	3900	555.4
BC 5	2.72	32.6	4.83	139.7	0.127	20.2	9	179	12200	90.9
BC 6	3.71	28.98	10.92	93.7	0.098	18.6	10.6	820	18700	236.1
BC 7	2.05	17.87	6.47	83.8	0.069	18.3	4	73	4400	19.5
BC 8	3.2	28.58	5.76	91.4	0.073	28.7	8.6	263	10900	41.9
BC 9	2.56	38.42	8.25	110.6	0.079	28.3	11.1	694	14700	35.5
BC 10	15.13	60.71	13.76	91.2	0.165	28.4	10.1	1210	14800	192.8
BC 11	7.07	16.3	7.83	41.8	0.032	14.6	4.5	100	10300	16.1
BC 12	1.55	20.35	7.79	103.3	0.083	24.8	11.8	440	8600	372.2
BC 13	6.16	38.48	10.6	130.1	0.152	26.4	9.7	401	17200	740.7
BC 14	1.77	29.88	9.24	109	0.148	18.5	5.8	193	8300	1063.8
BC 15	5.35	34.72	9.92	85.8	0.115	20.2	4.6	206	7000	290.4
BC 16	7.12	37.03	5.63	86.3	0.072	22.2	4.7	156	9300	92.3
BC 17	3.2	26.75	32.9	89.1	0.409	16.7	5.5	223	8300	4778.2
BC 18	8.35	37.27	9	105.3	0.137	17.4	6.9	503	8800	906.9
BC 19	9.06	44.1	41.99	120.3	0.449	16.3	6.3	116	5000	10000
BC 20	13.59	69.32	13.53	51	0.138	36	9.8	292	11300	160.4
BC 21	2.38	26.1	5.25	71.5	0.107	22.4	5.8	254	9100	368.4
BC 22	1.64	33.35	12.63	119.3	0.126	47.8	24.9	730	29900	43.7
BC 23	0.79	24.81	12.38	98.2	0.116	31.7	12	406	29100	30.2
BC 24	2.52	28.73	3.31	221.7	0.109	24.6	13.8	296	6300	71.9
BC 25	1.47	18.67	6.02	49.9	0.069	18.9	5.8	253	8400	39.6
BC 26	1.08	15.51	5.79	75	0.082	23.9	8.5	510	16500	30.3
BC 27	1.59	25.11	10.23	89	0.103	32.1	11.2	707	27100	112.6
BC 28	1.68	18.02	5.86	62	0.062	17.9	5.8	262	10600	93.9
BC 29	3.5	23.73	8.2	86.7	0.105	22.2	6.9	243	15100	397.5
BC 30	0.69	16.44	6.67	67.5	0.061	19.7	7.1	1342	18600	117.1
BC 31	1.44	22	6.26	116.4	0.096	24.8	10.8	391	10400	299.8
BC 32	2.18	21.24	10.35	77	0.143	23	7.4	312	9700	955.1
BC 33	3.7	34.93	3.8	89.9	0.083	25.4	7.5	344	8500	260
BC 34	6.09	41.22	12.44	152.4	0.141	35.1	14.1	292	19200	289.4
BC 35	0.64	45.09	11.08	99	0.113	52.5	11.9	718	23100	9.7
BC 36	1.14	37.33	9.14	103.4	0.146	42.9	5.9	272	4600	38.1
BC 37	0.77	27.24	8.55	80.3	0.092	38	11.9	896	16800	19
BC 38	1.43	46.4	13.76	95	0.115	51.3	13.2	847	25100	81.1
BC 39	0.47	25.47	11.37	60.2	0.081	27.2	10.7	1054	23200	26.2
BC 40	0.84	22.11	6.74	80.2	0.073	25.4	6.8	365	13400	37

Table 3.3 Continued

Sample	Mo	Cu	Pb	Zn	Ag	Ni	Co	Mn	Fe	As
BC 40	0.84	22.11	6.74	80.2	0.073	25.4	6.8	365	13400	37
BC 41	0.68	21.41	5.77	59.9	0.064	36	8.2	267	11400	22.3
BC 42	1.5	27.92	8.11	84.5	0.08	25.3	7.5	265	13600	35.6
BC 43	1.01	29.09	5.97	106.1	0.082	29.9	10.2	901	18700	115.7
BC 44	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
BC 45	1.29	35.2	14.1	87.1	0.119	40.7	14.6	431	33900	11.9
BC 46	1.89	31.7	14.93	78.5	0.129	33.1	12.2	304	22400	180.2
BC 47	2.4	37.93	14.9	76.7	0.111	27	10	244	21300	339.8
BC 48	0.92	23.67	5.09	81.9	0.091	16.1	6.2	760	10200	46.9
BC 49	0.47	12.04	5.05	31.6	0.036	12.6	4.1	2045	6400	27.4
BC 50	0.6	13.26	4.49	35.8	0.035	15.1	4.2	454	6900	21.1
BC 51	1.56	14.76	3.57	54.4	0.052	16.9	4.9	576	10300	27.5
BC 52	0.82	23.81	7.85	59	0.067	25.8	7.9	280	15900	6.3
BC 53	1.47	16.27	5.88	50.4	0.065	21	5.8	309	12900	27.9
BC 54	1.37	14.96	4.76	49.2	0.055	14.5	3.6	161	8200	31.3
BC 55	1.26	20.24	7.3	115.4	0.073	23.4	7	334	19100	44.1
BC 56	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
BC 57	0.36	22.35	10.65	86.7	0.072	29.7	11.8	672	30400	71.5
BC 58	1.19	32.68	8.32	69.1	0.075	34.6	10	386	17700	24.6
BC 59	1.46	21.22	5.79	43.8	0.058	20.4	6.1	967	13400	32.4
BC 60	0.26	13.1	4.83	26.9	0.025	14.4	4.7	142	9900	7
BC 61	0.52	25.45	11.2	73.2	0.086	36.1	12.5	1182	29200	38.4

Table 3.3 Continued

Sample	U	Th	Sr	Cd	Sb	V	Ca	P	La	Cr
BC 1	53.4	1.8	56.2	0.5	3.49	20	8300	980	33.1	22.2
BC 2	172.5	7.5	97.7	0.78	9.52	38	10500	2430	59.1	24.3
BC 3	33.4	2.6	52.6	0.65	4.37	20	9200	1040	30.7	17.9
BC 4	31.7	1	46.4	0.83	14.84	18	11300	1220	13.9	19.6
BC 5	10.9	1.1	39.6	0.65	2.67	25	6400	740	16.1	18.4
BC 6	36.2	1.8	49.1	0.48	14.82	15	9400	1640	13.5	18.9
BC 7	19.2	1.7	49.9	0.26	0.96	7	10500	860	10.8	11.5
BC 8	19.8	1.9	53.4	0.22	1.09	19	10900	930	15.1	21.2
BC 9	37.5	1.5	41.8	0.34	1.37	23	9800	780	20	30.3
BC 10	9.3	1.1	39.5	0.38	6.59	23	10200	1750	19.8	35.7
BC 11	1.3	3.6	14	0.07	3.28	16	4100	440	11.7	19.5
BC 12	3.5	0.4	62	0.38	5.78	17	14500	1160	6.1	15.7
BC 13	15	0.7	33.2	0.48	16.21	25	7300	1100	15.5	26.4
BC 14	8.2	0.9	41.6	0.42	20.4	12	11800	880	7.1	15.8
BC 15	13.2	0.8	37.9	0.48	10.96	16	8200	660	10.4	16
BC 16	13.4	1	40.7	0.39	2.3	15	8500	630	11.5	17.7
BC 17	11.6	0.7	48.1	0.47	44.4	10	10900	1030	6.6	12.7
BC 18	11.2	0.5	46.3	0.46	18.33	10	13700	590	5.9	11.7
BC 19	5.3	0.5	36.2	0.6	187.37	11	11100	770	4.5	9.7
BC 20	7.2	1.1	123.8	0.2	8.16	23	62300	1010	6.4	33.4
BC 21	6.3	1.2	71.2	0.4	7.82	14	18000	860	9.7	16.4
BC 22	3.9	8.8	51.2	0.33	2.1	50	6300	1060	32.2	49.9
BC 23	7.2	9.6	42.8	0.2	2.78	49	5000	910	33.3	48.5
BC 24	9	0.4	56.7	0.59	1.98	15	10100	1370	9.3	16.8
BC 25	6.7	1.3	56.6	0.14	3.38	19	9800	1220	12.7	18.5
BC 26	2	4.2	86.2	0.23	1.04	24	15400	830	14.3	23.5
BC 27	6.9	5.2	58.3	0.35	6.91	36	9200	1230	24	35.6
BC 28	4	2.3	45.7	0.2	2.74	21	8200	1250	11.4	18.3
BC 29	5.7	1.9	50.2	0.34	9.54	21	9100	720	11	17.4
BC 30	1.9	2.3	31.7	0.2	10.51	23	6700	1650	15.1	23.2
BC 31	4	0.8	49.9	0.47	8.63	18	10400	850	12.8	19.9
BC 32	2.9	1.2	55.5	0.45	18.21	15	14300	940	7.8	16
BC 33	5.6	1	40.4	0.53	12.01	14	9300	680	15.3	18.1
BC 34	16.1	2	35.1	0.44	13.05	29	7400	820	21.4	36.8
BC 35	4.1	3.8	62	0.22	0.66	39	10400	2080	34	48.2
BC 36	2.4	0.4	45.6	0.45	1.41	11	11200	1170	8.1	20.2
BC 37	2.5	2.7	61	0.31	1.26	30	11800	1860	21.4	35.7
BC 38	3	3.2	50.6	0.32	2.68	42	6500	1630	30.4	39.9
BC 39	1.8	7.7	36.6	0.19	0.52	37	4200	720	31.8	33.8

Table 3.3 Continued

Sample	U	Th	Sr	Cd	Sb	V	Ca	P	La	Cr
BC 40	2	1.9	57.9	0.23	2.27	20	10800	960	11.5	19.4
BC 41	2	1.8	78.3	0.18	0.96	18	12700	1270	13	22.5
BC 42	58.4	2.8	67.2	0.27	2.57	22	11100	1170	18.8	24.8
BC 43	2.6	1.3	73.9	0.47	1.29	28	13600	1070	13	27.7
BC 44	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
BC 45	6	18	52.9	0.21	0.52	57	5300	650	44.3	56.1
BC 46	3.1	6.2	42.1	0.17	13.48	35	10200	1350	24.1	38.3
BC 47	3.6	5.6	92.1	0.27	10.17	40	36100	940	23.7	34.7
BC 48	95	1	80.3	0.55	0.83	16	17800	870	10.3	16
BC 49	7.1	0.5	199	0.12	1.48	14	106400	1130	5.8	13.7
BC 50	4.8	0.8	118.3	0.14	0.65	12	42700	1350	7.9	13.4
BC 51	1.7	1.4	68.9	0.21	0.74	16	16200	980	9.7	13.5
BC 52	4	6.1	62	0.18	0.29	28	8500	800	25.2	29.4
BC 53	2.4	2.6	55	0.16	1.71	19	11000	1100	13	18.8
BC 54	2.1	1.5	56.3	0.26	1.92	12	13300	1040	7.7	11.1
BC 55	2.4	2.7	62.9	0.39	2.37	23	12000	1140	15	22.1
BC 56	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
BC 57	1.7	6.1	42	0.21	1.85	41	6600	1300	31	37.4
BC 58	3.3	4.2	48.6	0.24	1.68	32	9100	1120	22.2	36
BC 59	4	2.6	220.6	0.22	2.31	26	33800	1310	13.9	21.3
BC 60	1.3	4.4	24	0.06	0.75	17	3900	590	15.7	18.1
BC 61	1.8	9.1	46.7	0.18	2.15	46	6600	1080	31.4	44.3

Table 3.3 Continued

Sample	Mg	Ba	Ti	Al	Na	K	Sc	Tl	S	Hg
BC 1	2800	79.8	180	9200	220	900	1.5	0.17	18700	0.11
BC 2	1300	919.4	100	20100	100	500	3.3	0.56	3900	0.2
BC 3	2000	96.1	130	7700	300	600	1.1	0.16	19800	0.12
BC 4	1200	60.2	90	8700	120	300	0.7	0.17	8000	0.37
BC 5	2700	88.5	150	9800	220	700	1.6	0.12	9500	0.07
BC 6	2600	85.1	150	8100	300	800	1.5	0.16	12100	0.19
BC 7	1700	75.3	110	4100	190	500	1.3	0.06	8200	0.1
BC 8	3900	164.1	290	8400	200	1300	2.2	0.1	6200	0.1
BC 9	4600	163.1	280	13200	150	1300	2.6	0.12	4800	0.1
BC 10	5000	107.1	230	10000	190	2200	1.5	0.14	6700	0.11
BC 11	3900	49.2	240	6100	100	1100	1.5	0.07	2200	0.03
BC 12	2300	133.7	80	6100	240	700	0.7	0.08	6800	0.16
BC 13	3700	61.5	150	10600	190	1000	1.4	0.11	9300	0.13
BC 14	3000	85.5	120	5000	340	700	1.4	0.07	13600	0.11
BC 15	2600	77.1	150	5400	220	700	1.6	0.07	10600	0.08
BC 16	2800	79.7	160	5200	240	700	1.5	0.07	13900	0.06
BC 17	3500	71.5	100	4800	480	600	1.1	0.07	16500	0.2
BC 18	2700	99.1	70	4500	160	300	1.2	0.06	16000	0.11
BC 19	3000	62.9	70	3900	230	400	1.2	0.07	16600	0.35
BC 20	6200	71.9	190	6900	1330	1000	2.5	0.06	8500	0.06
BC 21	3500	103.8	140	6500	280	900	1.4	0.07	10200	0.09
BC 22	10000	252.2	650	21100	520	4400	5.2	0.28	3400	0.11
BC 23	8800	191.5	610	21500	350	3700	5.4	0.27	2200	0.09
BC 24	2500	92.6	70	6600	160	400	1	0.11	6100	0.14
BC 25	3900	134.9	170	7700	280	1300	1.3	0.08	5800	0.06
BC 26	5800	178.6	270	9200	420	2300	2.5	0.1	6300	0.05
BC 27	6500	200.3	380	14300	360	2700	3.4	0.17	6300	0.09
BC 28	3800	115	220	6700	400	1300	2	0.1	5900	0.04
BC 29	3500	98.9	190	7100	470	1000	1.6	0.11	13100	0.11
BC 30	3800	140.9	230	8400	200	1600	2	0.11	5400	0.06
BC 31	2800	138.9	140	8800	160	900	1.3	0.11	5400	0.11
BC 32	3200	133.4	130	6500	240	800	1.6	0.09	9500	0.12
BC 33	2800	59.9	130	6300	270	600	1.8	0.09	11400	0.1
BC 34	5300	112.1	310	14100	230	1900	2.9	0.15	10200	0.12
BC 35	8500	257.8	460	22200	360	4200	3.9	0.23	4000	0.13
BC 36	3200	106	130	4900	300	600	0.8	0.07	12500	0.21
BC 37	7000	215.4	330	14700	570	3200	2.7	0.17	7200	0.1
BC 38	6600	230	400	18000	430	3200	3.2	0.2	10100	0.13
BC 39	7600	178.2	670	14700	280	5000	3.7	0.2	2900	0.03

Table 3.3 Continued

Sample	Mg	Ba	Ti	Al	Na	K	Sc	Tl	S	Hg
BC 40	4600	116.1	230	8500	380	1400	1.4	0.1	8800	0.13
BC 41	4600	153.1	230	9700	580	1800	2.1	0.1	7300	0.15
BC 42	4800	127.3	250	10200	270	1500	2.1	0.13	6000	0.15
BC 43	6000	177.2	200	10000	630	2000	2.1	0.12	16600	0.07
BC 44	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
BC 45	12200	284.2	970	25500	650	5900	7	0.32	1000	0.02
BC 46	7700	166.2	470	15100	370	3100	3.5	0.18	7500	0.13
BC 47	6900	151.9	450	14900	400	2800	4	0.2	13900	0.08
BC 48	8800	156.3	200	8200	3910	3000	1.4	0.13	5800	0.09
BC 49	5300	170.5	160	4500	680	1400	0.9	0.07	6400	0.04
BC 50	4600	120.2	170	5000	760	1800	1	0.07	7900	0.05
BC 51	4400	120.1	150	5600	310	1300	1.3	0.06	6600	0.07
BC 52	6200	142.9	450	13500	350	2600	3.7	0.15	4100	0.04
BC 53	4400	126.4	210	7900	310	1400	2	0.07	5500	0.08
BC 54	3700	119.8	130	5000	480	1000	1.4	0.05	5000	0.08
BC 55	5000	148.7	260	10500	410	1900	2.5	0.11	4300	0.16
BC 56	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
BC 57	8700	230.8	500	17200	370	3900	3.7	0.19	2200	0.05
BC 58	6100	205.1	350	16200	300	2900	3.4	0.15	4300	0.08
BC 59	9900	209.7	250	10100	1490	3100	2.1	0.1	4100	0.07
BC 60	3500	85.6	290	7200	180	1700	1.8	0.08	1600	0.03
BC 61	10200	204	650	19100	510	4500	4.8	0.23	2500	0.06

Table 3.3 Continued

Sample	Ga	Cs	Hf	Rb	Sn	Zr	Y	Ce	Be	Li
BC 1	1.8	0.96	0.01	8.7	0.2	0.7	11.5	56.9	0.7	11.4
BC 2	3.6	0.74	0.12	4.5	0.2	4.4	25.7	107	2.6	8.1
BC 3	1.6	0.61	0.01	4.7	0.2	1.3	10.74	55.2	1	7.2
BC 4	1.3	0.46	0.09	2.9	0.2	2.1	13.38	27.9	0.4	3.5
BC 5	1.7	0.64	0.04	5.7	0.1	1	8.53	29.8	0.6	7.2
BC 6	1.9	0.88	0.04	6.8	0.2	1.2	7.32	24.6	0.3	11.4
BC 7	0.8	0.61	0.03	4.5	0.1	1.4	5.87	17.5	0.4	6.3
BC 8	2.7	1.76	0.07	13.5	0.3	3.4	6.39	29.1	0.5	13.2
BC 9	4	2.7	0.04	16.8	0.4	2.3	8.44	36.9	0.7	22.6
BC 10	3.2	2.02	0.05	24	0.7	1.7	8.83	35.7	0.2	18.5
BC 11	2.3	0.77	0.04	9.7	0.7	1.8	3.32	23.4	0.2	13.9
BC 12	1.1	0.43	0.08	5.1	0.2	2.4	4.07	13.4	0.1	3.8
BC 13	2.8	0.83	0.01	10.3	0.3	0.9	6.69	27.9	0.6	16.1
BC 14	1.2	0.6	0.01	5.1	0.3	1.8	3.48	14	0.2	7.8
BC 15	1.6	0.61	0.02	6.6	0.3	1.6	5.69	19.8	0.3	7.3
BC 16	1.4	0.58	0.01	6.6	0.2	1.6	5.64	21.3	0.4	8.7
BC 17	1	0.51	0.01	4.1	0.2	1.3	3.42	13	0.2	8.8
BC 18	0.8	0.33	0.03	2.3	0.1	1.4	3.88	10.5	0.1	3.8
BC 19	0.8	0.36	0.01	2.7	0.3	0.9	2.8	9.3	0.1	4.8
BC 20	2	0.47	0.05	4.7	0.2	1.6	2.93	12.4	0.1	12.6
BC 21	1.6	0.7	0.08	7.8	0.2	5.5	4.47	17.9	0.4	6.5
BC 22	7.6	1.99	0.22	39.4	1	10.2	9.91	66.5	0.8	34.7
BC 23	8.1	1.95	0.2	39.9	1	8.4	9.6	65.3	1.3	40
BC 24	1.3	0.5	0.02	4.8	0.2	1.2	4.97	19.4	0.2	2.9
BC 25	2.7	0.76	0.07	12.8	0.4	4.6	4.57	25.4	0.3	10.4
BC 26	3.1	0.95	0.24	17.7	0.4	10.5	5.31	30.2	0.4	15.3
BC 27	5	1.46	0.15	26	0.6	7.9	8.29	47.4	0.7	23.8
BC 28	2.1	0.75	0.08	11.5	0.3	3.7	4.09	22.1	0.4	11.4
BC 29	2.2	0.72	0.06	10	0.2	2.5	4.66	21.9	0.4	11.3
BC 30	2.8	1.03	0.01	15	0.2	1.6	6.9	31.3	0.5	14.9
BC 31	2	0.85	0.05	9.6	0.2	2.2	5.47	24.2	0.4	10
BC 32	1.7	0.67	0.09	8.1	0.2	4.3	3.68	15.3	0.4	6.9
BC 33	1.5	0.64	0.04	6.3	0.1	3	8.05	25.8	0.1	9.5
BC 34	4.2	1.68	0.05	22.1	0.5	3.7	8.29	42.1	0.8	25.4
BC 35	7.6	2.13	0.15	41.1	1	6.5	13.1	68.3	1	42.1
BC 36	1.1	0.59	0.03	5.2	0.1	1.2	3.84	14.9	0.1	8.3
BC 37	4.9	1.69	0.13	28.4	0.5	4.9	7.48	44.8	0.8	31.2
BC 38	6	1.83	0.06	29.9	0.9	4	11.26	62.3	0.9	32.7
BC 39	5.3	1.56	0.1	31.7	0.7	5.2	9.79	68.6	0.6	27.8

Table 3.3 Continued

Sample	Ga	Cs	Hf	Rb	Sn	Zr	Y	Ce	Be	Li
BC 40	2.7	0.92	0.07	13.9	0.3	4.2	4.91	23.8	0.5	12
BC 41	2.9	0.89	0.11	14.9	0.2	5.1	5.75	27	0.2	12.2
BC 42	3.2	1.15	0.18	17.5	0.5	8.8	7.8	36.8	0.7	16
BC 43	2.7	1.13	0.05	17.5	0.3	2.3	5.74	28.2	0.3	16.5
BC 44	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
BC 45	9.9	2.42	0.29	48.1	1.2	14.3	12.85	89.3	0.9	44.6
BC 46	5.4	1.68	0.17	27.4	0.8	8.1	7.73	49.9	0.5	27.4
BC 47	5.2	1.47	0.18	27.9	0.8	9.1	7.99	48.1	0.6	22.7
BC 48	1.8	0.87	0.02	11	0.1	1	4.81	22.2	0.1	17.3
BC 49	1.6	0.67	0.01	9	0.1	0.4	2.69	12.1	0.2	10.1
BC 50	1.6	0.71	0.04	10.5	0.2	1.6	2.99	16	0.4	12
BC 51	1.6	0.58	0.1	9.2	0.2	4.6	3.47	19.6	0.4	7.4
BC 52	5	1.26	0.23	26.4	0.6	10.3	8.28	50.8	0.6	22.1
BC 53	2.3	0.8	0.18	13.7	0.3	8	4.47	26.6	0.3	12.1
BC 54	1.4	0.49	0.09	7	0.2	3.7	2.96	15.5	0.6	5.9
BC 55	3	0.95	0.1	16.6	0.2	5.9	5.41	29.5	0.7	14.4
BC 56	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
BC 57	6.2	1.75	0.08	33.4	0.8	5.1	10.01	64.7	0.5	29.2
BC 58	5.4	1.53	0.14	28.2	0.7	7.9	8	45.4	0.6	25.6
BC 59	3.2	1.01	0.1	17.4	0.4	6.1	5.09	27.9	0.9	19.6
BC 60	2.4	0.8	0.05	14	0.3	2.6	5.19	32.1	0.4	13.1
BC 61	6.6	2.07	0.13	35.3	0.8	7.4	9.73	64.1	0.8	35.4

Table 3.4 Rock Eval. results

Sample	S1	S2	S3	TOC(%)
BC1	46.42	91.27	33.07	25.80
BC2	4.97	36.75	56.37	14.93
BC3	48.46	91.75	36.57	26.44
BC4	40.78	102.40	37.15	26.36
BC5	46.43	105.19	31.25	25.46
BC6	53.09	73.07	36.41	24.34
BC7	60.93	87.10	35.32	23.27
BC8	33.96	76.56	29.54	20.18
BC9	21.48	59.38	23.78	16.97
BC10	19.59	49.00	21.80	13.48
BC11	11.51	14.44	6.03	4.03
BC12	33.90	89.50	41.78	25.44
BC13	40.96	71.18	26.72	20.37
BC14	59.56	111.49	37.79	27.75
BC15	58.48	103.77	32.42	25.56
BC16	54.52	103.47	31.87	26.10
BC17	66.50	88.31	38.53	26.92
BC18	47.50	135.66	37.44	30.13
BC19	50.58	114.26	40.65	28.50
BC20	40.65	64.70	27.95	17.12
BC21	22.94	99.75	43.62	29.34
BC22	9.33	34.89	15.70	11.34
BC23	6.45	24.57	10.62	7.81
BC24	31.11	112.61	41.75	30.90
BC25	30.09	112.29	38.77	27.49
BC26	22.49	102.13	35.78	27.15
BC27	18.93	82.34	26.66	21.34
BC28	50.42	106.88	36.35	24.38
BC29	46.57	101.83	30.65	22.28
BC30	24.24	54.36	16.52	13.01
BC31	36.23	110.68	37.46	26.34
BC32	39.40	116.26	41.41	27.18
BC33	61.25	86.36	37.61	26.65
BC34	32.63	83.35	23.03	19.67
BC35	13.07	50.36	17.36	12.61
BC36	57.20	134.24	39.17	30.42
BC37	27.61	88.94	26.70	20.75
BC38	21.78	54.45	18.38	15.00
BC39	2.23	11.92	7.24	4.16
BC40	31.07	112.22	37.79	28.70
BC41	26.15	98.80	36.79	27.41
BC42	26.39	95.50	36.90	27.81
BC43	35.04	111.46	37.47	27.14
BC44	19.20	54.71	20.36	13.37
BC45	0.40	2.41	2.40	1.39
BC46	18.90	58.30	21.01	16.32
BC47	23.29	73.50	21.00	16.49
BC48	49.76	132.09	39.32	28.34
BC49	46.57	64.56	27.33	18.30
BC50	56.23	83.66	35.00	23.23
BC51	28.05	113.90	41.97	31.45
BC52	13.25	52.47	17.38	14.22
BC53	28.67	104.87	39.36	28.32
BC54	36.39	122.68	44.76	31.97
BC55	26.15	88.52	38.18	26.38
BC56	39.45	97.78	38.64	26.58
BC57	7.35	23.37	11.27	7.18
BC58	15.54	73.69	26.66	19.89
BC59	17.67	76.81	35.75	23.11
BC60	6.54	17.83	7.10	5.00
BC61	7.67	32.14	12.07	8.58

Table 3.5 Nutrient concentrations (in mg/l) of Nitrite, Nitrate, Ammonia, TKN and P

Variable	**Nitrite**	**Nitratre**	**Ammonia**	**TKN**	**P**
Units	**mg/L**	**mg/L**	**mg/L**	**mg/L**	**mg/L**
BC-1	0.052	0.077	8.17	19.6	0.65
BC-2	0.026	0.16	0.34	2.22	0.19
BC-3	0.051	0.027	5.38	14.2	0.55
BC-4	0.048	0.041	1.64	11.3	0.71
BC-5	0.048	0.041	7.44	21	0.94
BC-6	0.047	< 0.020	4.02	9.26	0.31
BC-7	0.027	< 0.020	1.31	1.79	< 0.01
BC-8	0.011	< 0.020	3.29	13.6	0.13
BC-9	0.03	< 0.020	2.97	4	0.13
BC-10	0.043	< 0.020	3.06	6.03	0.87
BC-11	0.021	0.025	1.16	2.58	0.18
BC-12	0.044	< 0.020	2.06	5.22	0.16
BC-13	0.027	0.068	0.88	2.95	0.17
BC-14	0.025	0.03	6.02	16.8	0.57
BC-15	0.034	0.026	7.58	18	0.55
BC-16	0.027	0.033	6.11	17	0.54
BC-17	0.027	0.063	6.08	3.12	0.17
BC-18	NA	NA	NA	NA	NA
BC-19	0.028	0.026	3.52	12	0.93
BC-20	0.02	< 0.020	7.42	13.5	0.14

Table 3.5 Continued

Variable Units	Nitrite mg/L	Nitratre mg/L	Ammonia mg/L	TKN mg/L	P mg/L
BC-21	0.103	< 0.020	34.2	5.09	0.05
BC-22	0.11	0.083	2.8	9.79	0.63
BC-23	0.049	0.106	0.68	3	0.21
BC-24	0.126	0.074	1.24	12	0.93
BC-25	0.047	< 0.020	7.81	17.3	0.12
BC-26	0.023	< 0.020	12.6	31.5	0.69
BC-27	0.021	0.079	1.43	3.64	0.1
BC-28	0.034	< 0.020	4.04	9.54	0.22
BC-29	0.037	< 0.020	5.43	11	0.3
BC-30	0.025	< 0.020	1.98	3.55	0.06
BC-31	0.035	0.028	1.93	10.4	0.42
BC-32	0.033	0.031	3.48	17.1	0.67
BC-33	0.064	0.027	4.07	34.1	1.24
BC-34	0.042	0.4	2.19	25.4	1.51
BC-35	0.016	0.037	0.04	1.82	0.15
BC-36	INS.	INS.	NA	NA	NA
BC-37	0.014	< 0.020	0.02	19.5	1.66
BC-38	0.125	< 0.020	6.36	14.2	1.09
BC-39	0.009	< 0.020	0.1	6.67	0.96
BC-40	0.029	0.278	2.54	20.9	0.74

Table 3.5 Continued

Variable Units	Nitrite mg/L	Nitratre mg/L	Ammonia mg/L	TKN mg/L	P mg/L
BC-41	0.024	< 0.020	4.98	8.9	0.18
BC-42	0.039	0.438	0.18	59	2.58
BC-43	0.07	0.409	2.07	18.2	0.68
BC-44	0.019	< 0.020	13	30.7	0.66
BC-45	0.044	0.154	0.57	3.53	0.55
BC-46	0.034	0.36	0.3	7.54	0.51
BC-47	0.064	0.046	2.03	15.4	1.09
BC-48	0.036	< 0.020	5.06	17.1	0.51
BC-49	0.089	0.073	3.58	26.6	1.14
BC-50	0.018	0.044	4.3	26.6	0.85
BC-51	0.027	0.031	4.84	13	0.35
BC-52	0.023	< 0.020	6.81	9.69	0.14
BC-53	0.03	< 0.020	4.53	7.45	0.1
BC-54	0.021	0.023	2.24	12.7	0.34
BC-55	0.019	0.191	1.18	3.48	0.1
BC-56	0.023	< 0.020	1.44	3.7	0.13
BC-57	0.018	0.036	0.21	0.91	0.07
BC-58	0.017	0.042	1.85	3.07	0.07
BC-59	0.022	0.04	0.03	2.79	0.1
BC-60	0.016	0.023	2.96	4.63	0.09
BC-61	0.015	0.185	0.31	3.22	0.23

3.3 Arcellacean analysis

Micropaleontological samples (2.5 cc) were first wet sieved through a 297-µm mesh. This was to remove any coarse debris such as grass and sticks. Then the sample went through a 37- µm mesh to retain arcellaceans and remove the fine material. Samples were immediately placed in formalin and were refrigerated in order to avoid decay.

Using the wet-splitter (Scott and Hermlin. 1993), the samples were subdivided into six aliquots for the purpose of quantitative analysis. The aliquots were then placed in a gridded Petri Dish and arcellaceans were counted using an Olympus SZH dissecting binocular microscope (7.5-64X magnification) until, whenever possible, a statistically significant number of specimens were quantified (Table 3.6; Patterson and Fishbein, 1989).

Identification of arcellaceans primarily followed standard reference key of Kumar and Dalby, 1998, although references were also made to photoplates and descriptions in various publications, notably Medioli and Scott, 1983, Reinhardt et al., 1998 and Roe et al., 2010. The infrasubspecific designation “strain” was used in the identification process. Although this level has no status under the Zoological Code of Nomenclature (ICZN, 1999), it has proved to be useful in distinguishing subenvironemtns not recognized by species (Reinhardt et. al., 1998; Kauppila et. al., 2006; Kumar and Patterson, 2000).

Scanning electron microscope images of common species and strains were obtained using a Tescan Vega-II XMU VP scanning electron microscope at the Carleton University SEM facility (Plate. 1, 2). All plates were digitally produced using Adobe Photoshop™ CS12 on an Apple Macintosh® computer.

Table 3.6 Relative abundances of 29 arcellacean species and strains.

Sample Code	Total Counts	Test/cc	SDI	CAA %	CAD %	CCA %	CCC %	CCS %	CP %	CT %
BC 1	302	120.8	1.91	0.000	0.020	0.225	0.175	0.013	0.000	0.040
BC 2	190	76	1.69	0.005	0.005	0.005	0.005	0.000	0.000	0.000
BC 3	225	90	1.73	0.013	0.000	0.299	0.112	0.000	0.000	0.018
BC 4	302	120.8	1.42	0.000	0.000	0.144	0.017	0.007	0.000	0.010
BC 5	341	136.4	1.96	0.012	0.003	0.355	0.157	0.006	0.012	0.006
BC 6	187	74.8	2.36	0.038	0.038	0.194	0.102	0.016	0.011	0.000
BC 7	226	90.4	2.07	0.013	0.009	0.279	0.142	0.018	0.009	0.022
BC 8	319	127.6	2.57	0.060	0.025	0.183	0.126	0.028	0.041	0.044
BC 9	300	120	2.6	0.021	0.028	0.124	0.131	0.010	0.017	0.062
BC 10	26	10.4	1.3	0.000	0.077	0.231	0.000	0.000	0.000	0.000
BC 11	304	121.6	2.1	0.007	0.010	0.188	0.112	0.030	0.003	0.000
BC 12	235	94	2.23	0.030	0.017	0.263	0.168	0.034	0.026	0.017
BC 13	268	107.2	2.16	0.007	0.000	0.224	0.093	0.007	0.004	0.015
BC 14	165	66	1.55	0.036	0.000	0.279	0.139	0.012	0.000	0.012
BC 15	351	140.4	1.49	0.000	0.000	0.333	0.140	0.009	0.000	0.037
BC 16	294	117.6	1.46	0.003	0.007	0.201	0.065	0.003	0.000	0.031
BC 17	291	116.4	1.53	0.010	0.031	0.368	0.203	0.151	0.007	0.003
BC 18	213	85.2	1.84	0.099	0.117	0.352	0.221	0.047	0.019	0.000
BC 19	226	90.4	1.7	0.013	0.090	0.345	0.157	0.045	0.040	0.000
BC 20	6	2.4	1.56	0.000	0.167	0.167	0.167	0.000	0.000	0.000
BC 21	257	102.8	2.62	0.137	0.141	0.051	0.055	0.078	0.012	0.047
BC 22	145	58	1.93	0.050	0.050	0.025	0.000	0.000	0.050	0.450
BC 23	198	79.2	2	0.000	0.005	0.046	0.000	0.000	0.000	0.036
BC 24	177	70.8	1.64	0.000	0.023	0.130	0.011	0.040	0.011	0.045
BC 25	196	78.4	2.47	0.087	0.026	0.015	0.062	0.015	0.000	0.313
BC 26	214	85.6	2.31	0.107	0.042	0.033	0.028	0.019	0.000	0.318
BC 27	299	119.6	1.88	0.023	0.003	0.000	0.003	0.003	0.000	0.030
BC 28	259	103.6	1.57	0.000	0.000	0.344	0.050	0.042	0.000	0.000
BC 29	203	81.2	1.11	0.000	0.010	0.232	0.054	0.000	0.000	0.000
BC 30	266	106.4	2.45	0.030	0.064	0.154	0.079	0.000	0.004	0.068
BC 31	279	111.6	2.37	0.011	0.004	0.133	0.058	0.000	0.018	0.072
BC 32	256	102.4	1.5	0.004	0.000	0.227	0.117	0.000	0.000	0.070
BC 33	218	87.2	1.32	0.032	0.014	0.459	0.092	0.000	0.000	0.009
BC 34	235	94	1.78	0.000	0.000	0.196	0.064	0.000	0.000	0.085
BC 35	210	84	1.65	0.005	0.000	0.070	0.000	0.000	0.000	0.313
BC 36	278	111.2	1.07	0.004	0.000	0.183	0.140	0.000	0.000	0.011
BC 37	307	122.8	1.89	0.000	0.000	0.333	0.000	0.000	0.205	0.103

Table 3.6 Continued

Sample Code	Total Counts	Test/cc	SDI	CAA %	CAD %	CCA %	CCC %	CCS %	CP %	CT %
BC 38	244	97.6	2.09	0.033	0.004	0.111	0.082	0.012	0.000	0.086
BC 39	287	114.8	2.12	0.035	0.017	0.181	0.122	0.038	0.007	0.021
BC 40	208	83.2	2.2	0.020	0.010	0.059	0.044	0.034	0.000	0.244
BC 41	181	72.4	2.33	0.033	0.000	0.039	0.067	0.000	0.000	0.244
BC 42	181	72.4	2.43	0.050	0.011	0.094	0.028	0.000	0.000	0.276
BC 43	224	89.6	1.75	0.022	0.000	0.027	0.076	0.027	0.000	0.192
BC 44	238	95.2	1.44	0.324	0.008	0.025	0.387	0.189	0.000	0.025
BC 45	204	81.6	2.25	0.078	0.052	0.031	0.047	0.047	0.005	0.383
BC 46	297	118.8	1.93	0.071	0.003	0.000	0.000	0.000	0.209	0.114
BC 47	313	125.2	2.07	0.249	0.006	0.064	0.265	0.070	0.000	0.125
BC 48	10	4	1.16	0.000	0.000	0.500	0.100	0.100	0.000	0.000
BC 49	245	98	1.79	0.106	0.000	0.188	0.216	0.012	0.000	0.049
BC 50	175	70	1.89	0.189	0.000	0.324	0.081	0.000	0.000	0.027
BC 51	238	95.2	2.17	0.207	0.017	0.017	0.042	0.004	0.000	0.338
BC 52	268	107.2	2.27	0.120	0.000	0.056	0.045	0.000	0.007	0.300
BC 53	254	101.6	2.31	0.020	0.008	0.004	0.028	0.000	0.008	0.281
BC 54	152	60.8	1.846	0.026	0.000	0.066	0.013	0.000	0.000	0.270
BC 55	143	57.2	2.09	0.140	0.000	0.007	0.028	0.000	0.000	0.343
BC 56	186	74.4	2.06	0.043	0.005	0.130	0.038	0.000	0.000	0.114
BC 57	271	108.4	1.97	0.007	0.000	0.063	0.004	0.000	0.004	0.107
BC 58	248	99.2	2.27	0.000	0.000	0.012	0.020	0.000	0.000	0.129
BC 59	302	120.8	1.94	0.017	0.003	0.115	0.014	0.000	0.153	0.477
BC 60	283	113.2	2.23	0.049	0.004	0.053	0.071	0.000	0.000	0.166
BC 61	325	130	2.11	0.006	0.000	0.006	0.000	0.003	0.096	0.253

Table 3.6 Continued

Sample Code	Total Counts	Test/cc	SDI	DB %	DM %	DGG %	DGD %	DOL %	DU %	DOO %
BC 1	302	120.8	1.91	0.017	0.000	0.000	0.000	0.000	0.000	0.089
BC 2	190	76	1.69	0.000	0.000	0.473	0.011	0.005	0.000	0.239
BC 3	225	90	1.73	0.000	0.000	0.049	0.004	0.000	0.000	0.040
BC 4	302	120.8	1.42	0.000	0.000	0.040	0.007	0.000	0.000	0.013
BC 5	341	136.4	1.96	0.000	0.000	0.003	0.003	0.000	0.000	0.071
BC 6	187	74.8	2.36	0.011	0.000	0.177	0.000	0.027	0.000	0.118
BC 7	226	90.4	2.07	0.000	0.000	0.080	0.000	0.000	0.000	0.040
BC 8	319	127.6	2.57	0.000	0.041	0.009	0.000	0.000	0.000	0.142
BC 9	300	120	2.6	0.000	0.010	0.010	0.000	0.000	0.000	0.214
BC 10	26	10.4	1.3	0.000	0.000	0.308	0.000	0.000	0.000	0.346
BC 11	304	121.6	2.1	0.000	0.003	0.112	0.003	0.000	0.000	0.139
BC 12	235	94	2.23	0.000	0.000	0.060	0.004	0.000	0.000	0.000
BC 13	268	107.2	2.16	0.000	0.000	0.131	0.000	0.000	0.000	0.022
BC 14	165	66	1.55	0.000	0.000	0.030	0.000	0.000	0.000	0.042
BC 15	351	140.4	1.49	0.000	0.000	0.026	0.000	0.000	0.000	0.006
BC 16	294	117.6	1.46	0.000	0.000	0.003	0.000	0.000	0.000	0.017
BC 17	291	116.4	1.53	0.000	0.000	0.003	0.000	0.000	0.000	0.000
BC 18	213	85.2	1.84	0.000	0.000	0.000	0.000	0.000	0.000	0.009
BC 19	226	90.4	1.7	0.004	0.000	0.000	0.000	0.000	0.000	0.004
BC 20	6	2.4	1.56	0.000	0.000	0.167	0.000	0.000	0.000	0.000
BC 21	257	102.8	2.62	0.000	0.012	0.012	0.000	0.012	0.000	0.074
BC 22	145	58	1.93	0.000	0.000	0.000	0.000	0.000	0.000	0.250
BC 23	198	79.2	2	0.021	0.000	0.185	0.000	0.067	0.005	0.446
BC 24	177	70.8	1.64	0.000	0.000	0.006	0.000	0.000	0.000	0.565
BC 25	196	78.4	2.47	0.000	0.062	0.031	0.005	0.010	0.000	0.082
BC 26	214	85.6	2.31	0.000	0.019	0.028	0.000	0.028	0.000	0.145
BC 27	299	119.6	1.88	0.000	0.000	0.003	0.000	0.431	0.000	0.167
BC 28	259	103.6	1.57	0.000	0.000	0.000	0.000	0.008	0.000	0.077
BC 29	203	81.2	1.11	0.000	0.000	0.000	0.000	0.000	0.000	0.039
BC 30	266	106.4	2.45	0.008	0.011	0.094	0.000	0.000	0.000	0.162
BC 31	279	111.6	2.37	0.000	0.000	0.040	0.000	0.000	0.000	0.079
BC 32	256	102.4	1.5	0.000	0.000	0.008	0.000	0.000	0.000	0.027
BC 33	218	87.2	1.32	0.000	0.000	0.000	0.000	0.000	0.000	0.018
BC 34	235	94	1.78	0.000	0.000	0.106	0.000	0.000	0.000	0.043
BC 35	210	84	1.65	0.000	0.000	0.413	0.000	0.000	0.000	0.109
BC 36	278	111.2	1.07	0.000	0.000	0.011	0.000	0.000	0.000	0.000
BC 37	307	122.8	1.89	0.000	0.000	0.128	0.000	0.077	0.000	0.077

Table 3.6 Continued

Sample Code	Total Counts	Test/cc	SDI	DB %	DM %	DGG %	DGD %	DOL %	DU %	DOO %
BC 38	244	97.6	2.09	0.000	0.000	0.238	0.000	0.012	0.000	0.041
BC 39	287	114.8	2.12	0.000	0.000	0.042	0.000	0.000	0.000	0.366
BC 40	208	83.2	2.2	0.000	0.000	0.107	0.000	0.000	0.020	0.034
BC 41	181	72.4	2.33	0.000	0.006	0.067	0.000	0.000	0.006	0.056
BC 42	181	72.4	2.43	0.000	0.000	0.144	0.000	0.000	0.000	0.066
BC 43	224	89.6	1.75	0.000	0.000	0.031	0.000	0.000	0.000	0.018
BC 44	238	95.2	1.44	0.000	0.000	0.000	0.000	0.000	0.000	0.004
BC 45	204	81.6	2.25	0.000	0.005	0.166	0.056	0.000	0.000	0.026
BC 46	297	118.8	1.93	0.000	0.000	0.017	0.000	0.384	0.000	0.020
BC 47	313	125.2	2.07	0.000	0.000	0.003	0.000	0.042	0.003	0.045
BC 48	10	4	1.16	0.000	0.000	0.000	0.000	0.000	0.000	0.000
BC 49	245	98	1.79	0.000	0.000	0.000	0.000	0.000	0.000	0.037
BC 50	175	70	1.89	0.000	0.000	0.000	0.000	0.000	0.000	0.000
BC 51	238	95.2	2.17	0.000	0.046	0.000	0.000	0.017	0.004	0.122
BC 52	268	107.2	2.27	0.000	0.007	0.011	0.003	0.015	0.000	0.139
BC 53	254	101.6	2.31	0.000	0.000	0.079	0.000	0.012	0.000	0.075
BC 54	152	60.8	1.846	0.000	0.000	0.000	0.000	0.000	0.000	0.066
BC 55	143	57.2	2.09	0.000	0.000	0.098	0.000	0.014	0.000	0.042
BC 56	186	74.4	2.06	0.000	0.005	0.000	0.000	0.000	0.000	0.292
BC 57	271	108.4	1.97	0.000	0.000	0.240	0.000	0.000	0.011	0.310
BC 58	248	99.2	2.27	0.000	0.000	0.073	0.000	0.222	0.016	0.169
BC 59	302	120.8	1.94	0.000	0.000	0.024	0.000	0.122	0.000	0.024
BC 60	283	113.2	2.23	0.000	0.004	0.046	0.000	0.000	0.000	0.276
BC 61	325	130	2.11	0.000	0.000	0.130	0.003	0.275	0.003	0.040

Table 3.6 Continued

Sample Code	Total Counts	Test/cc	SDI	DOS %	DOT %	DPP %	DPAC %	DPCL %	DPCR %
BC 1	302	120.8	1.91	0.000	0.000	0.007	0.033	0.003	0.000
BC 2	190	76	1.69	0.005	0.000	0.000	0.128	0.000	0.037
BC 3	225	90	1.73	0.004	0.000	0.013	0.000	0.000	0.004
BC 4	302	120.8	1.42	0.000	0.000	0.003	0.057	0.054	0.000
BC 5	341	136.4	1.96	0.009	0.006	0.003	0.015	0.041	0.000
BC 6	187	74.8	2.36	0.000	0.091	0.011	0.022	0.000	0.016
BC 7	226	90.4	2.07	0.000	0.000	0.062	0.031	0.000	0.022
BC 8	319	127.6	2.57	0.025	0.000	0.025	0.032	0.000	0.016
BC 9	300	120	2.6	0.048	0.000	0.007	0.048	0.014	0.000
BC 10	26	10.4	1.3	0.000	0.038	0.000	0.000	0.000	0.000
BC 11	304	121.6	2.1	0.007	0.023	0.036	0.013	0.000	0.017
BC 12	235	94	2.23	0.000	0.026	0.004	0.065	0.047	0.000
BC 13	268	107.2	2.16	0.000	0.097	0.019	0.037	0.000	0.034
BC 14	165	66	1.55	0.000	0.000	0.000	0.000	0.000	0.006
BC 15	351	140.4	1.49	0.000	0.000	0.023	0.000	0.000	0.009
BC 16	294	117.6	1.46	0.000	0.014	0.007	0.017	0.000	0.007
BC 17	291	116.4	1.53	0.000	0.000	0.000	0.000	0.000	0.000
BC 18	213	85.2	1.84	0.000	0.000	0.000	0.000	0.000	0.000
BC 19	226	90.4	1.7	0.000	0.004	0.000	0.000	0.000	0.000
BC 20	6	2.4	1.56	0.000	0.000	0.000	0.000	0.000	0.000
BC 21	257	102.8	2.62	0.004	0.043	0.000	0.027	0.008	0.027
BC 22	145	58	1.93	0.000	0.000	0.000	0.000	0.000	0.000
BC 23	198	79.2	2	0.031	0.036	0.000	0.010	0.036	0.005
BC 24	177	70.8	1.64	0.006	0.000	0.000	0.045	0.000	0.045
BC 25	196	78.4	2.47	0.005	0.021	0.051	0.026	0.010	0.067
BC 26	214	85.6	2.31	0.014	0.005	0.009	0.000	0.000	0.023
BC 27	299	119.6	1.88	0.027	0.003	0.107	0.067	0.023	0.080
BC 28	259	103.6	1.57	0.000	0.008	0.004	0.008	0.000	0.000
BC 29	203	81.2	1.11	0.000	0.000	0.000	0.000	0.000	0.000
BC 30	266	106.4	2.45	0.000	0.015	0.011	0.049	0.000	0.019
BC 31	279	111.6	2.37	0.000	0.025	0.018	0.072	0.011	0.040
BC 32	256	102.4	1.5	0.000	0.000	0.000	0.012	0.000	0.000
BC 33	218	87.2	1.32	0.000	0.000	0.000	0.005	0.000	0.000
BC 34	235	94	1.78	0.021	0.004	0.000	0.009	0.000	0.000
BC 35	210	84	1.65	0.000	0.000	0.000	0.025	0.000	0.000
BC 36	278	111.2	1.07	0.000	0.000	0.000	0.000	0.000	0.000
BC 37	307	122.8	1.89	0.026	0.000	0.000	0.000	0.000	0.000

Table 3.6 Continued

Sample Code	Total Counts	Test/cc	SDI	DOS %	DOT %	DPP %	DPA %	DPCL %	DPCU %
BC 38	244	97.6	2.09	0.000	0.041	0.000	0.008	0.012	0.008
BC 39	287	114.8	2.12	0.000	0.024	0.000	0.000	0.014	0.003
BC 40	208	83.2	2.2	0.005	0.010	0.005	0.039	0.005	0.000
BC 41	181	72.4	2.33	0.011	0.017	0.028	0.022	0.011	0.017
BC 42	181	72.4	2.43	0.011	0.006	0.000	0.028	0.011	0.050
BC 43	224	89.6	1.75	0.000	0.000	0.000	0.004	0.000	0.000
BC 44	238	95.2	1.44	0.000	0.000	0.000	0.000	0.000	0.000
BC 45	204	81.6	2.25	0.000	0.000	0.000	0.010	0.010	0.000
BC 46	297	118.8	1.93	0.007	0.000	0.034	0.061	0.017	0.017
BC 47	313	125.2	2.07	0.000	0.000	0.006	0.010	0.013	0.003
BC 48	10	4	1.16	0.000	0.000	0.000	0.000	0.000	0.000
BC 49	245	98	1.79	0.000	0.000	0.000	0.000	0.000	0.000
BC 50	175	70	1.89	0.000	0.000	0.000	0.000	0.000	0.000
BC 51	238	95.2	2.17	0.000	0.000	0.017	0.004	0.008	0.008
BC 52	268	107.2	2.27	0.000	0.000	0.007	0.064	0.056	0.004
BC 53	254	101.6	2.31	0.000	0.004	0.032	0.024	0.012	0.016
BC 54	152	60.8	1.85	0.000	0.007	0.007	0.000	0.000	0.000
BC 55	143	57.2	2.09	0.000	0.000	0.007	0.007	0.014	0.014
BC 56	186	74.4	2.06	0.000	0.000	0.011	0.005	0.000	0.000
BC 57	271	108.4	1.97	0.000	0.085	0.000	0.007	0.000	0.000
BC 58	248	99.2	2.27	0.000	0.000	0.129	0.036	0.012	0.028
BC 59	302	120.8	1.94	0.007	0.000	0.003	0.000	0.000	0.000
BC 60	283	113.2	2.23	0.004	0.000	0.004	0.014	0.004	0.049
BC 61	325	130	2.11	0.000	0.015	0.040	0.003	0.000	0.000

Table 3.6 Continued

Sample Code	Total Counts	Test/cc	SDI	DPSC %	DA %	DUU %	DUE %	HS %	LS %	LV %	PC %
BC 1	302	120.8	1.91	0.007	0.334	0.007	0.000	0.000	0.017	0.010	0.000
BC 2	190	76	1.69	0.000	0.011	0.005	0.000	0.011	0.021	0.000	0.037
BC 3	225	90	1.73	0.000	0.348	0.098	0.000	0.000	0.000	0.000	0.000
BC 4	302	120.8	1.42	0.000	0.625	0.017	0.000	0.000	0.000	0.000	0.003
BC 5	341	136.4	1.96	0.015	0.243	0.003	0.000	0.000	0.009	0.009	0.000
BC 6	187	74.8	2.36	0.000	0.118	0.005	0.000	0.000	0.000	0.005	0.000
BC 7	226	90.4	2.07	0.000	0.252	0.009	0.000	0.000	0.009	0.004	0.000
BC 8	319	127.6	2.57	0.013	0.132	0.022	0.000	0.000	0.028	0.006	0.000
BC 9	300	120	2.6	0.034	0.138	0.014	0.000	0.000	0.024	0.021	0.024
BC 10	26	10.4	1.3	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
BC 11	304	121.6	2.1	0.003	0.287	0.000	0.000	0.000	0.010	0.000	0.000
BC 12	235	94	2.23	0.004	0.207	0.004	0.000	0.000	0.000	0.000	0.000
BC 13	268	107.2	2.16	0.000	0.261	0.004	0.000	0.011	0.022	0.007	0.004
BC 14	165	66	1.55	0.000	0.430	0.000	0.000	0.000	0.006	0.006	0.000
BC 15	351	140.4	1.49	0.000	0.405	0.000	0.000	0.000	0.003	0.006	0.000
BC 16	294	117.6	1.46	0.024	0.578	0.000	0.000	0.000	0.020	0.003	0.000
BC 17	291	116.4	1.53	0.000	0.223	0.000	0.000	0.000	0.000	0.000	0.000
BC 18	213	85.2	1.84	0.000	0.056	0.000	0.000	0.000	0.005	0.005	0.000
BC 19	226	90.4	1.7	0.000	0.291	0.000	0.000	0.000	0.000	0.000	0.000
BC 20	6	2.4	1.56	0.000	0.333	0.000	0.000	0.000	0.000	0.000	0.000
BC 21	257	102.8	2.62	0.000	0.191	0.000	0.023	0.000	0.016	0.004	0.004
BC 22	145	58	1.93	0.000	0.050	0.000	0.050	0.000	0.025	0.000	0.000
BC 23	198	79.2	2	0.000	0.000	0.005	0.026	0.000	0.005	0.010	0.026
BC 24	177	70.8	1.64	0.000	0.051	0.000	0.006	0.000	0.000	0.011	0.000
BC 25	196	78.4	2.47	0.000	0.051	0.000	0.000	0.000	0.051	0.005	0.000
BC 26	214	85.6	2.31	0.000	0.121	0.005	0.009	0.000	0.014	0.014	0.000
BC 27	299	119.6	1.88	0.000	0.010	0.000	0.000	0.000	0.003	0.000	0.000
BC 28	259	103.6	1.57	0.000	0.402	0.000	0.000	0.000	0.012	0.015	0.031
BC 29	203	81.2	1.11	0.000	0.631	0.000	0.000	0.000	0.015	0.010	0.010
BC 30	266	106.4	2.45	0.000	0.173	0.008	0.000	0.000	0.038	0.008	0.008
BC 31	279	111.6	2.37	0.000	0.306	0.000	0.000	0.000	0.058	0.007	0.011
BC 32	256	102.4	1.5	0.004	0.500	0.004	0.000	0.000	0.012	0.012	0.000
BC 33	218	87.2	1.32	0.000	0.358	0.000	0.000	0.000	0.009	0.005	0.000
BC 34	235	94	1.78	0.000	0.426	0.000	0.000	0.000	0.026	0.013	0.004
BC 35	210	84	1.65	0.000	0.040	0.000	0.000	0.000	0.000	0.010	0.015
BC 36	278	111.2	1.07	0.000	0.633	0.000	0.000	0.000	0.000	0.000	0.000
BC 37	307	122.8	1.89	0.000	0.026	0.000	0.000	0.000	0.000	0.000	0.026

Table 3.6 Continued

Sample Code	Total Counts	Test/cc	SDI	DS %	DA %	DUU %	DUE %	HS %	LS %	LV %	PC %
BC 38	244	97.6	2.09	0.000	0.291	0.000	0.000	0.004	0.012	0.000	0.000
BC 39	287	114.8	2.12	0.000	0.028	0.000	0.000	0.007	0.045	0.028	0.017
BC 40	208	83.2	2.2	0.005	0.317	0.000	0.000	0.000	0.024	0.005	0.015
BC 41	181	72.4	2.33	0.000	0.250	0.000	0.000	0.000	0.000	0.011	0.006
BC 42	181	72.4	2.43	0.000	0.072	0.000	0.000	0.000	0.033	0.022	0.033
BC 43	224	89.6	1.75	0.000	0.478	0.000	0.000	0.000	0.045	0.000	0.027
BC 44	238	95.2	1.44	0.000	0.021	0.000	0.000	0.000	0.000	0.000	0.000
BC 45	204	81.6	2.25	0.000	0.016	0.031	0.000	0.000	0.021	0.005	0.021
BC 46	297	118.8	1.93	0.000	0.037	0.003	0.000	0.000	0.000	0.003	0.000
BC 47	313	125.2	2.07	0.000	0.086	0.000	0.000	0.000	0.000	0.000	0.000
BC 48	10	4	1.16	0.000	0.300	0.000	0.000	0.000	0.000	0.000	0.000
BC 49	245	98	1.79	0.000	0.335	0.000	0.000	0.000	0.033	0.012	0.012
BC 50	175	70	1.89	0.000	0.270	0.000	0.000	0.000	0.081	0.000	0.027
BC 51	238	95.2	2.17	0.000	0.034	0.046	0.000	0.000	0.042	0.013	0.000
BC 52	268	107.2	2.27	0.000	0.094	0.000	0.000	0.000	0.037	0.030	0.004
BC 53	254	101.6	2.31	0.000	0.229	0.008	0.000	0.000	0.028	0.008	0.004
BC 54	152	60.8	1.846	0.013	0.270	0.000	0.000	0.000	0.013	0.013	0.000
BC 55	143	57.2	2.09	0.000	0.126	0.000	0.000	0.000	0.028	0.035	0.000
BC 56	186	74.4	2.06	0.000	0.227	0.000	0.000	0.000	0.038	0.016	0.038
BC 57	271	108.4	1.97	0.000	0.103	0.000	0.000	0.000	0.011	0.011	0.022
BC 58	248	99.2	2.27	0.000	0.101	0.012	0.000	0.000	0.028	0.004	0.000
BC 59	302	120.8	1.94	0.000	0.007	0.000	0.000	0.000	0.000	0.000	0.000
BC 60	283	113.2	2.23	0.000	0.173	0.000	0.000	0.007	0.021	0.046	0.011
BC 61	325	130	2.11	0.000	0.025	0.003	0.000	0.000	0.000	0.022	0.019

3.4 Statistical analysis

Twenty-nine arcellacean species and strains were identified in the 61 quantified samples. The probable error (pe) was calculated for each sample using the following formula:

$$\mathrm{pe} = 1.96\left(\frac{s}{\sqrt{Xi}}\right)$$

where *S* is the standard deviation of the population count and *Xi* is the fractional abundance (Patterson and Fisbein, 1989). The sample was deemed statistically insignificant if the probable error exceeded the total count for a sample. Three samples (BC 10, 20 and 48) contained statistically insignificant population, and were therefore removed from ensuing multivariate data analysis. Standard error (Sxi) was calculated for each sample using the following equation:

$$Sxi = 1.96\sqrt{\frac{F1(1-F1)}{Ni}}$$

where *F*1 is the fractional abundance of species and *Ni* is the total number of counts. Species were considered to be present in insignificant number if the standard error exceeded the total counts for that species in all samples (Patterson and Fisbein, 1989). Out of the 29 species and strains, 24 were found present in statistically significant numbers.

The Shannon Diversity Index (SDI) was used to examine the faunal diversity of the species found in each sample to provide a general indication of the relative health of the lakes and ponds (Shannon, 1948). The SDI is defined as:

$$\text{S.I.} = -\sum_{i=1}^{S}\left(\frac{Xi}{Ni}\right) \times \ln\left(\frac{Xi}{Ni}\right)$$

where *Xi* is the abundance of each taxon in a sample, *Ni* is the total abundance of the sample, and *S* is equal to the species richness of the sample. Environments are considered to be stable if the SDI falls between 2.5 and 3.5, in transition between 1.5 and 2.5, and stressed between 0.1 and 1.5 (Magurran 1988; Patterson and Kumar 2000a)

3.4.1 Data screening

The species and environmental data (e.g. environmental parameters, ICP-MS, grain size, nutrients and Rock Eval.) were screened prior to statistical analysis. Because variables with missing values can cause problems for statistical data analysis (Riemann et al., 2008), they were removed from all data sets. In total, six samples were removed from the environmental data set due to missing values. For the environmental data sets, any variables with more than 25% values above or below detection limits were also removed. The same six samples were also removed from the species data to satisfy the requirement of the ordination analysis used in this study (requirement: the number of samples must be similar in all used data set). Any remaining value below detection limit was converted to half the detection limit of that variable, while anything exceeding the detection limit was changed to the detection limit maximum following Riemann et al., (2008). The units of all the variables in the environmental data were converted, wherever possible, to parts per million (ppm).

3.4.2 Data transformation

The Shapiro-Wilk normality test was performed on the ICP-MS data to determine if data transformation was needed. As expected from visual examination of the geochemical data, the distribution of the variables exhibited high skewness. Therefore, the data set was log-transformed to shift the distribution of the variables closer to normality. The Hellinger transformation was used on the arcellacean data set following analysis of the Detrended Correspondence Analysis (DCA) results (Section 3.4.6).

3.4.3 Variables reduction

Large ecological data sets are often problematic to analyze as they may contain; (1) redundancies in environmental information, and (2) environmental variables that have no influence on the distribution of arcellaceans (Birks et al., 1990). To deal with these problems, a protocol comprised of two stages statistical testing was implemented. In the first stage, highly correlated variables with no clear impact on arcellaceans were removed using visual inspection of a Principle Component Analysis (PCA) bi-plot (not shown). The collinearity of the remaining variables was then determined during the second stage of statistical testing using the Variance Inflation Factor (VIF), which is a component of the usdm package in R statistical programming environment. In this procedure the VIF (or collinearity) value is calculated for each variable. The program then removes any variables that exceed a pre-determined VIF cut off value. Any variables with VIF >10 were deemed to be highly collinear, and were thus eliminated from subsequent analysis.

3.4.4 Cluster analysis

Q-mode cluster analysis was used to group statistically similar populations using Ward's Minimum variance method, and recorded as squared-Euclidean distances (Fishbein and Patterson 1993). Following the same method, R-mode cluster analysis was utilized to determine which species are most closely associated with each other's and thus best characterized a particular assemblage (Fishbein and Patterson 1993). Q-mode and R-mode cluster analyses were carried out on the 24 arcellacean species and strains in the 52 samples determined to have statistically significant counts and not missing any values in the environmental data set. The results were organized into a hierarchical dendrogram.

3.4.5 Detrended Correspondence Analysis (DCA)

Detrended Correspondence Analysis (DCA) was used to compare the similarity between identified assemblages in multidimensional space. DCA revealed that the gradient length of the species data represented a unimodal response (>2). The species data were therefore Hellinger-transformed to satisfy the assumption of the linear-based ordination, Redundancy Analysis (RDA) used in this study.

3.4.6 Redundancy analysis (RDA)

Redundancy analysis (RDA) was used on the 52 samples and 24 statistically significant species and strains to assess the relationship between arcellacean assemblages and measured environmental variables. This analysis provided important insight for interpreting the cluster analysis and DCA results. A series of partial RDAs (pRDA) using the variance portioning test were carried out to identify the significance of the RDA axes. To determine the number of axes that needed to be retained, a scree plot was generated. A scree plot is a simple line segment plot that shows the fraction of total variance in the data represented by each RDA axis. The plot can also show an elbow-like separation between significant and less significant ones. Only axes above the separation were retained. Moreover, variance partition provided an additional quantification of the proportion of the variance in the arcellacean data set that can be attributed to the measured environmental variables. Variables with a p-value < 0.05 were deemed to be significantly contributing to the variance in the arcellacean assemblage.

4. Results

4.1 Cluster analysis

Previous research has demonstrated that there is a significant relationship between various arcellacean species and assemblage distribution, and limnological, climatic and environmental conditions (e.g. oxygen level, minimum average water temperature, levels of organics, clastics and pollution; Scott and Medioli, 1983; Patterson et al., 1985; Medioli et al., 1990; Collins et al., 1990; Asioli, 1996; Patterson et al., 1996; Reinhardt, 1998). Similar environmental relationships were recognized in this study.

The Q-mode cluster analysis, performed on 52 sediment/water interface samples, strongly suggested that five distinct arcellacean assemblages are represented in the data: (1) Arsenic Contaminated Substrate Assemblage; (2) Deep Transitional Assemblage; (3) Shallow Transitional Assemblage; (4) *Difflugia oblonga* Assemblage; and (5) Deep Substrate Assemblage (Figure 4.1). Each assemblage was named after the most characteristic variables, species or environmental condition that described the grouping. Although 24 species of arcellaceans were included in the analyzed data, R-mode cluster analysis indicated that only six species and strains significantly influence assemblage composition; *Difflugia protaeiformis* (Ehrenberg 1830) strain "amorphalis", *Centropyxis constricta* (Ehrenberg 1843) strain "aerophila", *Centropyxis constricta* (Ehrenberg 1843) strain "constricta", *Curcurbitella tricuspis* (Carter, 1985), *Difflugia glans* (Penard 1902) strain "glans" and *Difflugia oblonga* (Ehrenberg 1832) strain "oblonga" (Figure 4.1).

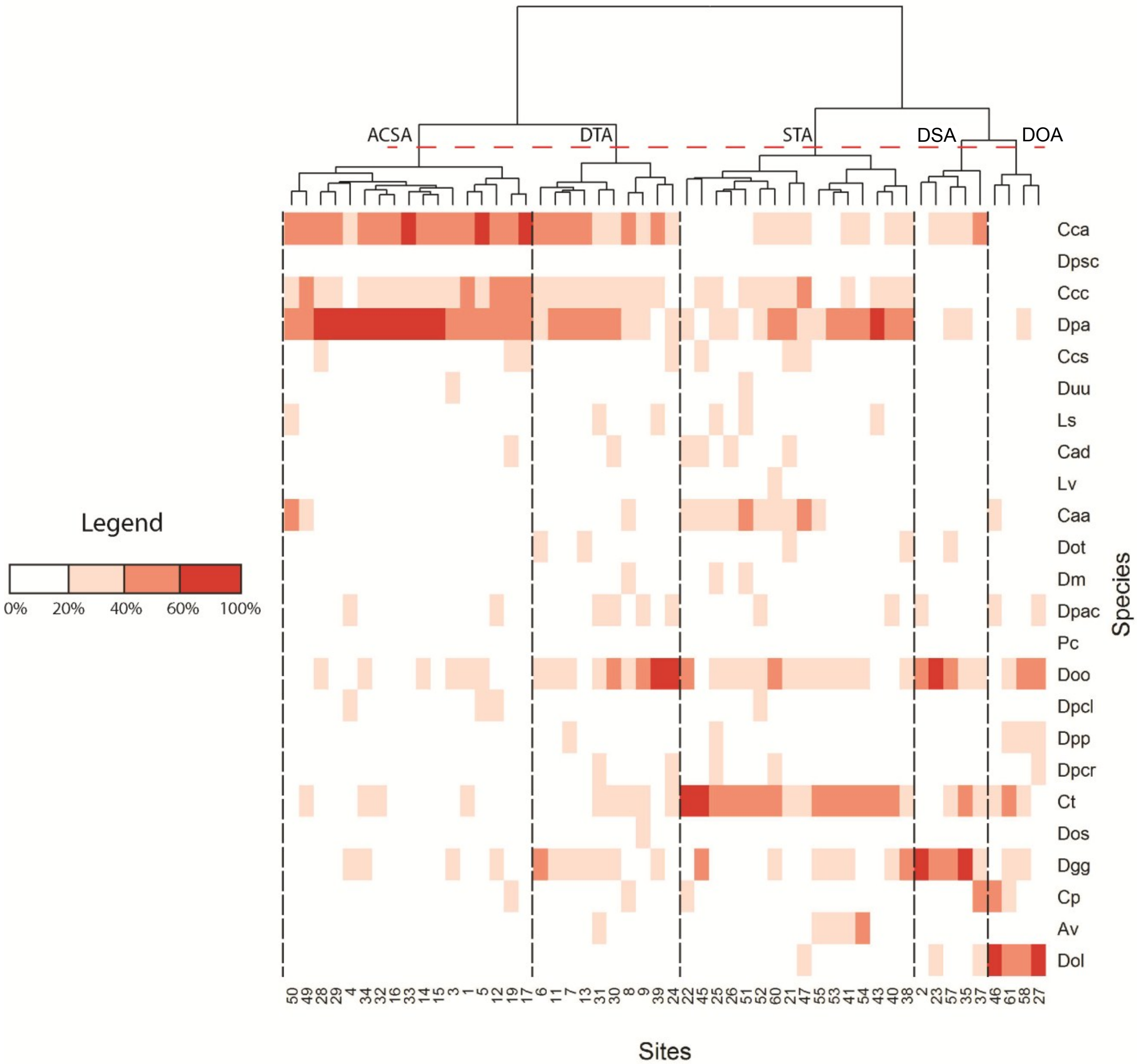


Figure 4.1 Q-mode (horizontal) vs. R-mode (vertical) cluster analysis dendrogram for the 52 samples and 24 statistically significant species and strains showing five sitinct arcellacean assemblages. The colors represented in the dendrogram reflect the relative abundances of the arcellacean species and strains.

4.2 DCA

Results produced by DCA analysis were mostly similar to that obtained by cluster analysis, delineating five distinct arcellacean assemblages (Figure 4.2). The only difference between the results of these analyses was the loadings associated with samples BC 38, 40 and 43. The samples are closely clustered near the Deep Transitional Assemblage on the DCA plot, while they are found associating with the samples of the Shallow Transitional Assemblage in cluster analysis (Figure 4.1; Figure 4.2). The reason for such sharply contrasting results will be discussed below (Section 4.4).

4.3 RDA and pRDA

The RDA analysis results are in general agreement with the results of the DCA and cluster analysis, as they show the same distinct arcellacean assemblages (Figure 4.3). Variance partition of the partial RDA results shows that the first five axes are significant at $P < 0.005$. However, only the first three RDA axes were retained based on the scree plot. Axes one (Eigenvalue= 0.09835), two (Eigenvalue= 0.033279), and three (Eigenvalue=0.01916) explain 75.1% of the variance in the arcellacean data. Moreover, variance partitioning provided confirmation that several variables influenced the faunal distribution. The results show that arsenic is the most statistically significant among the selected variables, explaining 10.7% of the variance of the observed arcellacean faunal distribution (Figure 4.4). Other significant variables include sedimentary P (8.5%), Ba (6.2%), S1 (6%), Ca (4.5%) and S3 (3.4%).The measured environmental variables, selected with the help of variance partition, explained 57% of the variance in the arcellacean data.

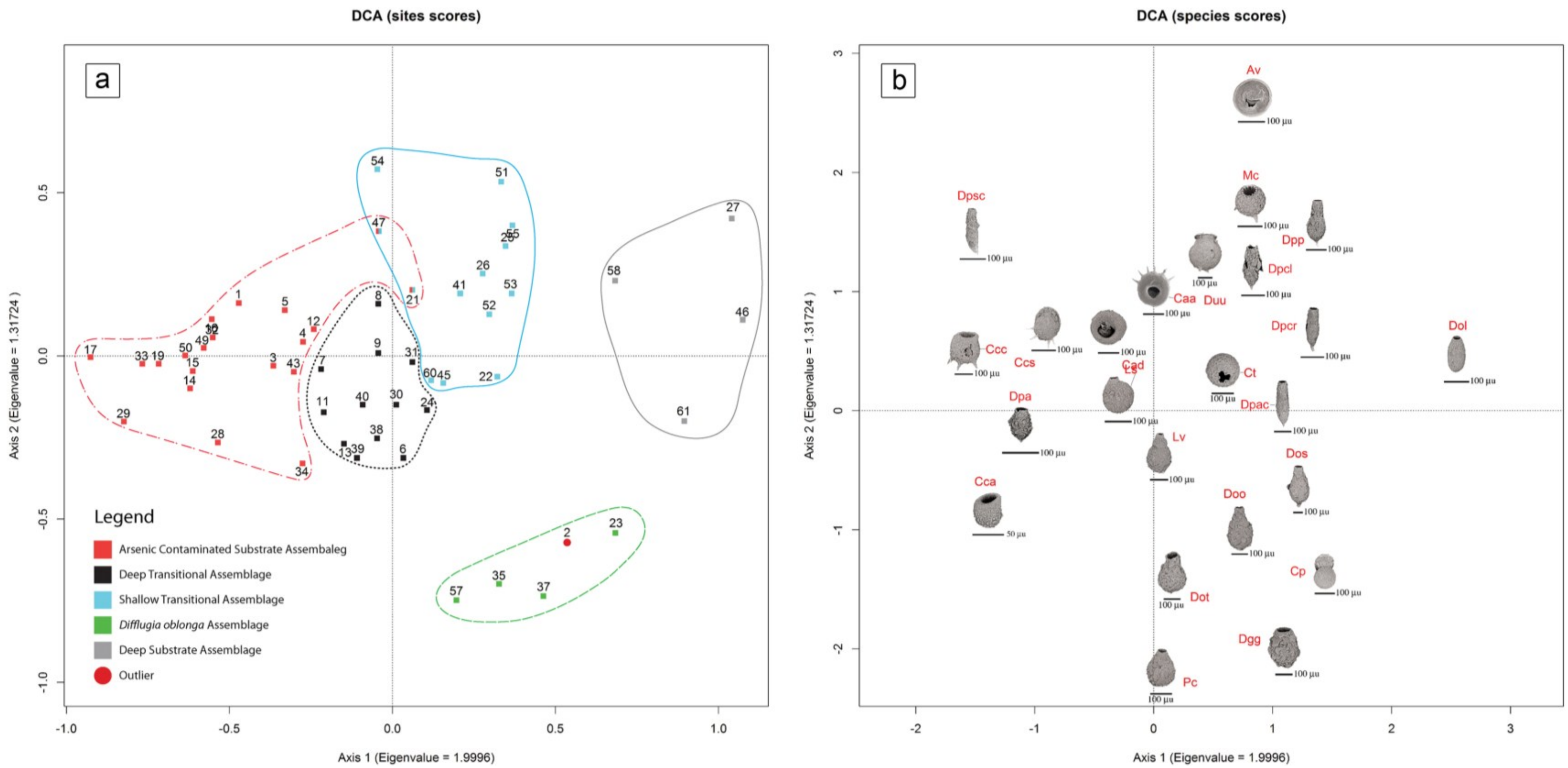


Figure 4.2 Detrended Correspondence Analysis (DCA), where (a) DCA bi-plot based on sites scores showing five distinct arcellacean assemblages, and (b) DCA bi-plot based on species scores showing the faunal distribution of arcellacean species and strains.

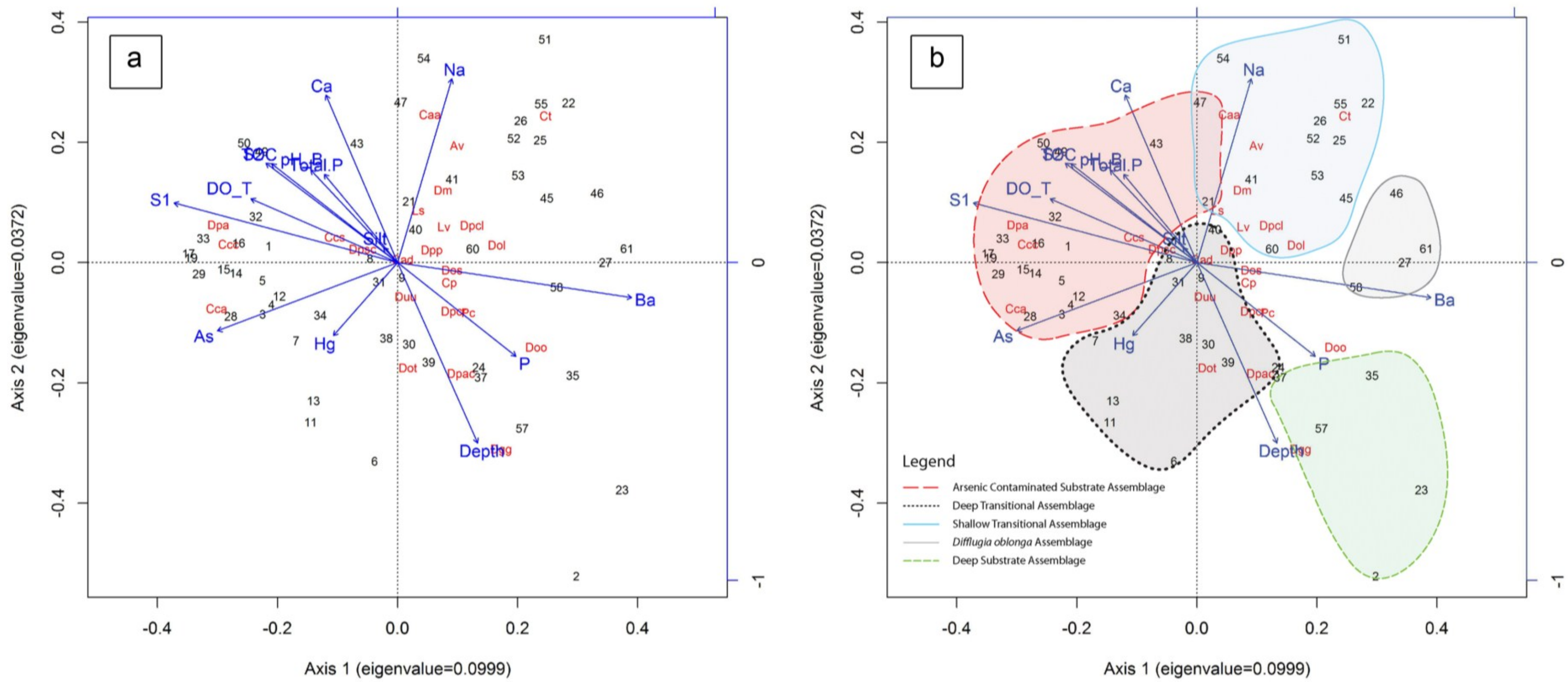
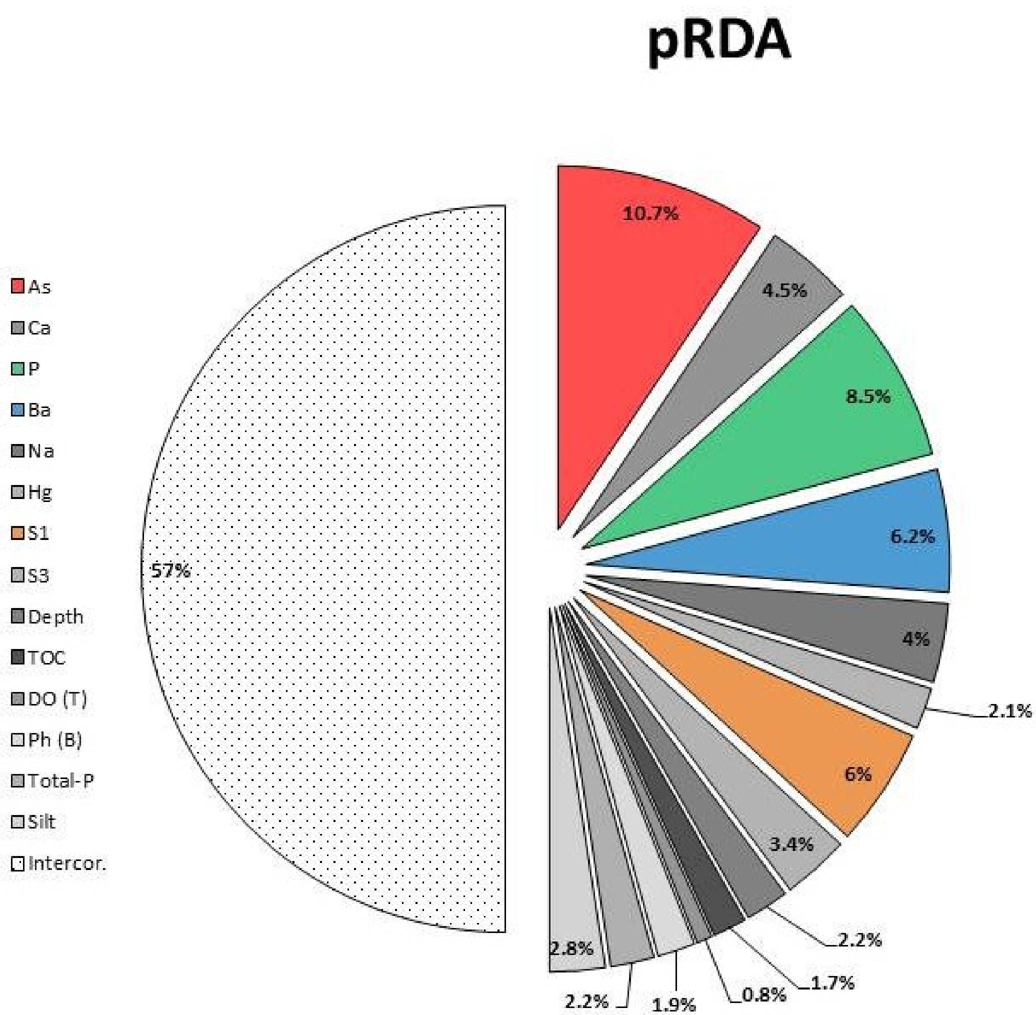


Figure 4.3 (a) Redundancy Analysis (RDA) species-environment-sample tri-plots for the 52 sediment-water-interface samples that yielded arcellaceans and no missing values. (b) RDA species-environment-sample tri-plot showing five distinctive arcellacean assemblages and their relation with the measured environmental variables.



Variable	% Variance	Pr(>F)
As	10.7	0.01
Ca	4.5	0.01
P	8.5	0.01
Ba	6.2	0.01
Na	4	0.01
Hg	2.1	0.1
S1	6	0.01
S3	3.4	0.02
Depth	2.2	0.02
TOC	1.7	0.2
DO (T)	0.8	0.6
Ph (B)	1.9	0.2
Total-P	2.2	0.1
Silt	2.8	0.03
Intercor.	57	NA

Figure 4.4 Partial Redundancy Analysis (pRDA) with variance partitioning test showing the percentage variance in the arcellacean data set that is explained by the measured environmental variables and p-value (Pr(>F)).

4.4 Arcellacean assemblages

The arcellacean assemblages recognized from cluster and DCA analysis demonstrate that the study area is a good natural laboratory to study different environmental conditions that occur in contaminated, transitional and remediated lakes.

4.4.1 Arsenic Contaminated Substrate Assemblage (ACSA)

Samples comprising the ACSA were collected mainly from lakes in transects N, W, and to a lesser extent E. The distances between the sampled lakes and the Giant Mine ranges from 2 to 20 km . These samples were found in cold ($\bar{x} = 9.6$ C°), shallow ($\bar{x} = 2.49$m) and well-oxygenated water ($\bar{x} = 10.9$ mg/l at the sediment-water interface). The sediments of the sampled lakes were dominated by silt ($\bar{x} = 73.3\%$). The calculated SDI for the majority of the samples ranges from 1.11 to 2.23, with the exception of sample BC 21 (SDI = 2.6), reflecting a transitional ecosystem (Magurran 1988; Patterson and Kumar 2000).

The DCA analysis results indicate that the ACSA samples have a high degree of similarity as they are closely grouped (Figure 3.2). However, samples BC 21 and 47, and sample BC 43 grouped differently than what is found in cluster analysis. On the DCA plot, samples BC 21 and 47 plots between the ACSA and the Shallow Transitional Assemblages (STA; Figure 4.2). These samples seem to show a similar faunal structure to that observed in the STA samples, thus explaining their coexistence with that assemblage in the Q-mode cluster results (Figure 4.1). However, these samples are also characterized by high arsenic concentrations (BC21 = 368.4 ppm and BC 47 = 339.8 ppm) that make them similar to those found in the ACSA. Due to these similarities, the two assemblages were deemed to be overlapping in these samples. Similarly, sample BC 43 is grouped with the Deep Transitional Assemblage (DTA) in cluster analysis, while grouping with samples from the ACSA assemblage in the DCA analysis results. The explanation for this apparent contradiction will be presented below in the results for the Deep Transitional Assemblage.

Geochemical analysis shows that samples of the ACSA are typically characterized by very high arsenic concentrations ($\bar{x}$ = 1017 ppm, σ = 2354 ppm), with only two samples (BC 49 and 50) having concentrations lower than 90 ppm (Table 3.3). These values are well above the interim sediment quality guidelines (ISQG; 5.9 ppm) and the probable effect levels (PEL; 17 ppm; CCMA, 2001). Results from the RDA analysis confirmed these results by showing that ACSA positively correlated with As, S1 S3, Ca, TOC, DO, pH, and Total-P (Figure 4.3) and negatively correlated with depth, P and Ba.

The faunal makeup of this assemblage was dominated by *Difflugia protaeiformis* (Lamarck 1816) strain "amorphalis" ($\bar{x} = 21.4\%$), *Centropyxis constricta* (Ehrenberg 1843) strain "aerophila" ($\bar{x} = 17.7\%$) and *Centropyxis constricta* (Ehrenberg 1843) strain "constricta" ($\bar{x} = 11.3\%$; Figure 4.1). Other species like *Difflugia oblonga* (Ehrenberg 1832) strain "oblonga" ($\bar{x} = 5.6\%$), and *Curcurbitella tricuspis* (Carter 1856) ($\bar{x} = 5.8\%$) were also common, but not in all samples.

4.4.2 Deep Transitional Assemblage (DTA)

This assemblage consisted of samples from transects N, E and W, and a sample from transect S. The distance between the Giant Mine and the sampled lakes comprising this assemblage vary from 5.5 to 19 km. The samples were restricted to cold ($\bar{x} = 9.1$ C$^{\circ}$), relatively deep ($\bar{x} = 4.7$m) and well-oxygenated water ($\bar{x} = 8.6$ mg/l at the sediment-water interface). Similar to what was observed in the ACSA, the sedimentary texture was dominated by silt ($\bar{x} = 68\%$), but with a higher percentage of sand ($\bar{x} = 18\%$). The SDI values obtained for ACSA samples (SDI = 1.64 to 2.55) fall within the transitional range. (Magurran 1988; Patterson and Kumar 2000)

As observed in the ACSA, the results of the DCA and cluster analysis differed slightly. Cluster analysis grouped samples BC 38 and 40 and 43 with the Shallow Transitional Assemblage (STA), while DCA analysis grouped BC 37 and 40 with the DTA, and BC 43 with the ACSA (Figure 4.1; Figure 4.2). The faunal makeup of these samples is most similar to STA, but the deeper water origin of the samples is more characteristic of the DTA. This explains the contrast between cluster analysis and DCA for BC 38 and 40, but not BC 43 which was grouped with the ACSA. Regardless of the similarities shared between the three samples, BC 43 was characterized by elevated arsenic and conductivity (Table 3.1; Table 3.3) making it more characteristic of the ACSA, and justifying the grouping of this sample with that assemblage.

Geochemical analysis indicated that the samples of this assemblage were also characterized by high arsenic concentrations ($\bar{x}$ =148 ppm, σ = 208 ppm), which are still higher than the acceptable levels (CCMA, 2001) but lower than observed in the ACSA. Samples of the DTA correlated well with As, Hg, silt, depth and P in the RDA analysis bi-plot (Figure 4.3). The analysis was also characterized by a negative correlation with Na, Ca, TOC, pH, S2 and DO.

As observed from the ACSA assemblage, the relatively high proportion of *D. protaeiformis* (Lamarck 1816) strain "amorphalis" ($\bar{x} = 12.2\%$), *C. constricta* (Ehrenberg

1843) strain “aerophila” ($\bar{x} = 11.5\%$) and *C. constricta* (Ehrenberg 1843) strain “constricta” ($\bar{x} = 8.5\%$) (Figure 4.1) were also present in the DTA but were less dominant. Notable additional components of the DT assemblage included *D. oblonga* (Ehrenberg 1832) strain “oblonga” ($\bar{x} = 10.4\%$), and most notably *Difflugia glans* (Penard 1902) strain “glans” ($\bar{x} = 7.71\%$). *C. tricuspis* (Carter 1856) was also a common contributor to the assemblage ($\bar{x} = 5.7\%$).

4.4.3 Shallow Transitional Assemblage (STA)

Samples corresponding with the STA were collected from all transects. The closest sampled lake to the Giant Mine is B 21 (5.2 km from the mine), while the furthest is BC 51 (31.5 km from the mine). The STA assemblage characterizes cold ($\bar{x} = 9.9$ C$^{\circ}$), very shallow ($\bar{x} = 2$ m) and well-oxygenated water ($\bar{x} = 10.6$ mg/l at the sediment-water interface). As with the previously discussed assemblages, the fauna in this assemblage were found living in substrates characterized by a silt-dominated sedimentary texture ($\bar{x} = 70.2\%$). The observed SDI values for the STA assemblage range from 1.93 to 2.47, indicating an ecosystem that is transitioning to healthy environmental conditions (Magurran 1988; Patterson and Kumar 2000).

The results of the DCA analysis were similar to that obtained by cluster analysis, except for the overlapping samples BC 21 and 47 (see section 4.4.1). The DCA plot show that samples within the ST assemblage are relatively closely correlated (Figure 4.2). With the exception of the overlapping samples BC 21 and 47, the samples of this assemblage are characterized by the lowest arsenic concentrations ($\bar{x}$ =26 ppm, σ = 123 ppm). However, these concentrations remain above the acceptable guidelines for the protection of aquatic life **(**CCMA, 2001**).** Examination of the RDA analysis results indicate that this assemblage best correlates with Na, and more weakly correlates with As, Hg, and silt (Figure 4.3).

This assemblage is similar to the ACSA and DTA in that it was dominated by *D. protaeiformis* (Lamarck 1816) strain "amorphalis" ($\bar{x} = 11.6\%$). This assemblage, however, is co-dominated *C. tricuspis* (Carter 1856) ($\bar{x} = 14.3\%$). The centropyxids, especially *C. constricta* (Ehrenberg 1843) strain "aerophila" ($\bar{x} = 6\%$)*, C. constricta* (Ehrenberg 1843) strain *"*constricta" ($\bar{x}$ =7.8%) and additionally *C. aculeata* (Ehrenberg 1832) strain "aculeata" are an important assemblage component, albeit present in moderate abundances*. D. oblonga* (Ehrenberg 1832) strain "oblonga" ($\bar{x} = 8.6\%$) was also present but not in all samples.

4.4.4 *Difflugia oblonga* Assemblage (DOA)

Samples comprising the DOA included a sample from transect W, another from transect E and two samples from transect S. The distances between the sampled lakes and the Giant Mine ranges from 2.3 to 20 km. The samples were found in cold ($\bar{x}$ =10.9 C$^{\circ}$), relatively deep ($\bar{x} = 4$m) and well-oxygenated water ($\bar{x} = 9.98$ mg/l at the sediment-water interface). The sediments comprising these samples was dominated by silt ($\bar{x} = 63\%$), but also by an elevated percentage of clay ($\bar{x} = 25\%$). The calculated SDI for the assemblage reflects transitional ecosystems, as it ranges from 1.88 to 2.27 (Magurran 1988; Patterson and Kumar 2000).

The results of DCA indicates that the samples of the DOA are closely correlated, thus corroborating the results of the cluster analysis (Figure 4.2). The average arsenic concentration in the DOA is similar to that of the STA ($\bar{x}$ =88.9 ppm, σ = 72 ppm) and is still well above the recommended Canadian acceptable guidelines (CCMA, 2001). Results of the RDA analysis indicate that samples comprising this assemblage correlate positively with Ba, and negatively with S1, As, Hg and silt (Figure 4.3).

As the name suggests, the DOA is dominated by Difflugid species and strains, especially *D. oblonga* (Ehrenberg 1832) strain "lanceolata" ($\bar{x} = 14.5\%$), and *C. tricuspis* (Carter 1856) ($\bar{x} = 14.3\%$). Other common species and strains include *D. oblonga* (Ehrenberg 1832) strain "oblonga" ($\bar{x} = 9.6\%$), and *D. protaeiformis* (Lamarck 1816) strain "protaeiformis" ($\bar{x} = 7.5\%$), and *D. glans* (Penard 1902) strain "glans" ($\bar{x} = 5.8\%$), and *D. protaeiformis* (Lamarck 1816) strain "amorphalis" ($\bar{x} = 6.8\%$). Notably, the presence of centropyxids is reduced in this assemblage, with only *C. aculeata* (Ehrenberg 1832) strain " aculeata" ($\bar{x} = 5.7\%$) and *C. constricta* (Ehrenberg 1843) strain " constrict" ($\bar{x} = 5.6\%$) present in moderate proportions.

4.4.5 Deep Substrate Assemblage (DSA)

Samples comprising the DSA Assemblage were collected from lakes found in all four transects. The samples were among the furthest from the site of the Giant Mine, with a distance range from 17 to 22 km. The sampled lakes had cold ($\bar{x} = 11.4$ C$^{\circ}$), deep ($\bar{x} = 6.8$m) and oxygenated water ($\bar{x}$ =6.5 mg/l). The samples of this assemblage were also characterized by a silt-dominated sedimentary texture ($\bar{x} = 79.4\%$). The calculated SDI for the assemblage ranges from 1.69 to 1.95, indicating a transitional ecosystem (Magurran 1988; Patterson and Kumar 2000).

On the DCA plot, the samples of the DSA were closely grouped away from the other assemblages (Figure 4.2). RDA analysis show that samples from this assemblage positively correlate with water depth and sedimentary P (Figure 4.3) while negatively correlating with S1, DO, TOC, pH, Ca, Total-P (Figure 4.3). DSA has the second lowest average concentration of arsenic ($\bar{x}$ =32.6 ppm, σ = 27.2 ppm). One outlier within the DSA, BC 2, was characterized by a very high arsenic concentration ($\bar{x} =$ 905.2%). This sample clusters with this assemblage because it has a similar faunal structure that is also dominated by *D. glans* (Penard 1902) strain "glans" ($\bar{x} =$ 69%).

This assemblage is dominated by *D. glans* (Penard 1902) strain "glans" ($\bar{x} =$ 18.9%), *D. oblonga* (Ehrenberg 1832) strain "oblonga" ($\bar{x} =$ 16.8%), and *C. tricuspis* (Carter 1856) ($\bar{x} =$ 10.1%). *C. constricta* (Ehrenberg 1843) strain "constricta" ($\bar{x}$ =27.5%), *D. protaeiformis* (Lamarck 1816) strain "amorphalis" ($\bar{x}$ =15.6%), and *D. protaeiformis* (Lamarck 1816) strain "acuminata" ($\bar{x}$ =14%) were also common, but in all the samples.

5. Discussion

5.1 Arsenic Contaminated Substrate Assemblage (ACSA)

The ACSA is a prime example of an assemblage typically found in shallow, heavily contaminated substrates. It is relatively similar to the Contaminated Substrate Assemblage – CS of Patterson et al., (1996) and the Shallow Contaminated Substrate Assemblage (5) of Reinhardt et al., (1998). This correlation is supported by the strong positive correlation between arsenic and the ACSA samples in the RDA analysis tri-plot (Figure 4.3). Furthermore, the geochemical analysis revealed that samples of this assemblage are characterized by very high, albeit variables, concentrations of arsenic ($\bar{x}$ = 1017 ppm, σ = 2354 ppm). The results therefore suggest a strong influence of arsenic on the arcellacean distribution in this assemblage.

The faunal structure of this assemblage is dominated by *D. protaeiformis* strain "amorphalis" ($\bar{x} = 21.4\%$). In northern Italy and sites in Ontario, various strains of *D. protaeiformis* were recognized to be good indicators of contaminated substrates (Asioli et al., 1996; Patterson et al., 1996; Reinhardt et al., 1998). Reinhardt et al. (1998) linked the relatively high abundance of *D. protaeiformis* strain "amorphalis" in their Diatom Mud Assemblage (1) with residual contamination that penetrated the surface of the sampled lakes. The abundance of *D. protaeiformis* strain "amorphalis" in the ACSA seems to reflect similar conditions. Processing of ore during the early years of the Giant Mine's operation (1948 to 1958) resulted in very high atmospheric emission of As_2O_3 (thousands of kg per day), which subsequently decreased significantly in later years as more stringent environmental laws came into effect (INAC, 2007). The atmospheric emission of arsenic trioxide finally ended with the cessation of the Giant Mine's activities in 2004. Therefore, we assume that the measured arsenic concentrations are, in fact, residuals from the historical release of As_2O_3 and, possibly, other sources (e.g. bedrock geology ; Webster, 1999; Ollson, 1999).

Centropyxids, like *C. constricta* strain "aerophila" ($\bar{x} = 17.7\%$) and *C. constricta* strain "constricta" ($\bar{x} = 11.3\%$), were also present in high proportions. These opportunistic species are capable of withstanding harsh environmental conditions, including cold temperature (Decloire, 1956), low salinity conditions (<5‰; Decloîre, 1956; Scott and Medioli, 1980; Patterson et al., 1985; Hoing and Scott, 1987), oligotrophic conditions (Schönborn, 1984) and sites heavily contaminated by arsenic and mercury (Patterson et al., 1996; Reinhardt et al., 1998). Therefore, the abundance of centropyxids in the ACSA is an indication of the environmental stress caused by the excessive presence of arsenic in these samples. However, the calculated SDI values of the ACSA (SDI = 1.11 – 2.23) seems to reflect signs of remediation as it fall between stressed and transitional ecosystems (Magurran 1988; Patterson and Kumar 2000). A similar contrast between the geochemical analysis and arcellacean diversity was reported in a previous study and was considered as a sign of ongoing remediation in stressed ecosystems (Reinhardt et al., 1998). In addition, the moderate proportions of *D. oblonga* strain "oblonga" ($\bar{x} = 5.6\%$), and *C. tricuspis* ($\bar{x} = 5.8\%$) in these samples suggest that a slow, but steady, and ongoing remediation process going on in lakes of the ACSA, as these species are usually linked with eutrophic conditions or substrates comprised of elevated organic matter (Collins et al., 1990).

5.2 Deep Transitional Assemblage (DTA)

The DTA seems to be a transitional assemblage between the ACSA and the Shallow Transitional Assemblage (STA), and is similar to the type of assemblage found in relatively deep, contaminated substrates that are transitioning toward more hospitable conditions. The transition is reflected by the measured SDI for this assemblages (SDI = 1.64 – 2.55), which falls in the range of transitional to relatively healthy ecosystems (Magurran 1988; Patterson and Kumar 2000). The RDA analysis (Figure 4.3) results confirmed the lasting, albeit reduced, influence of arsenic on the distribution of arcellacean in DTA samples ($\bar{x}$ = 148 ppm, σ = 208 ppm).

The transition is also observed from the relative abundance of the dominant species in the DTA. Similar to the ACSA, *D. protaeiformis* (Lamarck 1816) strain "amorphalis" ($\bar{x} = 12.2\%$), *C. constricta* strain "aerophila" ($\bar{x} = 11.5\%$) and *C. constricta* strain "constricta" ($\bar{x} = 38\%$) dominate the faunal makeup of the DTA, but are present in lower proportions (Figure 4.1). While many of the arguments presented for the ASCA hold true for the DTA, the reduced abundance of the dominant species seem to suggest more hospitable conditions in the DTA. Additionally, the increase in the abundance of *D. oblonga* strain "oblonga" ($\bar{x} = 10.4\%$) corroborates the assumption of reduced environmental stress. *D. oblonga* was reported to thrive from tropical to Arctic conditions in virtually any lake environments as long as the sediments are sufficiently organic rich (Collins et al, 1990). Although the average TOC ($\bar{x}$ = 18.9%, σ = 8.7) is relatively low in this assemblage, the high diversity of arcellaceans in this assemblage (SDI = 1.64 – 2.55) suggests the presence of sufficient amounts of organics to sustain a considerable diversity of arcellacean species.

Notably, the DTA is also characterized by elevated proportions of *D. glans* (Penard 1902) strain "glans" ($\bar{x} = 7.7\%$). Reinhardt et al. (1998) reported high proportions of this species in deep, contaminated substrates as well. Similarly in the DTA, *D. glans* (Penard 1902) strain "glans" was found to colonize relatively deep ($\bar{x}$ = 4.76m) and contaminated ($\bar{x}$ = 148 ppm, σ = 208 ppm) substrates. The RDA analysis further supports this assessment and indicates a very close correlation between depth and *D. glans* (Penard 1902) strain "glans" (Figure 4.3).

5.3 Shallow Transitional Assemblage (STA)

The environmental conditions indicated by the STA are similar to those found in lakes exhibiting mesotrophic environmental conditions, and is very similar to the High Diversity Assemblage (2) of Reinhardt et al. (1998). This assessment is supported by the high arcellacean diversity in this assemblage (SDI = 1.93 – 2.47), which is expected for relatively healthy ecosystems. The RDA bi-plot suggests a weak correlation between arsenic and the STA samples (Figure 4.3). The geochemical analysis indicates that STA samples are characterized by the lowest arsenic concentrations ($\bar{x}$ = 26.5 ppm, σ = 13.5 ppm) when excluding the concentrations of samples BC 21 and 47. These low arsenic concentrations are not surprising since the samples were collected from lakes located at considerable distance from the Giant Mine (Figure 2.1). This fact might lead to the assumption that arsenic concentrations were low to begin with in these sampled lakes. However, the significant presence of the opportunistic centropyxids suggests that these lakes are still recovering from continued environmental stress (Patterson et al., 1996).

D. protaeiformis strain “amorphalis” ($\bar{x} = 11.6\%$), C. aculeata strain “aculeata” ($\bar{x} = 7.8\%$), *C. constricta* strain “aerophila” ($\bar{x} = 6\%$) and *C. constricta* strain “constricta” ($\bar{x} = 5.5\%$) are present in the STA, but in decreasing order of frequency. The significant reduction in the abundances of the opportunistic centropyxids and *D. protaeiformis* “amorphalis” in this assemblage corresponds well with the drastic reduction of environmental stress.

A notable distinction between the faunal structure of the STA and that of the previously discussed assemblages is the dominance of *C. tricuspis* ($\bar{x} = 14.3\%$). Elevated abundances of *C. tricuspis* were reported to inhabit water bodies characterized by the presence of a bright green algal mat comprised of *Spirogyra* (Schönborn, 1984; Patterson, 1985; Medioli et al., 1987; Collins et al., 1990). Medioli et al., (1987) determined a parasitic relationship between *C. tricuspis* and *Spirogyra*, upon which the first preferentially feeds on the latter. In our study, the high proportion of *C. tricuspis* could not be attributed to this parasitic relation because *Spirogyra* was not studied in lakes sampled for the STA. However, high abundances of *C. tricuspis* in the absence of *Spirogyra spp*. have also been reported from highly eutrophic lakes in New Brunswick and Nova Scotia (Patterson et al., 1985; Scott and Medioli, 1983). While average TOC measured for the samples of this assemblage ($\bar{x} = 21\%$, $\sigma = 10.4\%$) does not reflect eutrophic conditions, the high arcellacean diversity in STA suggests the presence a source of organics sufficiently high to sustain this assemblage. The high organic content of these samples is also reflected in the presence of a relatively high proportion of *D. oblonga* “oblonga” ($\bar{x} = 8.6\%$) which indicates the presence of sufficient organic matter in the sampled lakes (Collins et al., 1990).

5.4 *Difflugia oblonga* Assemblage (DOA)

The moderately diverse DOA is characteristic of transitional lakes that are showing signs of remediation. This assessment is supported by the calculated SDI values (SDI = 1.88 – 2.27), which reflects transitional ecosystems, and the moderate arsenic concentrations ($\bar{x}$ = 80 ppm, σ = 72 ppm) characteristic of DOA lakes. Both DCA and RDA results suggest that the DOA is subject to more than one source of environmental stress. The RDA analysis reveals a strong correlation between DOA samples and barium on RDA axis 1 and, as would be expected, a weak correlation between the assemblage and arsenic (Figure 4.3).

Barium is an alkaline-earth metal that is found in over 80 minerals, principally barite (barium sulfate) and witherite (barium carbonate; CCMA, 2001). Soluble, and some insoluble, barium salts and compound are deemed as highly toxic (CCMA, 2001). The samples of the DOA are indeed characterized by significantly higher barium concentration ($\bar{x}$ = 193 ppm, σ = 18 ppm) when compared to the other assemblages recognized in this study. Therefore, the presence of elevated concentrations of barium may be considered as another source of stress in addition to arsenic in the DOA.

The faunal makeup of this assemblage provides additional evidence of transitional environmental conditions. The dominant species in the DOA, *D. oblonga* "lanceolata" ($\bar{x} = 14.5\%$), *D. oblonga* "oblonga" ($\bar{x} = 9.6\%$), and *C. tricuspis* ($\bar{x} = 10.9\%$), suggest a recovery to more normal conditions in the sampled lakes. However, the presence of *C. aculeata* strain "aculeata" ($\bar{x} = 5.7\%$), *C. constricta* strain "constricta" ($\bar{x} = 5.6\%$), and *D. protaeiformis* strain "protaeiformis" ($\bar{x} = 7.5\%$) indicates continued environmental stress. Recently, difflugids vs. centropyxids ratio was used as a mean to evaluate ecosystem health of constructed wetlands in Alberta, Canada (Neville et al., 2011). This metric approach proved to be both robust and simple, as it only required genus-level identification. Healthy ecosystems were characterized by an abundance of difflugids, while centropyxids were found in stressed environments. The difflugids vs. centropyxids ratio in the DOA clearly suggest that although stress is present, remediation in the sampled lakes is well underway due to the elevated proportion of the difflugids compared to the centropyxids.

5.5 Deep Substrate Assemblage (DSA)

The moderately diverse (SDI = 1.8 – 2.2) DSA is characteristic of transitional systems that are found in depth-stressed substrates and is relatively similar to the Deep Water Contaminated Assemblage (3) of Reinhardt et al., (1998). The RDA analysis results show that samples of this assemblage are correlating positively with depth, and negatively with arsenic (Figure 4.3). This result suggests that environmental stress, as reflected by the DSA, is primarily cause by depth rather than arsenic. However, arsenic can still be considered as a secondary source of stress since the generally low concentrations ($\bar{x}$ = 32 ppm, σ = 27 ppm) in the DSA falls above the ISQG (5.9 ppm) and the PEL (17 ppm) limits (CCMA, 2001).

The influence of depth on this assemblage is most evident on the faunal distribution, which is characterized by a significant increase in the abundance of *D. glans* strain “glans” ($\bar{x} = 18.9\%$). As observed in the DTA, elevated presence of *D. glans* strain “glans” was linked with deep, contaminated substrates. In the case of the DSA, however, arsenic contamination is much milder than that found in the DTA. Nonetheless, *D. glans*

(Penard 1902) strain “glans” did not seem to be impacted by this significant reduction of arsenic contamination. This suggests that depth rather than the level of arsenic contamination is controlling the distribution of *D. glans* (Penard 1902) strain “glans”.

In addition, the presence of significant abundance of *D. oblonga* strain “oblonga” ($\bar{x} = 16.8\%$) and *C. tricuspis* ($\bar{x} = 10.1\%$), moderate proportions of *C. constricta* strain “constricta” ($\bar{x} = 10\%$), *D. protaeiformis* strain “amorphalis” ($\bar{x} = 5.6\%$), and *D. protaeiformis* “acuminata” ($\bar{x} = 5\%$) provides confirmation that these ecosystems are incrementally transitioning back to more hospitable conditions while experiencing some continued environmental stress.

6. Conclusions

This study has provided new insight into the sensitivity of the distribution of lake-arcellaceans to arsenic, based on sixty-one sediment-water-interface samples collected from the region around the Giant Mine. Five distinct arcellacean assemblages, recognized by Q-mode cluster analysis, have been linked with a number of driving variables, including arsenic, barium and depth. The majority of the sampled lakes show evidence of environmental stress due to arsenic concentrations that are elevated above the acceptable Canadian guidelines, but many show evidence of remediation.

Ordination (RDA) analysis has confirmed that 14 measured environmental variable explain 57% of the variance in the arcellacean distribution. Partial RDA analysis has further confirmed that arsenic has the largest influence on the assemblage variance, explaining 10.7% ($p < 0.01$) of the total variance. This result underlies the sensitivity of arcellaceans to environmental stress caused by arsenic. Other factors influencing faunal distribution are sedimentary P, which explains 8.5% ($p<0.01$) of the total variance, Ba 6.2% ($p<0.01$), S1 6% ($P<0.01$), Ca 4.5% ($p<0.01$).

The preliminary data presented in this study suggest that arcellaceans hold considerable potential as indicators for arsenic and heavy metal contamination and remediation progress in sites surrounding mining activities. These results will be of use to policy makers and planners when evaluating the merit of various remediation programs (e.g. The Giant Mine Remediation Project) and will provide valuable insight into the impact of Giant Mine on the BCW region.

7. Future directions

7.1 Enhancing sampling resolution

This step can be achieved simply by collecting more sediment-water-interface samples from lakes that were not sampled in the current Four-transects. Such enhancement in the spatial resolution of the study will facilitate the following:

- A better and more accurate spatial mapping of arsenic distribution in the region using different interpolative methods such as Inverse Distance Weighing (IDW) and/or Kriging via geospatial software like ArcMap
- The spatial mapping of the distribution of arsenic will aid in identifying possible arsenic gradients in the region surrounding the Giant Mine, which will help in identifying sites characterized by elevated arsenic concentration in the region.

7.2 Freeze cores

Arcellaceans will be potentially examined in freeze cores collected from the same region. The cores will be subsampled at a very high temporal resolution (1mm), using a purpose-made sledge microtome (Macumber et al., 2011), to determine the following:

- Pre-mining baseline environmental conditions
- Response of lakes to mining impact
- Gauging the success of post-mining remediation

7.3 Transfer function

The significant taxa-environment result for arsenic indicated in the RDA and pRDA warrant the development of a transfer function for arsenic based on the current dataset. Such transfer function will produce calibrated quantitative estimates of the past arsenic concentrations in the region in the vicinity of the Giant Mine.

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Arcellacean SEM Plates

PLATE 1

1. *Arcella vulgaris* (Ehrenberg 1830). Apertural view of a specimen from sample BC 54.

2-3. *Centropyxis aculeata* (Ehrenberg 1832) stain “aculeata”. Apertural (2) and back (3) views for specimens from sample BC 44.

4-5. *Centropyxis aculeata* (Ehrenberg 1832) stain “discoides”. Apertural views for specimens from samples BC 9 and BC 30.

6-7. *Centropyxis constricta* (Ehrenberg 1843) stain “aerophila”. Side (6) and apertural (7) views for specimens from samples BC 8 and BC 17.

8-10. *Centropyxis constricta* (Ehrenberg 1843) stain “constricta”. Side (8) and apertural (9 and 10) views of specimens from samples BC 30 and BC 3. 11 from sample BC 9.

11. *Centropyxis constricta* (Ehrenberg 1843) stain “spinosa”. Specimen from sample BC 11.

12-15 *Centropyxis pontigulasiformis* (Beyens, Chardez and De Bock 1986). Back and side views of specimens from samples BC 25 and BC 46. 14 from sample BC 46. 15 shows the details of the visor surrounding the aperture.

16-18. *Cucurbitella tricuspis* (Carter 1956). Side (16) and apertural (17) views of specimens from samples BC 8 and 51. 18 shows the characteristic lobes of the aperture.

PLATE 1

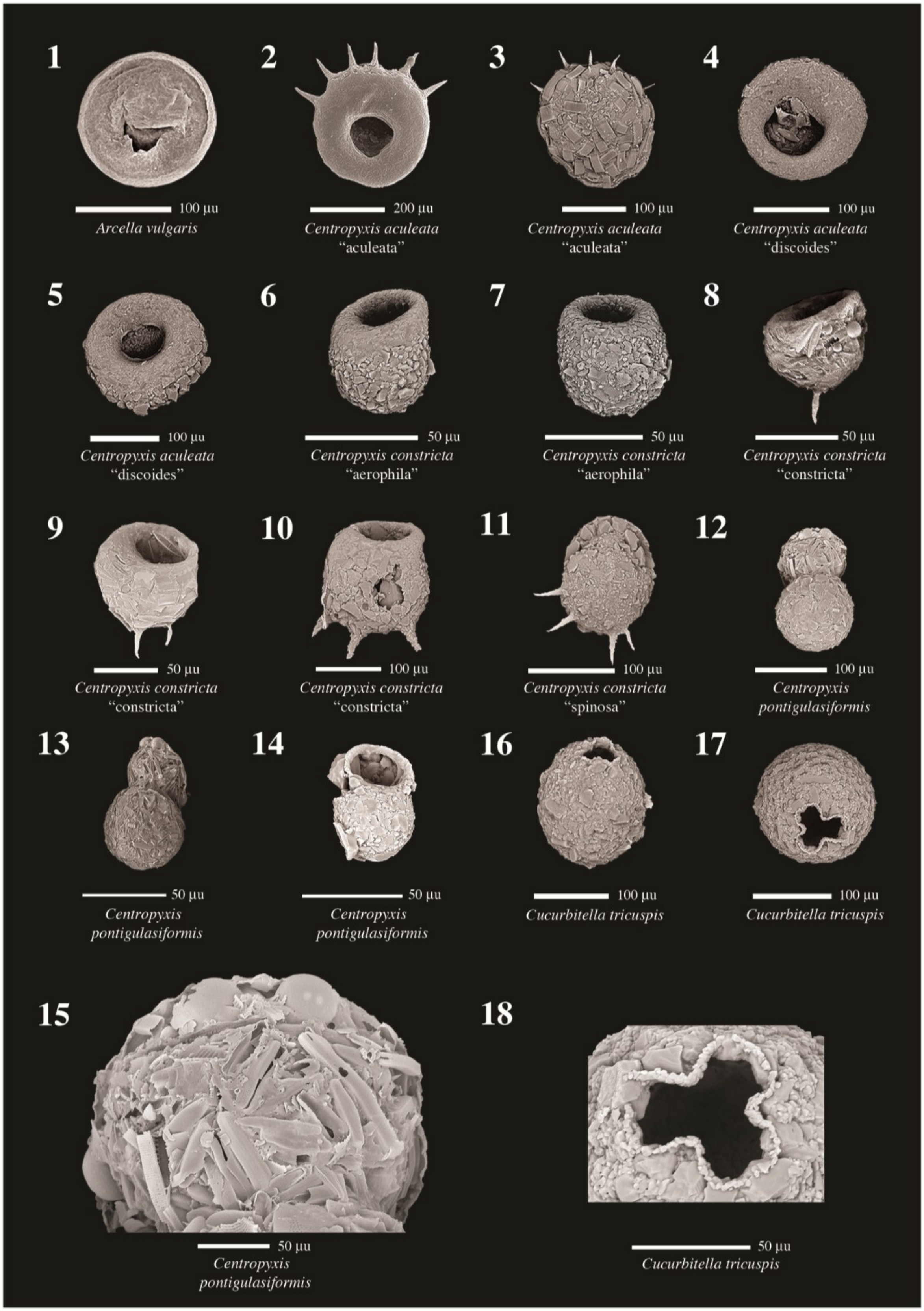

1
100 µu
Arcella vulgaris
2
200 µu
Centropyxis aculeata
"aculeata"
3
100 µu
Centropyxis aculeata
"aculeata"
4
100 µu
Centropyxis aculeata
"discoides"
5
100 µu
Centropyxis aculeata
"discoides"
6
50 µu
Centropyxis constricta
"aerophila"
7
50 µu
Centropyxis constricta
"aerophila"
8
50 µu
Centropyxis constricta
"constricta"
9
50 µu
Centropyxis constricta
"constricta"
10
100 µu
Centropyxis constricta
"constricta"
11
100 µu
Centropyxis constricta
"spinosa"
12
100 µu
Centropyxis
pontigulasiformis
13
50 µu
Centropyxis
pontigulasiformis
14
50 µu
Centropyxis
pontigulasiformis
16
100 µu
Cucurbitella tricuspis
17
100 µu
Cucurbitella tricuspis
15
50 µu
Centropyxis
pontigulasiformis
18
50 µu
Cucurbitella tricuspis

PLATE 2

1. *Mediolus corona* (Patterson, and Wallich 1986). Apertural view of a specimen from sample BC 51.

2. *Heleopera sphagni* (Leidy 1874). Side view for a specimen from sample BC 9.

3. *Lesquereusia spiralis* (Ehrenberg 1840). Side view for a specimen from sample BC52.

4. *Pontigulasia compressa* (Carter 1864). Side view for a specimen from sample BC 30.

5. *Lagenodifflugia vas* (Leidy 1874). Side view of a specimen from sample BC 9.

6. *Difflugia bidens* (Penard 1902). Specimen from sample BC 6.

7. *Difflugia protaeiformis* (Lamarck 1816) strain “amphoralis”. Side view of a specimen from sample BC 52.

8. *Difflugia urens* (Patterson, MacKinnon, Scott, and Medioli 1985). Side view of a specimen from sample BC 27.

9. *Difflugia urceolata* (Carter 1864) strain “urceolata”. Side view of a specimen from sample BC 24.

10. *Difflugia glans* (Penard 1902) strain “glans”. Side view of a specimen from sample BC 52.

11. *Difflugia glans* (Penard 1902) strain “distenda”. Side view of a specimen from sample BC 46.

12. *Difflugia oblonga* (Ehrenberg 1832) strain “oblonga”. Side view of a specimen from sample BC 23.

13. *Difflugia oblonga* (Ehrenberg 1832) strain “’spinosa”. Side view of a specimen from sample BC 35.

PLATE 2

14. *Difflugia oblonga* (Ehrenberg 1832) strain "tenuis". Side view of a specimen from sample BC 39.

15. *Difflugia oblonga* (Ehrenberg, 1832) strain "lanceolata". Side view of a specimen from samples BC 46.

16. *Difflugia protaeiformis* (Lamarck 1816) strain "protaeiformis". Side view of a specimen from samples BC 46.

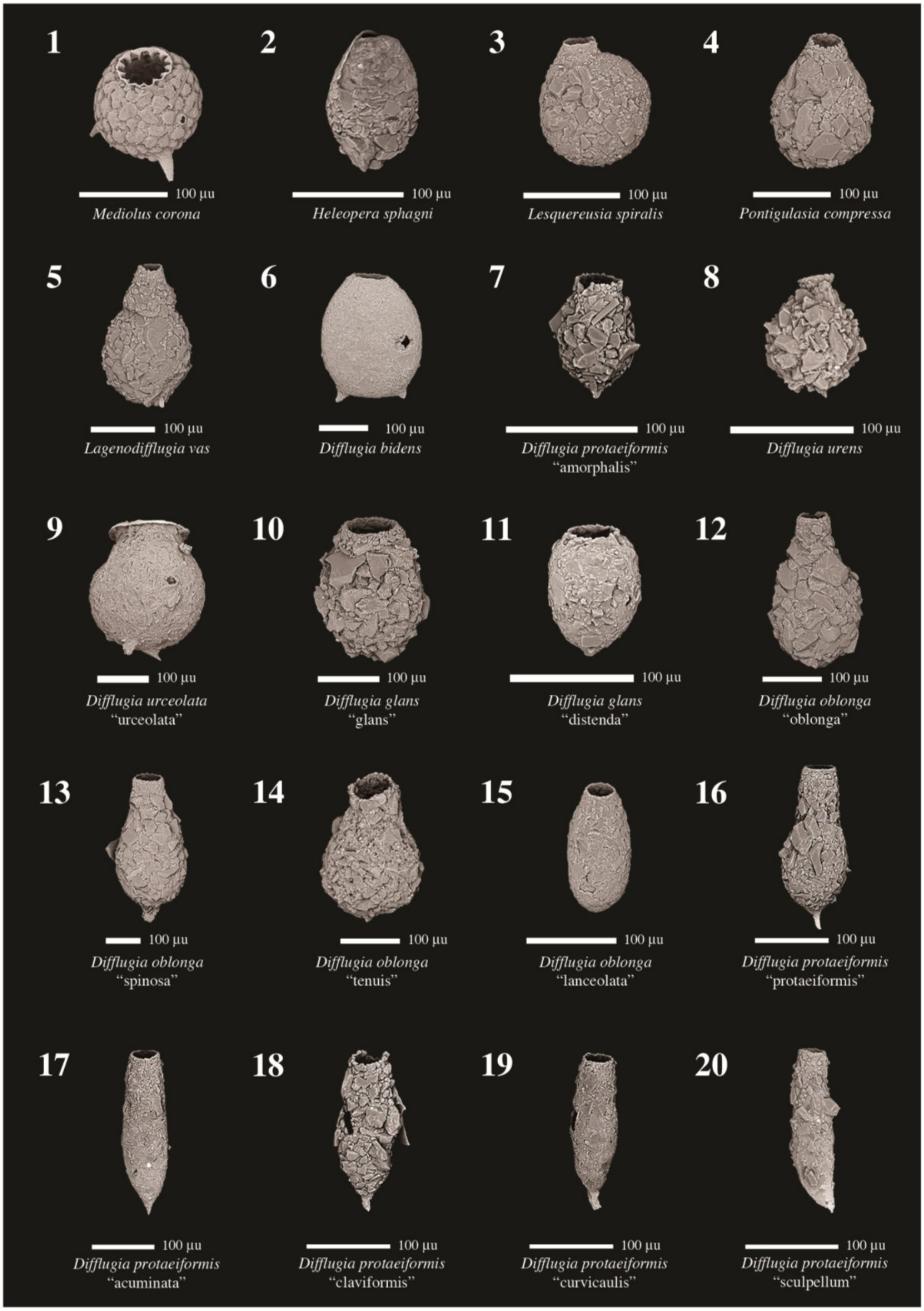
17. *Difflugia protaeiformis* (Lamarck 1816) strain "acuminata". Side view of a specimen from samples BC 38.

18. *Difflugia protaeiformis* (Lamarck 1816) strain "claviformis". Side view of a specimen from samples BC 52.

19. *Difflugia protaeiformis* (Lamarck 1816) strain "curvicaulis". Side view of a specimen from samples BC 9.

20. *Difflugia protaeiformis* (Lamarck 1816) strain "sculpellum". Side view of a specimen from samples BC 9.

PLATE 2


1
100 µu
Mediolus corona
2
100 µu
Heleopera sphagni
3
100 µu
Lesquereusia spiralis
4
100 µu
Pontigulasia compressa
5
100 µu
Lagenodifflugia vas
6
100 µu
Difflugia bidens
7
100 µu
Difflugia protaeiformis
"amorphalis"
8
100 µu
Difflugia urens
9
100 µu
Difflugia urceolata
"urceolata"
10
100 µu
Difflugia glans
"glans"
11
100 µu
Difflugia glans
"distenda"
12
100 µu
Difflugia oblonga
"oblonga"
13
100 µu
Difflugia oblonga
"spinosa"
14
100 µu
Difflugia oblonga
"tenuis"
15
100 µu
Difflugia oblonga
"lanceolata"
16
100 µu
Difflugia protaeiformis
"protaeiformis"
17
100 µu
Difflugia protaeiformis
"acuminata"
18
100 µu
Difflugia protaeiformis
"claviformis"
19
100 µu
Difflugia protaeiformis
"curvicaulis"
20
100 µu
Difflugia protaeiformis
"sculpellum"

Appendices

Appendix A: Multivariate ordination code in R

The code can be pasted and used in R with no complications. Any italic part of the code is to be appropriately changed by the user,

#Importing data (CSV file)
spe.raw<-read.csv("*spe*.csv", row.names=1)
Env.raw<-read.csv("*Env.raw*.csv", row.names=1)

#Exploratory data analysis
##Uploading libraries
Library (vegan)

##Environmental data
Summary (Env.raw) #Descriptive statistics for the data (e.g. mean, median, min, max…etc.)

###Barplots
barplot(Env.raw$*Mo*,main="Mo")
barplot(Env.raw $*Cu*,main="Cu")
barplot(Env.raw $*Pb*,main="Pb")
barplot(Env.raw $*Zn*,main="Zn")

###Boxplots
boxplot(Env.raw$*Mo*,main="*Mo*")
boxplot(Env.raw $*Cu*,main="*Cu*")
boxplot(Env.raw $*Pb*,main="*Pb*")
boxplot(Env.raw $*Zn*,main="*Zn*")

###Barplots and Boxplots
Par(mfrow=c(2,2) #Show graphs in two rows and two columns
barplot(Env.raw$*Mo*,main="*Mo*")
boxplot(Env.raw$*Mo*,main="*Mo*")
barplot(Env.raw $*Cu*,main="*Cu*")
boxplot(Env.raw $*Cu*,main="*Cu*")

##Shapiro-Wilk normality tests
#null hypothesis = data is normally distributed. Chosen alpha level = 0.05.
#If p-value < 0.05 then null hypothesis is rejected (data is not normally distributed)
#If p-value is > 0.05 then null hypothesis is supported (data is normally distributed)

Shapiro.test(Env.raw$*Mo*,main="*Mo*")
Shapiro.test(Env.raw $*Cu*,main="*Cu*")

##Data transformation

###Environmental data set
Log.Env.raw <- log(Env.raw) #log transformation
Summary (Log.Env.raw) #Descriptive statistics for the log-transformed data
write.csv(log.Env.raw, file="*Env.log*.csv") # Saving in CSV file. Make sure to set the working directory to the desired destination
###2.4.2 Species data set
spe.hellinger<-decostand(*Spe.raw*,"hellinger") # Hellinger transformation
write.csv(spe.hel, file="*Spe.hel*.csv")

#Variance Inflation Factor
##Upload libraries
library(raster)
library(sp)
library(usdm)

##Calculate VIF
vif1<-vif(Env.log)
vif1
write.csv(vif1, file="Calculated VIF.csv") #Save calculated VIF in CSV file

Reducing variables
##Method1: vifcor
vifcor1<-vifcor(Env.log, th=0.9)
vifcor1

##Method2: vifstep
vifstep1<-vifstep(Env.log, th=10)
vifstep1

##Structure
str(vifstep(acme1, th=10))

##Saving results
write.csv(t(vifstep1@excluded),file="*vif_excluded*.csv")
write.csv(t(vifstep1@corMatrix),file="*vif_corMatrix*.csv")
write.csv(t(vifstep1@results),file="*vif_results(raw)*.csv")
write.csv(t(vifstep1@results$variables),file="*vif_results$variables*.csv")

#Cluster analysis

##Uploading libraries (make sure to download missing libraries)

```
library(ade4)
library(vegan)
library(gclus)
library(cluster)
library(RColorBrewer)
library(labdsv)
library(mvpart)
library(Formula)
library(Hmisc)
library(latticeExtra)
library(fBasics)
library(timeDate)
library(stabledist)
library(MVPARTwrap)
library(timeSeries)
library(rdaTest)

spe.ch<-vegdist(spe.hel, "euc") # use Euclidian distance
t.spe<-t(spe.hel) #tanspose species data for R-mode cluster analysis
t.spe.ch<-vegdist(t.spe, "euc")
```

#Ward's Minimum Variance method

```
spe.ch.ward<-hclust(spe.ch, method="ward.D")
t.spe.ch.ward<-hclust(t.spe.ch, method="ward.D")
plot(spe.ch.ward, ,main="Q-mode: Ward's Minimmum Variance (Hellinger)")
plot(t.spe.ch.ward, ,main="R-mode: Ward's Minimmum Variance (Hellinger)")
```

#Q- vs. R-mode cluster analysis (heatmap)

```
spe.chwo<-reorder.hclust(spe.ch.ward, spe.ch)
t.spe.chwo<-reorder.hclust(t.spe.ch.ward, spe.ch)
dend<-as.dendrogram(spe.chwo)
heatmap(t(spe.hel[rev(or$species)]), Rowv=NA, Colv=dend, col=c("white",
brewer.pal(3,"Reds")), scale="none", margin=c(4,4), ylab="Species", xlab="Sites")
```

#Detrended Correspondence Analysis (DCA)
##Upload libraries

```
library(vegan)
```

##DCA using the Hellinger-transformed species data set

```
spe.dca<-decorana(Spe.hel)
spe.dca # Notice axial length of axis 1 (gradient length). Is it more or less than 2?
plot(spe.dca, display= c("both"), xlim=c(-2,3), ylim=c(-2,2), main="DCA (Hellinger)")
spe.dca
```

#Redundancy Analysis (RDA)
Upload libraries

```
library(ade4)
library(vegan)
library(packfor)
library(MASS)
library(ellipse)
library(FactoMineR)
```

#RDA using log-transformed environmental data + Hellinger-transformed species data (Full model)

```
spe.rda<-rda(Spe.hel~., Enc.log)
plot(spe.rda)
summary (spe.rda)
```

#pRDA with variance partitioning tests (full model)

```
axis<-anova(spe.rda,by="axis", perm= 999)     # Significance of axes
term<- anova(spe.rdat,by="term", perm= 099)     # Significance of variables
axis
term
```

#Save anova results

```
write.csv(axis, file="axis.csv")
write.csv(term, file="term.csv")
```

#pRDA with variance partitioning tests (on selected variables)

```
spe.rda <- rda(Spe.hel ~ As + Ca + P + Ba + Na + Hg + S1 + S2 + Depth + TOC + DO_T
+ pH_B + Total.P + SILT,  Env.log)
plot(spe.rda)
summary (spe.rda)

axis<-anova(spe.rda,by="axis", perm= 999)     # Significance of axes
term<- anova(spe.rdat,by="term", perm= 099)     # Significance of variables
axis
term
```

#Save anova results

```
write.csv(axis, file="axis.csv")
write.csv(term, file="term.csv")
```