WATER DEPTH AND SALINITY CONTROL OF THECAMOEBIAN (TESTATE AMOEBAE) ASSEMBLAGES IN COOTES PARADISE, SOUTHERN ONTARIO, CANADA

WATER DEPTH AND SALINITY CONTROL OF THECAMOEBIAN (TESTATE AMOEBAE) ASSEMBLAGES IN COOTES PARADISE, SOUTHERN ONTARIO, CANADA

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ABSTRACT

High density sampling (n=50) was conducted in Cootes Paradise, a shallow wetland on the western shoreline of Lake Ontario near the city of Hamilton. Cootes Paradise is an urban wetland that has been affected by pollutants and nutrients and invasive carp. Thecamoebian analyses paired with site specific environmental measurements (depth, sp. conductivity, temperature, DO and pH) and substrate characteristics (textural and organic content -LOI) show relationships (R^2 = 0.6) with depth (0-1m) and corresponding sp. conductivity (0.5 to 0.65 mS/cm) and temperature (26.5 to 30.5 °C). Q-mode cluster analysis recognized two biofacies. Biofacies 1 samples (n= 26) are found in the deeper areas (0.70 ± 0.27 m) and dominated by *C. tricuspis* 36 ± 8% (1 std), *L. vas* 18 ± 13% and *D. protaeiformis "claviformis"* 14 ± 6%. Mean water temperature is 28.0 ± 0.6 °C and conductivity at 0.56 ± 0.04 mS/cm. This assemblage has low species diversity (SDI=1.9 ± 0.3) which indicates a transitional environment. Biofacies 2 contains samples (n= 24) which are found in shallower areas (0.38 ± 0.15 m) and the assemblage is characterized by *C. constricta "aerophila"* 25 ± 8%, *C. tricuspis* 18 ± 5%, *Cyclopyxis sp.* 9 ± 6 % and *L. vas* 9 ± 4 %. The SDI for Biofacies 2 is 2.2 ± 0.2 and like Biofacies 1 shows a

transitional environment. The average temperature is and 29.0 ± 1.0 °C with mean sp. conductivity also slightly higher than Biofacies 1 at 0.6 ± 0.04 mS/cm.

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1. General Introduction

The taxonomy of testate amoebae is somewhat confused with biologists and paleontologists taking a very different approach to classification (Lumpers vs splitters; eg. Medioli et al. (1990) vs Ogden and Hedley (1980) fully discussed in Patterson and Kumar (2002). The relationship between taxa and environmental variables has also been problematic for the group as there are no clear species associations that are entirely reproducible. For the most part, the group together with foraminifera, has been useful as an indicator of salinity in estuaries and marshes, but it has also been used in environmental impacts as an indicator of stress (eg. Patterson and Kumar, 2000). The stress is normally expressed by relative changes of diversity (i.e. Shannon Diversity Index; Patterson and Kumar, 2000) both in an areal extent within lake systems or in cores measuring impacts through time (eg. Reinhardt et al., 2005). However, isolating the exact stressor and the thecamoebian response has not always been possible or reproducible (Patterson and Kumar, 2002). Possible stressors that have been examined include temperature (eg. McCarthy et al., 1995; Dalby et al., 2000), dissolved oxygen (eg. Roe et al., 2010), salinity (eg. Neville et al., 2011) and metals (eg. Reinhardt et al., 1998). Linking species or trend has been difficult as the results from one location are not always repeated elsewhere. The

introduction of the strain concept by Dr. F. Medioli (Dalhousie University) and tested in Reinhardt et al., (1998) was in part an attempt to compare the utility of thecamoebians as a group - i.e. the taxonomy was the problem for identifying clear trends (Patterson and Kumar, 2002). There has been some success with this, but its application is still new and universal trends have yet to be defined. The results presented here from Cootes Paradise suggest that sampling biases may also be playing a part, as the relationships with depth (and corresponding salinity and temperature) have never been reported in previous studies.

2. Introduction

Thecamoebians (testate amoebae) are single-cell micro-organisms that exist in fresh to slightly brackish water (Medioli and Scott, 1983; Patterson et al., 1985). Due to their high sensitivity to environmental fluctuations, thecamoebians are used extensively as a paleo-environmental indicator in lacustrine, marine marsh and bog settings (Patterson and Kumar, 2002; Patterson et al., 1985; Charman et al., 1998; Mitchell et al., 2008; Warner et al., 2007). In marine marshes, foraminifera and thecamoebians assemblages are zoned (i.e. high vs low marsh) with elevation. The lower reaches of the marsh consist of agglutinated foraminifera (eg. *Trochammina inflata, Trochammina macrescens*) while

the less saline upper marsh consists of thecamoebians (Scott and Medioli, 1980b; Scott and Medioli, 1986; Kumar and Patterson, 2000; Scott et al., 2001; Charman et al., 1998; Roe et al 2002; Charman et al., 2002; Roe et al., 2010).

Thecamoebians can tolerate low salinity (1-2 ppt; Patterson et al., 1985; Scott and Medioli, 1980c) and they have been found in brackish assemblages in salt marshes up to 6.1 ppt (Patterson et al., 2007). In marine marshes the transition between foraminifera and thecamoebians is a useful marker for reconstructing past sea-level changes in peats. Recently, more detailed work (Charman et al., 1998) examining the transition of thecamoebians communities in the high marsh zone found a secondary zonation between thecamoebians and elevation in South Wales, UK.

There are no specific studies that attempt to use thecamoebians to reconstruct water levels (< 1 m) in lacustrine settings like Gehrels et al. (2001) and Roe et al. (2002). General trends have been defined (eg. Fig. 5-1 in Scott et al., 2001) which include: a wide range of agglutinated thecamoebians such as *Difflugia, Pontigulasia, Centropyxis, Lagenodifflugia, Bullinularia, Hoogenradia, Cyclopyxis, Hyalosphenia, Nebela, Pseudodifflugia, Cucurbitella* in ponds and lakes while wet niches like forest mosses, wet soil and vegetation and sphagnum bogs are characterized by organic walled *Plagiopyxis, Pontigulasia, Euglypha,*

Tracheleuglypha, Lequereusia, Nebela, Heleopera, Archerella and *Amphitrema.* However, there have been no studies that have examined thecamoebians as a water level indicator as they have been used in marine marshes.

The relationship between thecamoebians and environment has been difficult to define with no few clear associations that have proven broadly applicable. Certain taxa have been found to inhabit stressed conditions but often the diversity of thecamoebians is often used as an indicator of ecosystem health (Kumar and Patterson, 2000; Neville et al., 2008, 2011; Roe et al., 2010). Generally, the relationship between distribution of thecamoebian and environmental variables is studied on the lake basin scale with low number of samples collected over a large area which are often then compared to other local or regional lake basins (eg. Roe et al., 2010). Changes in diversity within these systems or in cores are largely used as the indicator of impact, rather than the presence/absence of species. In order to try and clarify some of the species/ environmental relationships, strains were used to characterize water quality in Peterson and Crosswise lakes, near Cobalt, Ontario (Reinhardt et al., 1998). Strains are artificial taxonomic groupings of species with similar morphologies that are thought to reflect eco-phenotypes. In the Cobalt example, thecamoebian assemblages provided an assessment on the level of contamination (cyanide and

mercury from silver ore processing) and remediation in the lakes. The study found some associations with contamination as found in previous work (eg. Asioli et al., 1996). Kumar and Patterson (2000) studied the relationship between acidity levels and thecamoebian distributions James Lake in northeast Ontario and found a good trend with *Arcella vulgaris* and diversity.

More elusive have been clearly defining the relationships with 'natural' variables. Many of the contamination studies examine heavily polluted settings often at extremes with impacts on all life forms (e.g. Asioli et al., 1996; Patterson et al., 1996; Reinhardt et al., 1998; Kumar and Patterson, 2000; Dumaresq, 1993). However, there have been few if any clear relationships between natural variables such as temperature or nutrients. McCarthy et al. (1995) examined lake cores from Newfoundland and Nova Scotia and tried to make associations between pollen and thecamoebian records but did not find any strong relationship. Differences in the response of the indicators was thought to play a role (terrestrial vs aquatic), but even the stronger associations (e.g. *Pontigulassia compressa*) have not been reproduced in other studies. One strong association with eutrophication is with *Cucurbitella tricuspis* and *Spirogyra* algae which have been found in numerous studies (eg. Reinhardt et al., 2005; Scott et al., 1983)

Roe et al. (2010) examined interrelationships between environmental

controls and thecamoebian distribution in seventy-one surface samples from twenty-one lakes and ponds in the Greater Toronto Area. They examined water properties (eg. pH, temperature, DO, conductivity, water depth and lake area), substrate type and bioavailable metals and sediment based phosphorous. They found that thecamoebian assemblages had a strong association with phosphorous in the lakes and on a more localized scale, conductivity played a role.

Compared to other proxies (eg. diatoms; Stoermer and Smol, 1999) the association between assemblages and environment is weak and an effort with the strains (ecophenotypes) is not leading to any major discoveries ('strain concept' is discussed in Reinhardt et al, 1998). It could be that microenvironmental trends play a large role in thecamoebian distribution and trends are missed due to low sampling density. In many of the previous studies, water quality data was collected for the lake as a whole with less emphasis on the sample specific characteristics and the samples are often spread over multiple lakes often with low sample density (eg. Roe et al., 2010 mostly used between 1-6/lake). This study examines whether a higher sampling density may prove worthwhile in finding environmental controls on thecamoebian distributions by pairing high

density sampling with site specific water quality measurements in Cootes Paradise.

2.1. Cootes Paradise Wetland

Cootes Paradise is a natural wetland located at the westernmost end of Hamilton Harbor, Lake Ontario and is part of the Royal Botanical Gardens (RBG). The wetland is separated from Hamilton Harbor by Highway 403 and the Burlington Bridge. Spencer, Borer and Chedoke Creeks are the three main tributaries which are draining into Cootes Paradise wetland. Spencer creek is the largest one, which provides approximately 80% of Cootes Paradise water (Chow-Fraser et al., 1998). Several smaller creeks such as Delsey Creek, Westdale Creek, Long Valley Brook, Hickory Brook and Highland Creek drain from the surrounding escarpment into Cootes with the Dundas wastewater treatment plant (WTP) discharging at the western end of Desjardins Canal via West Pond (Poulton et al., 1996; Chow-Fraser et al., 1998).

Cootes Paradise is one of the forty-two 'Areas of Concern' for the Hamilton Harbor Remedial Action Plan according to the Great Lakes water quality agreement between Canada and United States government instituted in 1972. The area is managed by the Royal Botanical Gardens which contains a 840

ha wildlife sanctuary containing 250 ha of coastal wetland. The area is the largest marshland in the west end of Lake Ontario (Whillans, 1982) and plays a vital role for migrating birds and conservation of several fish, mammals and amphibians. Cootes Paradise is ranked a first class wetland by the Government of Ontario's Ministry of Natural Resources (Peros et al., 2005). Aquatic plants had extensive coverage across the wetland prior to the 20th century. However, by the 1930's, marsh vegetation was reduced to 85% and declined to 15% by 1985. Twenty-four submergent plant species were recognized in Cootes Paradise in 1949, but only 10 species were found in 1970 (Chow-Fraser et al., 1998; Chow-Fraser 2005) .

Cootes Paradise has experienced a variety of stresses affecting biodiversity since European settlement (circa 1840); a result of agricultural, industrial and commercial development (Whillans 1982). Early settlers drained wetlands in the watershed by constructing dams and clearing forests for agricultural and residential purposes (Whillans 1982; Bowlby, McCormack and Heaton, 2009). During 1827, the Desjardins Canal was constructed and completed in ten years (1837) to provide ship access to the Dundas Valley from Lake Ontario via Hamilton Harbor (Judd, 1953). The canal extends from the east side of Cootes Paradise and the edge of the town of Dundas with Hamilton Harbor through the Burlington Heights ship canal. The canal went out of use in the late nineteenth

century although remnants of the canal can still be seen in aerial photos, the rotten wood pilings still protruding above the substrate (Judd, 1953).

Today, the area surrounding Cootes Paradise is heavily urbanized with high sediment inputs and nutrients (Holems, 1988). The Dundas wastewater treatment plant (WTP) and sewer overflows (CSOs) (at Sterling Street) provide excess nutrients (nitrogen and phosphorous) to Cootes (Semkin et al. 1977; Quinn et al., 2004; Mayer et al., 2008). Semkin et al. (1976) estimated the phosphorous loading from the WTP to be at approximately 45 kg d^{-1} in the early 1970s. Chow-Fraser in unpublished data estimated that mean total external phosphorous that has been arrived into wetlands between 1989 and 1995, 56 kg d^{-1}. The high levels of nitrogen and phosphorous have caused eutrophication and decreased oxygen levels affecting submerged plants (Harris and Smith 1977; Ontario Ministry of the Environment, 1985). WTP and the CSOs discharges are not only major sources of nutrients but are also contribute pharmaceuticals, hormones, and other wastewater contaminants (Mayer et al., 2007; Mayer et al., 2008).

Spencer Creek discharges large volumes of sediments during the spring and summer through the ice melt, flooding and storm events (Chow-Fracer, 1998). Semkin et al. (1977) calculated that Spencer Creek discharged approximately 1.600 kg d^{-1} of sediment into the wetland between 1974 to 1975.

Suspension of fine sediments has caused increased water turbidity especially in the open water area (east part of the wetlands; Chow-Fraser, 1998) which has been amplified with the decline of emergent vegetation and increased wave action and re-suspension (Chow-Fraser, 1998). Increased eutrophication has also caused water turbidity problems with increased phytoplankton blooms (Chow-Fraser et al. 1998; Lougheed et al., 2004; Bruekelaar et al., 1994)

Hamilton Harbor is another source of pollutants for Cootes Paradise. Hamilton Harbor is still an active port with an area of 21.5 km^2 and is known as a one of the most contaminated sites at the western end of Great Lakes and communicates. Hamilton Harbor is connected to Cootes Paradise through the Burlington ship canal (Bachtiar et al., 1996). Steel production since the early twentieth century, wastewater treatment plants and untreated urban sewers and industrial discharges have caused widespread contamination (Bachtiar et al., 1996; Pozza et al., 2004). The contaminants include: polycyclic aromatic hydrocarbons (PAH), polychlorinated biphenyls (PCB), heavy metals, phenols and a range of other toxins (Pozza et al., 2004; Poulton, 1987; Poulton et al., 1996; Mayer and Nagy, 1992; Mayer et al., 2008; Table. 1).

The Hamilton Harbor Remedial Action Plan (HH-RAP) included Hamilton Harbor and Cootes Paradise and was initiated in 1986. Since

environmental impacts in Hamilton Harbor affect Cootes Paradise as well, RBG and HH-RAP have had numerous partnerships to decrease water contamination, re-establishing vegetation, control invasive Asian carp (*Cyprinus carpio*) and promoting wildlife habitat (Environment Canada, 1996; Bowlby, McCormack and Heaton, 2009). Carp has been a major problem for Cootes Paradise with its introduction to Lake Ontario in 1896 (Painter et al. 1989). The Carp prefers to spawn in weedy shallow water (less than a meter depth) in late spring and early summer when water temperature reaches between 17 to 24 °C with Cootes Paradise representing an ideal habitat for spawning and as a nursery (Crivelli, 1981; Scott and Crossman 1973; Panek, 1987). The carp caused increased water turbidity through foraging and feeding on mollusks, insects, worms, crustaceans, algae, seeds and plant material in the bottom sediments disrupting plant roots (Scott and Crossman 1973; Billard, 1995). Carp spawning also increases nutrient loading (i.e. total phosphorus) adding to the eutrophication and phytoplankton problem (Lougheed et al., 1998). Approximately 90,000 carp were removed between 1952 and 1956 (Simser, 1982) and a survey in 1959 indicated that the emergent plants coverage had increased by 40% (Chow-Fraser et al., 1998); however continued efforts have not produced a full remediation back to vegetation levels prior to the 1930s. A more recent control effort started in 1996

with the construction of control weirs (fishways) to restrict access between Hamilton Harbor and Cootes Paradise preventing carp and (other non-native fish) from entering (McCrimmon, 1986; Environment Canada, 1996). However, full exclusion of the carp has not been possible as they occasionally breach the fishway during storms or floods (Court and Bowman, 2011).

Re-establishment of native vegetation in Cootes Paradise has been a major challenge for the RBG. In the existing wetland there exist two different ecological systems; the impacted open water which represents 74% of the area and the vegetated wetland which comprises the remaining 26% (Lee et al., 1998). Emergent cattails (*Typha*), bulrushes (*Scirpus*), arrowhead (*Sagitaria*), sedges (*Carex*) and submergent pondweeds (*Potamogeton*), wild celery (*Vallisneria*), Canada waterweed (*Elodea*) and the floating leaf water lilies (*Nymphaea*) are dominant indigenous species in Cootes (Bowen, 1998; Hagen, 1996). Actions to control the invasive species including purple loosestrife (*Lythrum salicaria*) and reed manna grass (*Glyceria maxima*) started in 1993 with an active cultivation and replanting program of indigenous plants (Bowen, 1998; Hagen, 1996). At present, the dominant plant species in Cootes Paradise is the invasive Eurasian manna grass (*Glyceria maxima*) which is mostly found on the open shorelines, deciduous swamps and swamp thickets and covers ~31.6 hectares. Cattails (*Typha sp.*) and

willows (*Salix sp.*) are found in the marsh meadow and woodland deciduous communities and occupy ~26 ha and 13.4 ha respectively (Daw, 2011; Lee et al., 1998).

The maximum water depth (1.8 m) is found near the Desjardins Canal on the western side of the wetland. Annual water levels vary by ~ 0.5 meters (Painter et al., 1989; Figure 1) and fluctuate through inter-annual and seasonal changes in precipitation and evaporation in the Great Lakes region and episodic events such as floods, storm surges and ice jams (Quinn, 2002). Water level on Lake Ontario is measured by Canadian Hydrographic Services at the Burlington gauge station (relative to mean sea-level; http://www.waterlevels.gc.ca/C&A/Graphs/burlington.htm) and is identical to Cootes Paradise. A water-level measuring board is located at the Fishway which is matched to the Burlington gauge. Water depths are referenced (1918-1999) at the deepest part of the wetland and ranged from ~ 0.8 m in the beginning of the year and reached their highest level at 1.2 m in the months of April and May. The water levels decline through the summer in the fall due to evaporation which continues into the fall and the inflowing creeks are lower as the landscape plants are growing and utilizing the precipitation until they become dormant in the fall (Court and Bowman, 2011). Average water levels and the data from year 2010

show that water levels were lower than average in the months of April to June peaking at 0.3 m due to less rainfall. During the rest of 2010, water levels remained very close to previous years values and overall fluctuated by ~ 0.55 m (Court and Bowman, 2011) (Fig. 3).

2.2. Long Term History of Cootes Paradise Wetland

Cootes Paradise and Hamilton Harbor are in the Dundas Valley which is located on the extreme West end of Lake Ontario. The Dundas Valley is ~ 12 km long and extends from Copetown in the west to Hamilton Harbor and Lake Ontario to the East (Edgecombe, 1999). Dundas Valley is urbanized (population ~ 650,000) and as mentioned, suffers from surface and ground water contamination from disposal, industrial and buried chemical storage sites (Brikes and Eyles, 1996). The Dundas Valley is delineated to the north and south by the Niagara Escarpment which is an approximately 40 m high outcrop of Paleozoic rocks (Ordovician to Silurian; Eyles et al., 1997; Eyles, 2002). The escarpment stretches north-westward from New York State, through Ontario, Michigan, and Wisconsin to Illinois (Richard and Kosydar, 1989). The Dundas Valley is a small disjunction in the escarpment (MacCormak, 2002). The Paleozoic strata cover

igneous and metamorphic rocks of the North American craton (Karrow 1987; Eyles, 2002).

The Dundas Valley is infilled with approximately 180 m and 210 m of sediment (Greenhouse and Monier-Williams, 1986; Karrow, 1987; Singer et al., 2003) and probably contains early or pre-Wisconsin deposits (Karrow, 1967; Eyles and Williams, 1992) of glacial, lacustrine and fluvial origin (Karrow, 1963, 1987; Greenhouse and Monier-Williams, 1986).

There are many local and regional aquifers in the Dundas Valley which supply Cootes Paradise, Hamilton Harbor and finally Lake Ontario water through these sediments (Brikes and Eyles, 1996). Lake Ontario during the late Wisconsin was repeatedly influenced by fluctuating lake levels from episodic ice-damming of basin outlets (Karrow, 1974; Eyles, 2002; Barnett, 1992). Dundas Valley accumulated silty clays and immature fine-grained sediments during the high water levels of proglacial Lake Iroquois at approximately 13 ka BP due to washout of suspended and ice-rafted sediments (Eyles and Eyles, 1983; Hicock and Dreimanis, 1992). Much of the sediment deposited in the Cootes today is derived from the erosion of these higher lakestand deposits (eg. van Hengstum et al., 2007). At 11 ka BP Lake Ontario water level was 90 m below its current elevation (Coakley and Karrow, 1994) and has been rising gradually in the

western part the basin due to postglacial uplift at the eastern portion at the outlet near Kingston. Isostatic rebound and Lake Ontario water level in the mid-Holocene was not enough to flood Cootes Paradise (Anderson and Lewis, 1985) but likely formed during the transgression of the western part of Lake Ontario during the Nippissing Highstand (4 - 5 ka BP; Flint et al., 1988; Coakley and Karrow, 1994). Since the end of the Middle Holocene Hypsithermal 2000 ^{14}C years ago climate has been relatively stable in southern Ontario (Yu et al., 1997). Also the Little Ice Age was occurred between 13th and 19th centuries and had influenced forest vegetation significantly (Peros et al., 2005).

Cootes Paradise had a human presence at least by the Late Woodland (AD 500 to 1000; Crawford and Smith, 1996). The Princess Point Complex is known to be a prominent archaeological site for south-central Ontario with approximately 80 settlement locations around the perimeter of Cootes Paradise (Crawford and Smith, 1996; Smith et al. 1997). Pollen and diatom analysis has been performed on cores from Cootes Paradise to understand the ecological changes to the wetland and their link with the archaeological record (Gyoung-Ah Lee et al., 2004, Peros et al., 2005).

2.3. Thecamoebian Characteristics

Thecamoebian assemblages have been used as paleo-environmental indicators in lacustrine, wetlands (eg. bogs) and marine marshes. The following summarizes some of the major ecological relationships.

One of the most abundant thecamoebians in nutrient-rich environments is *Cucurbitella tricuspis* (Medioli and Scott, 1983; Scott et. al. 2001). Investigations show that this species has a parasitic relationship with *Spirogyra* algae which grows in nutrient rich, eutrophic environments (Scott et al., 2001). *C. tricuspis* is not known to have a tolerance for metal rich environments, but they have been identified in high numbers in some studies (eg. Torigai et al., 2000; Reinhardt et al., 1998). However, this is likely due to their 'pseudo' planktic life-habit with *Spirogyra* and living in the less polluted upper water column but these high numbers are due to a lack of other benthic species living on the bottom (Medioli et al., 1987).

The dominance of centropyxids in assemblages is often used as an indicator of ecosystem stress. Centropyxids have been observed in waters with pHs higher than 6.2 (Ellison 1995). They can be found in waters with high concentration of heavy metals, low temperature, brackish, oligotrophic and eutrophic conditions (Kumar and Patterson 2000; Monteiro et al. 1995; Burbidge

and Schröder-Adams 1998; Reinhardt et al., 1998; Trappeniers et al. 1999). Studies have shown that *Centropyxis* spp. can survive and even thrive in various harsh ecosystems such as brackish environments (salinity less than 5 ppt; Medioli and Scott, 1988; Honig and Scott, 1987; Roe et al. 2002) or areas with high concentration of Hg and As (Patterson et al., 1996; Reinhardt et al., 1998). Centropyxids were found in high percentages (*Centropyxis aculeata* and *Centropyxis constricta*) in brackish sediments from isolation basins in Journeays and Gibson Lakes, New Brunswick (Scott and Medioli, 1980; Medioli and Scott, 1988; Patterson et al., 1985; Honig and Scott, 1987). Dallimore et al., (2000) found *C. aculeata* in salinity between 0.1 - 2.0 mg/l in cold (mean annual air temperature -10 °C; mean annual ground temperature -6 to -10 °C) thermokarst lakes on Richards Island, Northwest Territories, Canada and they have been found in the upper reaches of estuaries (Bartlett, 1966; Laidler and Scott, 1996; Gehrels et al., 1998). There has also a relationship with oxygenation that was discussed in Reinhardt et al. (1998) where *C. aculeata* and *C. constrica* dominated shallow vegetated areas with low oxygen levels but also high concentrations of Hg and As in Crosswise and Peterson Lakes, Ontario, Canada.

Arcella vulgaris has been found to dominate acidic environments. Kumar and Patterson (2000) studied the relationship between acidity levels and

thecamoebian distributions in James Lake located in northeast Ontario and found a good trend with diversity. The lake is contaminated by waste material from pyrite processing near the south west of the lake where pH ranges from 2 to 5.5 while the other areas have values at ≈ 6.7 pH. In the low pH south western area with high levels of Fe, Al and SO_4 *A. vulgaris* dominates the assemblage. Correlations between pH, *A. vulgaris* and Shannon Diversity Index show the importance of pH for thecamoebian distributions.

Difflugia protaeiformis has been found in ecosystems with low oxygen levels, high percentage of organic materials, sulfides and sulfites (Asioli et. al 1996). This morph tends to live in alkaline environments (a pH in the range of 6.5 - 7.5) (Kumar and Patterson, 2000) and also have been found in a mine sites with high levels of pollutants (e.g. Hg, As, Cd, Cr, Cu, and Pb; Reinhardt et al., 1998) (Patterson et al., 1996). Reinhardt et al. (1998) found similar results in Peterson Lake (near Cobalt, Ontario) showing that the strain *D. protaeiformis "claviformis"* inhabited contaminated (Hg & As) deepwater lake environments. Similarly, the strain *D. protaeiformis* "rapa" was found in low pH (between 3.9-4.5) (Reinhardt et al., 1998), contaminated areas with copper sulphate and high concentrations of ammonia nitrogen and nitric nitrogen (Asioli et. al 1996).

Thecamoebians (or testate amoebae) have been increasingly used in sea-level research. Initially, thecamoebians were used to distinguish the upland or highest astronomical tide (HAT) which was characterized by a diverse thecamoebian assemblage (e.g. see Scott et al., 2001). However, recent research has found that this zone can be further divided and provide higher resolution sea-level data. The approach uses the <63 micron fraction which was not used in the previous studies (Charman et al., 1998). Studies on both UK and North American (US and Canada) coastlines show the potential of this methodology (Charman et al., 1998; Gehrels et al., 2001; Gehrels et al, 2006). Gehrels et al, (2006) analyzed the distribution of thecamoebian in three sites along the East coast of North America; two sites in USA and one in Canada. They found similar vertical zonation of saltmarsh assemblages in both side of Atlantic Ocean (UK and East coast of North America). They found *Centropyxis cassis, Centropyxis platystoma* and *Difflugia pristis* in lower parts of the salt marsh while *Euglypha tuberculata* type, *Trinema linear* and *Tracheleuglypha dentata* occur in the highest part of the salt marsh. *Centropyxis cassis* is equivalent to *C. constricta,* in Medioli and Scott (1983) and Reinhardt et al., (1998) and *C. constricta "aerophila"* in Kumar and Dalby (1998). *Centropyxis platystoma* is equivalent to *D. oblonga* in Scott and Medioli (1983) and *Difflugia oblonga "glans"* in Kumar and Dalby (1998). The

controls on thecamoebian distribution in marine marshes are not known for certain, although salinity is likely playing a large role combined with wetness (tide level), acidity and substrate type (Gehrels et al., 2001; Gehrels et al. 2006) study.

3. Method of Analysis

3.1. Field Sampling

Forty-four surface samples were collected from the water-sediment interface using an Ekman grab sampler in the middle of August, 2010 (Appendix 1) (Figs. 2a, 2b). Approximately 2 cm of upper sediment was sampled (≈30cm^3) for microfossil analysis. For each location, water depths were recorded with a measuring tape and environmental parameters were recorded with a HydroLab water quality multi probe (MS5). The following parameters were recorded: temperature (± 0.10 °C), dissolved oxygen (± 0.01 mg/L for 0-8 mg/L; ±0.02 mg/L for > 8 mg/L), redox (± 20 mV), pH (± 0.2 units), specific conductivity (± 1% of reading; ± 0.001 mS/cm) and salinity (± 0.2 ppt). In addition, twelve surface samples were collected from three different transects to identify thecamoebian distribution with elevation above lake level (Fig. 2a). Elevations of these samples

was recorded with tape and line level with the horizontal distance of the transects at approximately 4 - 5 meters. Sample locations were recorded using a Garmin eTrex® GPS .

3.2. Microfossils Analysis

Samples (1- 5 cm^3) were sieved with a 38 μm screen for microfossil analysis and a wet splitter was used to subdivide the samples for analysis (Scott and Hermelin 1993). Approximately 250 to 350 thecamoebians were identified in each sample using an Olympus SZX12 at 60 TO 90X magnification. Species and strains were identified using Medioli and Scott (1983); Kumar and Dalby (1998) and Reinhardt et al. (1998) (Fig. 4). Specimens were gold sputter-coated and imaged with a Philips 515 SEM in the Canadian Center for Electron Microscopy - Brockhouse Institute for Material Research at McMaster University.

3.3. Particle Size Distribution (PSD)

Particle size was measured to provide the physical characteristics of the sediment. All samples were analyzed for their particle size distributions using a Beckman - Coulter LS 230 machine and statistical data computed using the Fraunhofer optical model (Murray, 2002; Van Hengstum et al., 2007). Approximately 0.5 cm^3 of sediment samples were first mixed with a 1% sodium

hexametaphosphate solution to disperse the sediment particles before analysis. Conventional particle size statistics such as mean, mode and standard deviation were calculated in μm and converted to phi (ϕ) units (Sambridge et al., 1995).

3.4. LOI

Loss on Ignition (LOI) was used to determine the measure the organic content of the sediments (Dean, 1974; Bengtsson & Enell, 1986). Wet samples (≈ 2 g) were kept in the oven (105 °C) for 24 hours and cooled in a desiccators for 2 - 3 hours and the dry weight (W_{dry}) was measured. Samples were transferred to a muffle furnace, were burnt at 550°C for 3 hours, cooled in a desiccator and again weighed (W_{550} °C). Loss on ignition was calculated by following formula:

$$LOI = \frac{W_{dry} - W_{550C}}{W_{dry}} \times 100$$

3.5. Statistical Analysis

Relative fractional abundance was calculated for each species/strain in the samples used by following formula:

$$X_i = \frac{Ci}{Ni}$$

Where:

C_i is the number of specimens of species;

N_i is the total number of specimens in the sample.

3.5.1. Standard Error

The standard error was calculated by following formula:

$$S_{Xi} = 1.96\sqrt{\frac{X_i(1-X_i)}{N}}$$

Where:

N is the total number of specimens in a sample;

X_i is the fractional abundance of each species/strain.

The standard error was used to eliminate statistically insignificant taxa (Patterson and Fishbein; 1989). If the standard error was greater than the fractional abundance (i.e. the abundance could be < 0) then it was eliminated from the multivariate analysis. Q-mode and R-mode cluster analysis was performed using the statistical computer software PAST - PAST free software package - (Hammer et al., 2001). Ward's minimum variance method was used and the analysis results were displayed as a hierarchal dendrogram showing sample (Q-mode) and species (R-Mode) groupings (Patterson and Fishbein 1989).

3.5.2. SHANNON Diversity Index

The SHANNON diversity index (SDI; Shannon and Weaver, 1949) was calculated for each sample using the formula:

$$\text{SDI} = -\sum_{i=1}^{S}\left(\frac{X_i}{N_i}\right)\times \ln\left(\frac{X_i}{N_i}\right)$$

Where:

X_i is the abundance of each taxon in a sample;

N_i is the total abundance of the sample;

S is equal to the species richness of the sample.

SDI was used as a measure of environmental stability (Shannon and Weaver, 1949) (Appendix 2). SDI values for a stable environment have been found to range between 2.5-3.5, transitional from 1.5-2.5 and stressed are <1.5 (Magurran 1988; Patterson and Kumar 2002).

4. Results

Generally, the eastern area of Cootes Paradise close to the channel connecting it with Hamilton Harbor is the deepest at 1m water depth and it also has the lowest temperatures at 27.5 °C (Appendix 1; Fig.2a). Moving westward the depths become shallower < 0.5 m and the temperature tends to be higher at

28-29 °C along with sp. conductivity (0.50-0.59 vs >0.60 mS/cm; Appendix 1). pH and DO do not seem to follow any consistent pattern and the substrate is uniformly muddy (12-18 μm; Appendix 1; Table 3) with LOI values ranging mostly between 3-8% (Appendix 3). Generally, temperature and salinity decreased with increasing depth in the wetland - shallow margins particularly in the west had more emergent vegetation had higher temperatures and salinities (Fig. 15).

4.1. Biofacies

Standard error calculations eliminated 12 of the 27 species/strains and the remaining taxa include: *Arcella vulgaris, Centropyxis aculeata "aculeata", Centropyxis aculeata "discoides", Centropyxis constricta "aerophila", Centropyxis constricta "spinosa", Centropyxis constricta "constricta", Cucurbitella tricuspis, Cyclopyxis sp., Difflugia oblonga "oblonga", Difflugia oblonga "bryophila", Difflugia oblonga "tenuis", Difflugia oblonga "lanceolata", Difflugia oblonga "linearis", Difflugia protaeiformis "claviformis", Lagenodifflugia vas.*

The Q-mode cluster analysis of the reduced data set determined two assemblages (Biofacies 1 and Biofacies 2; Fig. 5a) while the R-mode cluster analysis showed several groupings of species that followed the Q-mode results

(Fig. 5b). The strong species associations of *D. protaeiformis "claviformis"* with *Lagenodifflugia vas* and *Centropyxis constricta "aerophila"* with *Cyclopyxis sp.* largely defined the Q-mode derived biofacies while the other taxa tended to be less significant.

Biofacies 1 includes 26 samples and is dominated by *C. tricuspis* 36 ± 8%, *L. vas* 18 ± 13% and *D. protaeiformis "claviformis"* 14 ± 6% (Table 2). Most of the samples in Biofacies 1 are located in the east, northeast and central part of the wetland (Fig. 2a). This assemblage has low species diversity (SDI=1.9 ± 0.3) which based on Kumar and Patterson (2002), would indicate a transitional environment. The average depth and water temperature for Biofacies 1 is 0.70 ± 0.27 m and 28.0 ± 0.6 °C and the mean salinity and sp. conductivity is 0.27 ± 0.02‰ and 0.56 ± 0.04 mS/cm (Table 3). pH and DO are 8.39 ± 0.18, 11.0 ± 1.4 mg/l and not appreciably different than the Biofacies 2 values.

Biofacies 2 contains 24 samples which are found in the western and southwestern locations of Cootes Paradise (Fig. 2a). The assemblage is characterized by *C. constricta "aerophila"* 25 ± 8%, *C. tricuspis* 18 ± 5%, *Cyclopyxis sp.* 9 ± 6 % and *L. vas* 9 ± 4 % (Table 2). The SDI for Biofacies 2 is 2.2 ± 0.2 and like Biofacies 1 shows a transitional environment (Table 2). The average depth and temperature is 0.38 ± 0.15 m and 29.0 ± 1.0 °C shallower and a slightly higher

temperature (~1 °C) than Biofacies 1. The mean salinity and sp. conductivity was also slightly higher at 0.29 ± 0.02 ppt, 0.6 ± 0.04 mS/cm (Table 3).

4.2. Species vs. Environmental Variables

The dominant taxa, including *C. tricuspis, Centropyxis aculeata "aculeata", C. constricta "aerophila", Centropyxis constricta "spinosa", D. protaeiformis "claviformis", L. vas, Cyclopyxis sp.,* were cross-plotted to examine their inter-relationship with environmental parameters (Figs 6-10). Overall there were good relationships with water depth, and specific conductivity (salinity), moderate relationships with temperature, but poor relationships with DO, pH and sediment characteristics (mean, mode, std, LOI; Figs. 10-14). Not all taxa showed good relationships with these environmental parameters (depth, sp. conductivity, temperature) but there were a couple species (*C. tricuspis, D. protaeiformis "claviformis"*) that had consistently high R^2 values.

Water depth ranged from 0.05 to 1.1 m and showed the best relationships with four taxa (*C. tricuspis, C. aculeata "aculeata", C. constricta "aerophila", D. protaeiformis "claviformis"*) which have R^2 values > 0.3 (Fig. 6). *D. protaeiformis "claviformis"* had the strongest relationship with water depth, with an $R^2 = 0.6$ and was more abundant at deeper depths which was also the case with *C.*

tricuspis and *L. vas. C. aculeata "aculeata", C. constricta "aerophila"* and *Cyclopyxis sp.* had inverse relationships and tended to dominate shallower depths. SDI values showed little relationship with water depth ($R^2 = 0.2$)

The overall the specific conductivity values are low and have a small range of values (0.48 - 0.65 mS/cm). However, despite this short range, there is a relatively good relationship between several taxa and specific conductivity (salinity) particularly *D. protaeiformis "claviformis"* and *C. tricuspis* which have a respective $R^2 = 0.4$ and 0.3 (Fig. 7). These taxa tend to decrease in abundance with increasing conductivity. The other taxa and the SDI don't show any strong relationship with conductivity with R^2 values less than 0.1 (Fig. 7).

Water temperature ranged from 27- 30 °C and had moderately good R^2 values (≈ 0.3) with several species (*C. constricta "aerophila", C. tricuspis, D. protaeiformis "claviformis"*). *Cyclopyxis sp.* showed a good relationship with an $R^2 = 0.4$ and its abundance increasing with temperature.

Measured DO values had a narrow range from 8 to 14 mg/l, with most of the values between 10 and 12 (Fig. 10). The pH ranged from 7.8 to 8.9 but most of the values were from 8.4 to 8.6 (Fig. 9). There were very poor relationships with pH and DO - the best R^2 with pH was 0.07 and it was slightly better with

DO where *D. protaeiformis "claviformis"* had an $R^2 = 0.2$ but most of the others it was < 0.1 .

No significant relationship was observed between thecamoebians and particle size. R^2 values tended to be low with most of the values ranging from 0.1 - 0.3. Sediment was all very fine (12-18 μm) and muddy (Fig. 11, Fig. 12, Fig. 13) with no significant range in size amongst the sample locations.

5. Discussion

5.1. Biofacies

The Q-mode cluster analysis found two distinct biofacies that were largely distinguished by their *D. protaeiformis "claviformis"*, *C. constricta "aerophila"*, *C. tricuspis*, *Cyclopyxis sp.* and *L. vas* content. Water depth seems to be a strong control on the distribution of thecamoebians as the water depth range was quite different when considering the standard deviation of the values in the biofacies (depth values; Fig. 6; Table 3). Associated with water depth are the other parameters such as salinity and temperature which generally follow the water depth trend (Fig. 15). This would be expected in the wetland particularly in late summer (August) when the climate is the driest and the air temperature is at the

peak of the season (Court and Bowman, 2011). Many of the shallow areas, particularly in the west have emergent plants (eg. *Typha, Scirpus*) which prevent circulation and interchange with the open water areas causing increased temperatures and evaporation throughout the summer and salinities (conductivity) to rise (Fig.16). The colder and deeper part of the basin close to the Burlington Channel has inputs of colder lake water from Hamilton Harbor which circulates with the open water areas of the basin where there is no vegetation to restrict mixing via waves or currents. This distinction is largely seen in the distribution of thecamoebians in the wetland with Biofacies 2 found in the shallower areas of the western basin and Biofacies 1 in the deeper eastern portions. The controlling factor(s) for the biofacies distribution is hard to determine as there is a strong inter-relationship between water depth, salinity and temperature (Figs. 15). pH and DO are highly variable and do not have good relationships with the biofacies or the individual taxa so they do not seem to be a significant factor, likewise with substrate and OM (LOI) content, they don't seem to be a significant factor in thecamoebian distribution. Depth has the greatest separation between the biofacies, while temperature and salinity have considerable overlap (values; Table 3). Samples in the narrow southern arm by McMaster University campus should cluster with Biofacies 2, as it would appear

to be restricted and vegetated, however, the area is relatively deep (average water depth = 0.8m) compared to Biofacies 1 (<0.5m) and the temperature is lower at approx. 28 °C, but conductivity is slightly high at 0.6 mS/cm which is closer to the Biofacies 2 average. Mayer et al. (2007) showed that the marshy areas near the CSOs and WTP discharge had elevated levels of alkylphenolic contaminants and nutrients which correspond somewhat with the distribution of Biofacies 2 except in narrow southern arm near McMaster University. Some of the conductivity patterns maybe due to this effluent and possibly road salt (Eyles and Meriano, 2010; Roe et al., 2010) but we would expect this type of contamination during the spring run-off vs later in the summer. Neville et al. (2011) found good correlations (R^2 = 0.707) with conductivity with *C. constricta* and *C. aculeata* dominating the elevated conductivities (and also naphthenic acid concentrations) which is a similar case with Biofacies 2. Regardless of the exact cause, Biofacies 2 represents a stressed shallow area vs Biofacies 1 which represents a deeper and more stable environment but the stress level between the two areas is not large as the SDI shows a transitional value for both biofacies (≈ 2; Kumar and Patterson, 2000).

The cross-plots follow the biofacies results and provide insight into the gradational aspect of the relationship between thecamoebians and environment.

Depth has the best relationship in the cross-plots with *D. protaeiformis "claviformis"* having an $R^2 = 0.6$ but other taxa show good R^2 values (i.e. *L. vas; C. constricta "aerophila"*). Conductivity has relatively good relationships ($R^2 = < 0.4$) and temperature to a slightly lesser degree. The relationship with water depth is surprising as it has never been reported before. Depth control is seen in marine marshes as it relates to wetness of the substrate and thus tidal inundation, but it has not been reported in continually submerged lacustrine environments (Gehrels et al. 2001).

The good *D. protaeiformis "claviformis"* relationship with environmental variables and with depth in particular is unusual as it has never been reported as depth limited in previous studies (Fig. 6). *D. protaeiformis "claviformis"* is traditionally been considered a contamination indicator as it has been found in abundance (≈ 60%) in deep As and Hg contaminated settings (Cobalt, Ontario; Reinhardt et al., 1998). Similarly, the *D. protaeiformis* strain "rapa" has been found in contaminated settings in Italy (Copper sulphate; 10 - 80 %; Asioli et al., 1996). The abundance of *D. protaeiformis "claviformis"* in Cootes Paradise is lower (< 20%) and the wetland is not as deep or as contaminated as a mine tailings lake with high As and Hg (Reinhardt et al., 1998; Asioli et al., 1996). Cootes is not a 'pristine' urban water shed, but the metals contamination is not at

the levels seen in the previous studies, although wastewater sewer overflows are a major source of nutrients (e.g., phosphorus) and contributors of pharmaceuticals, hormones, and other wastewater contaminants (Mayer et al., 2008). Mayer et al., 2008 showed that most of these contaminants remain close to their points of entry marshy areas (shallower) and don't seem to disperse in the wetland. It also doesn't appear in this case that *D. protaeiformis "claviformis"* is indicative of contamination as *L. vas* also increases with water depth and it is not normally associated with ecosystem stress. van Hengstum et al. (2010) used the presence of *L. vas* in Mexican cave sediments as an indicator of lower salinity which maybe the case in Cootes Paradise as well, although the salinity variation is much smaller than the Mexican example. *L. vas* was also shown to decline in sediments from Frenchman's Bay after European contact and the beginning of eutrophication (see below). Vegetation may also play a role; Roe et al. (2010) in their examination of lakes in the northern Toronto area examined the role of *Typha* coverage on thecamoebian distributions but found it did not have a significant role and *D. protaeiformis "claviformis"* was not found in great abundance. In the case of Cootes Paradise there was little relationship with LOI or substrate to suggest that *Typha* coverage is a significant factor. The lack of previous data showing *D. protaeiformis "claviformis"* as depth dependant maybe

due to sampling bias, as shallow lake margins are not typically the focus of studies with most sampling occurring at > 1m water depths (eg. Patterson et al., 2002; Reinhardt et al., 1998; Asioli et al., 1996).

The freezing of Cootes Paradise may also play a factor in the biofacies distribution. The thickness of ice in the winter is highly variable but typically ranges between 10-30 cm (personal communication Tys Theysmeyer, Royal Botanical Gardens, Nov 1st 2011) which would affect the taxa in the shallower Biofacies 2 which is mostly centropyxids. In previous research, centropyxids tend to dominate colder environments (eg. cold temperature, Decloitre 1956; Scott and Medioli 1980; Dallimore et al., 2000).

C. tricuspis is a known eutrophication indicator and is found in high nutrient loading settings (Scott and Medioli, 1983; Reinhardt et al., 2005). Work in Frenchman's Bay east of Toronto, Ontario which is a shallow wetland like Cootes Paradise on the margin of Lake Ontario had very high levels of *C. tricuspis* in post-European contact sediments which increased with urbanization in the 1950-60s from eutrophication (up to 70%; Reinhardt et al., 2005). In the case of Cootes Paradise, *C. tricuspis* follows this trend with eutrophication but there is also a depth relationship that has not been previously reported. *C. tricuspis* tends to be found in deeper areas of the wetland (Biofacies 1) and has a

similar relationship as *D. protaeiformis "claviformis"*. Phosphorous levels are high in the wetland (Mayer et al., 2007; 2008) - *Project Paradise* water quality report (2009) indicates the average total phosphorous in Cootes Paradise water and the streams (CSO and WTP) was 140 and 120 mg/l respectively, between the months of May to October, 2009. Phosphorous was found to be a controlling factor in Toronto area lakes as found by Roe et al., (2010). The deeper water probably presents a better habitat for *Spyrogyra* upon which *C. tricuspis* depends in its growth cycle or it could be that the open water areas have more phosphorous compared the more restricted margin areas. *C. tricuspis* was also found in the less impacted areas in Neville et al. (2011) although *C. tricuspis* is often found in contaminated settings because its pseudo-planktic life mode living in the less contaminated upper water column.

C. constricta "aerophila" and to a lesser degree *C. aculeata "aculeata"* were important for distinguishing the biofacies but also showed good relationships with depth increasing in abundance in shallower areas. Others that followed a similar trend were *Cyclopyxis sp.* and also *C. constricta "spinosa"*. *C. aculeata "aculeata"* is often found in marginal environments where the stress is high - Roe et al., (2010) found it in cold, low oxygen environments and it is often found in marginal marine settings (Scott and Medioli 1980, van Hengstum et al., 2008;

Scott et al., 2001) but also in heavily metal contaminated sites and low pH (eg. Reinhardt et al., 1998; Patterson and Kumar, 2000b; Neville et al., 2011). Roe et al., also found *C. aculeata "aculeata"* and to some degree *C. constricta "spinosa"* in a road salt contaminated biofacies indicating the salinity effect. Neville et al. (2011) found it also in elevated conductivities in their wetlands from the oil sands in Alberta and, likewise, van Hengstum et al. (2008) found *C. constricta "aerophila"* was the most euryhaline thecamoebian in Mexican cenotes. The abundance of *C. aculeata* and *C. constricta* strains has also been found to dominate shallow freshwater marshy settings in response to low oxygen conditions that often characterize the high OM content sediments (Sonnenburg et al., 2009; Reinhardt et al., 2010). In the Cootes Paradise results there is an effect with conductivity and possibly contamination with alkylphenolic compounds as the shallow areas are points of concentration which tend to correspond with the Biofacies 2 as discussed (Mayer et al., 2007). However, in Cootes Paradise, low-oxygen conditions and pH do not seem to be a controlling factor and there is no relationship with sediment texture and OM content (LOI). This indicates that salinity and higher temperatures with possibly alkylphenolic contamination are likely a combined stressor. It could also be that stress is also provided through seasonal changes due to water level change (50 cm) occurring on a yearly basis (≈

0.5m), temperatures (- 18.4 °C - 34.5 °C) and associated freezing of the water column and salinity from the spring-runoff to the height of summer - the shallower areas would be more affected by these changes than the deeper open water areas.

5.2. Depth Relationship

The depth relationship and its associated parameters (specific conductivity and temperature) are invoking a strong control on the distribution of thecamoebians in Cootes Paradise that have not been reported in other studies (eg. Scott et al., 2001). The lack of findings may relate to the density of sampling and water depth interval used in this study which differs from previous research. Previous studies (eg. Reinhardt et al., 1998) have typically focussed on deeper lake basins rather than shallow and marshy areas, and the shallow areas are not sampled at a high resolution as they can be difficult to access. Also, in most instances the sampling resolution in any given lake basin is relatively low and samples are often scattered among several lake basins. Environmental information is often provided by several measurements in the basin but not necessarily at each sampling site (eg. Reinhardt et al., 1998). In this study, the sampling resolution was fairly high (n=50), over a narrow depth range (<1m) and

environmental information was collected at each site allowing trends to be seen that may have been overlooked in the past. The lack of previous data makes it difficult to compare depth trends with other locations and assess their reproducibility, however further work may show that thecamoebians can be a useful indicator of changing water depths in lacustrine systems on par with sea-level and marine marshes (Scott et al., 2001; Gehrels et al., 2001; Gehrels et al. 2006).

6. Conclusions

D. protaeiformis "claviformis", *L. vas* and *C. tricuspsis* tend to increase in abundance with depth (range: 0-1m), and correspondingly lower salinity (sp. conductivity range: 0.5 to 0.65 mS/cm) and temperatures (range: 26.5 to 30.5 °C). In contrast, *C. constricta "aerophila"*, *C. aculeata "aculeata"* and *Cyclopyxis sp.* tend to dominate the shallow vegetated areas that have elevated temperatures and salinities relative to the deeper open water areas. The direct relationship between temperature and salinity suggests that evaporation is dictating the depth trends. The lack of previous results maybe due to sampling strategy both in the density of samples in individual lake basins but also the depth range (0-1m). More research is needed to demonstrate the reproducibility of these results

from Cootes Paradise by examining other shallow wetland basins around the Great Lakes and other parts of the world.

7. References

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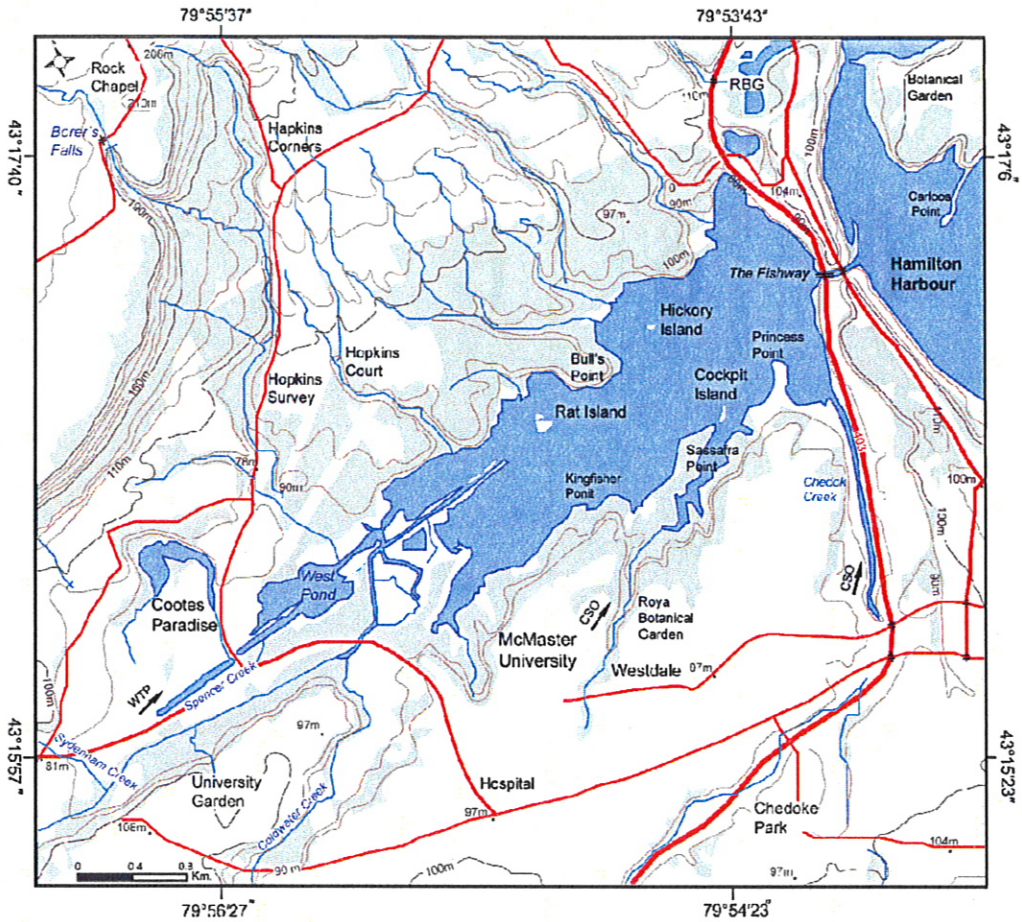


FIGURE 1. Cootes Paradise topography contours are at 10 m intervals.

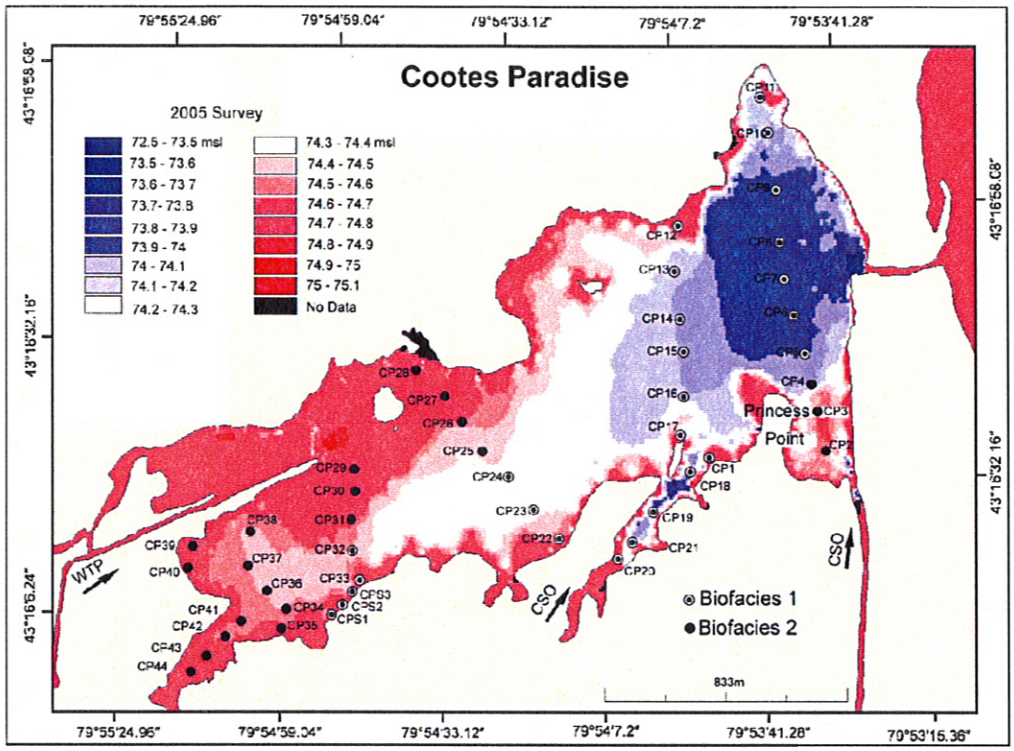


FIGURE 2a. Cootes Paradise bathymetry map (RBG – 2005) with sample and biofacies locations.

FIGURE 2b. Cootes Paradise aerial photograph (Google earth image accessed on September 2009).

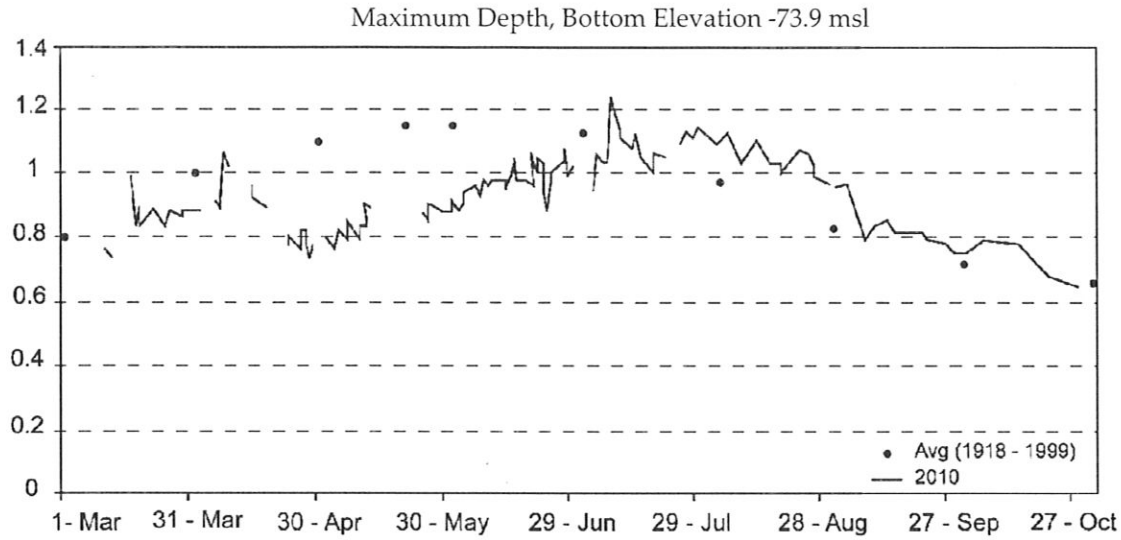


FIGURE 3. Cootes Paradise water depths during 2010, based on Fishway water level data (RBG Project Paradise Report, 2011).

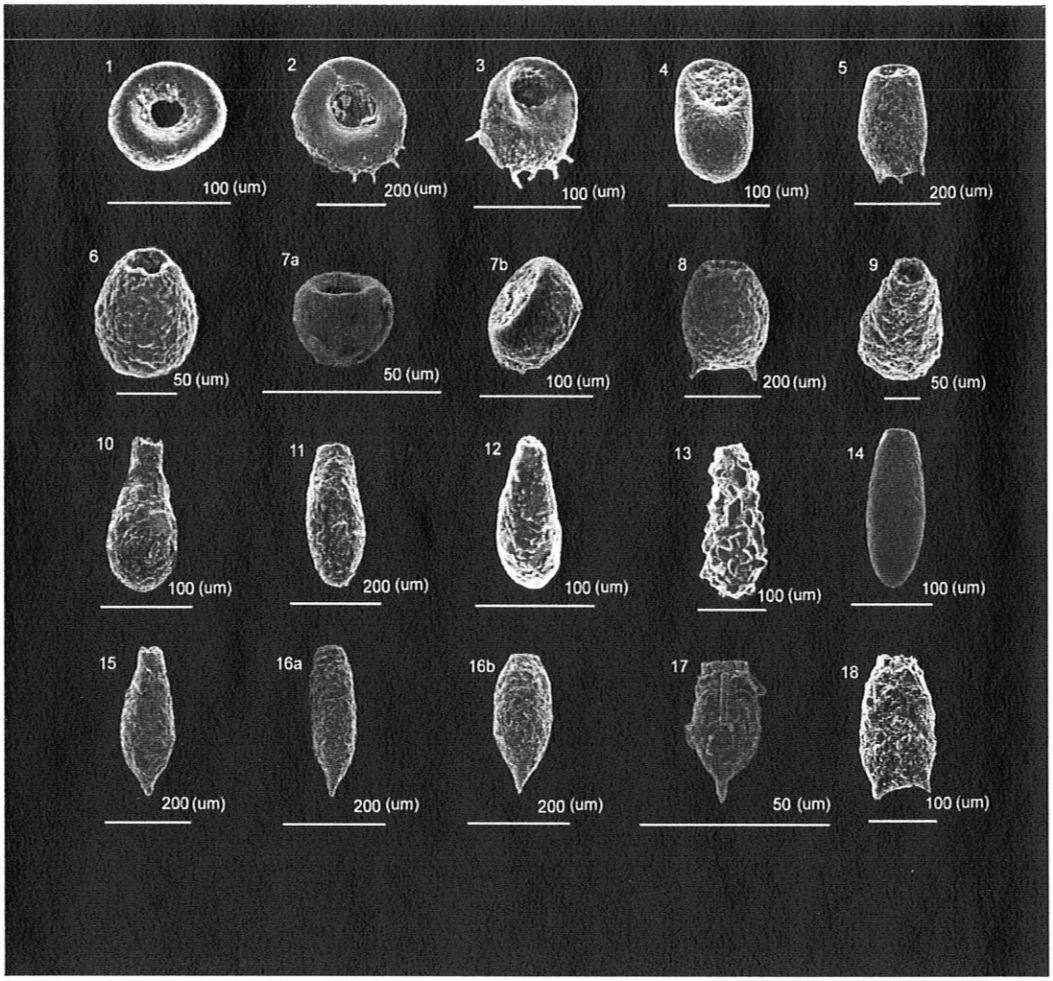


FIGURE 4. 1. *Centropyxis aculeata "discoides"*, 2. *Centropyxis aculeata "aculeata"*, 3. *Centropyxis constricta "spinosa"*, 4. *Centropyxis constricta "aerophila"*, 5. *Centropyxis constricta "constricta"*, 6. *Cucurbitella tricuspis*, 7a. & 7 b. *Cyclopyxis sp.*, 8. *Difflugia corona*, 9. *Lagenodifflugia vas*, 10. *Difflugia oblonga "oblonga"*, 11. *Difflugia oblonga "tenuis"*, 12. *Difflugia oblonga "linearis"*, 13. *Difflugia oblonga "bryophila"*, 14. *Difflugia oblonga "lanceolata"*, 15. *Difflugia oblonga "spinosa"*, 16a. & 16b. *Difflugia protaeiformis "claviformis"*, 17. *Diffugia protaeiformis "amphoralis"*, 18. *Difflugia bidens.*

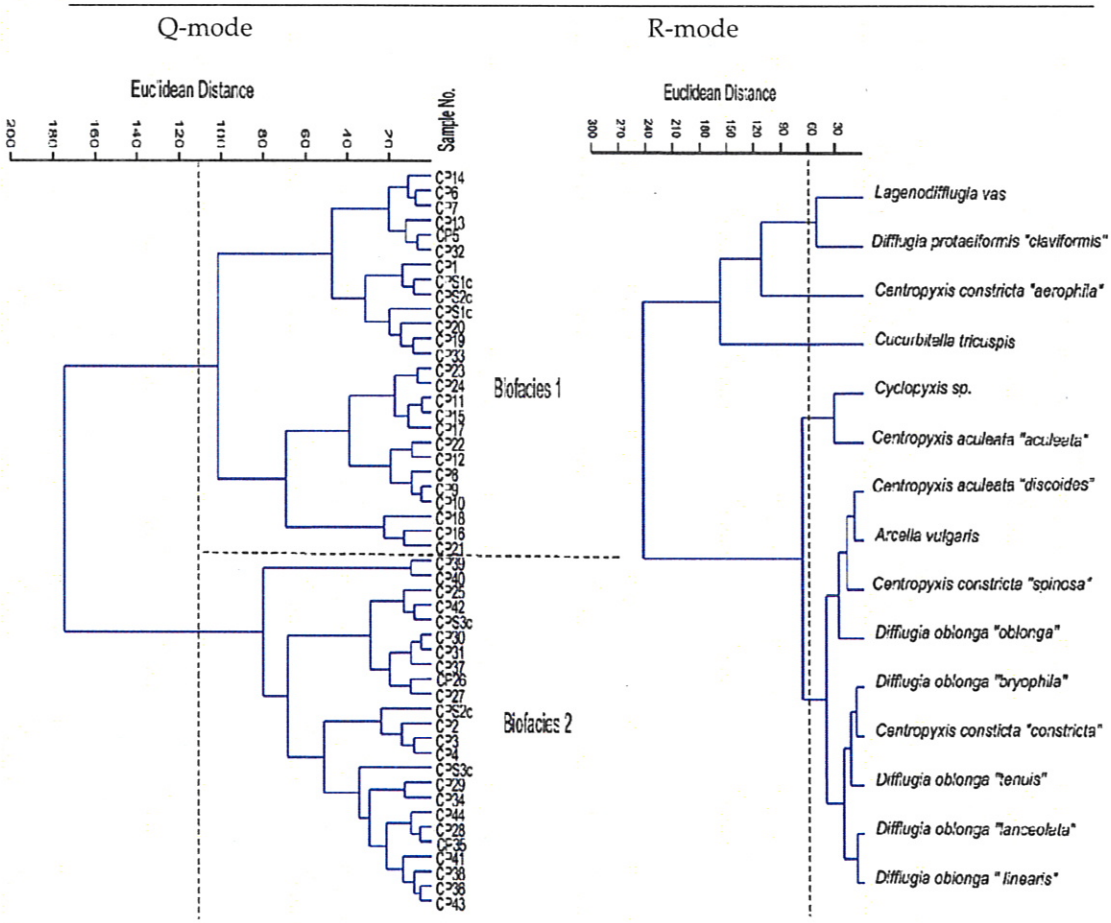


FIGURE 5. a) Q-mode dendrogram and b) R-mode dendrogram

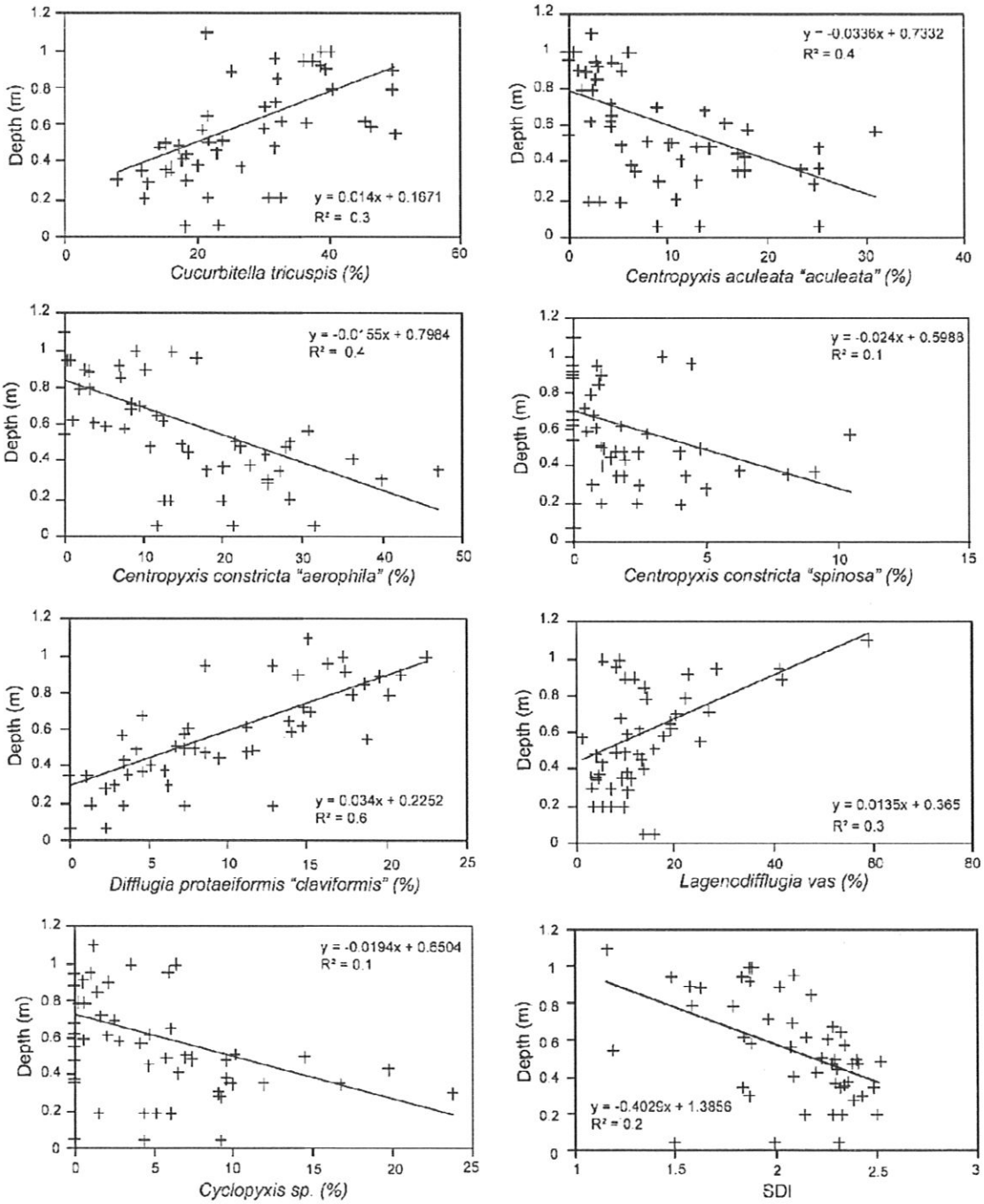


FIGURE 6. Relationship between depth & taxa.

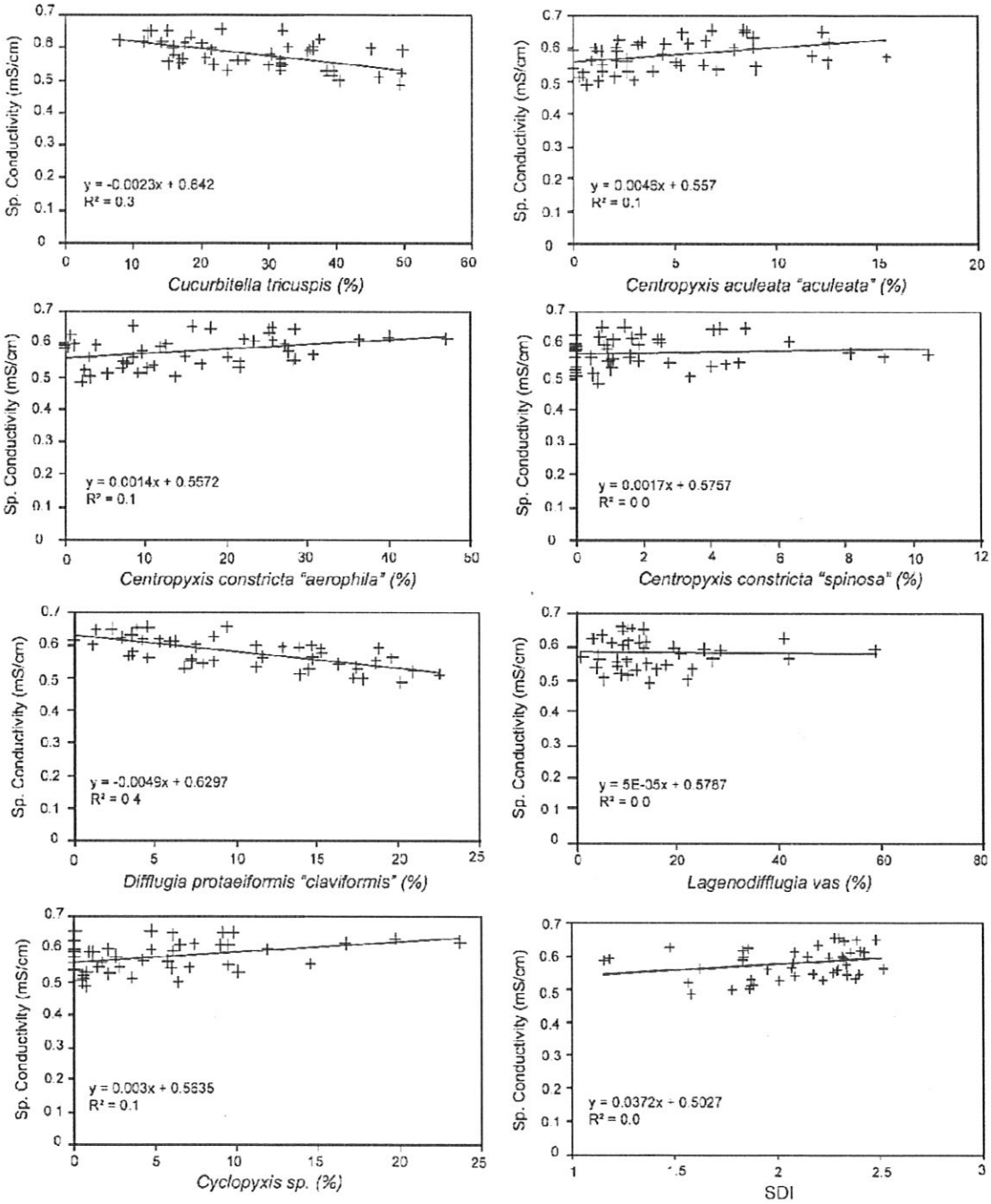
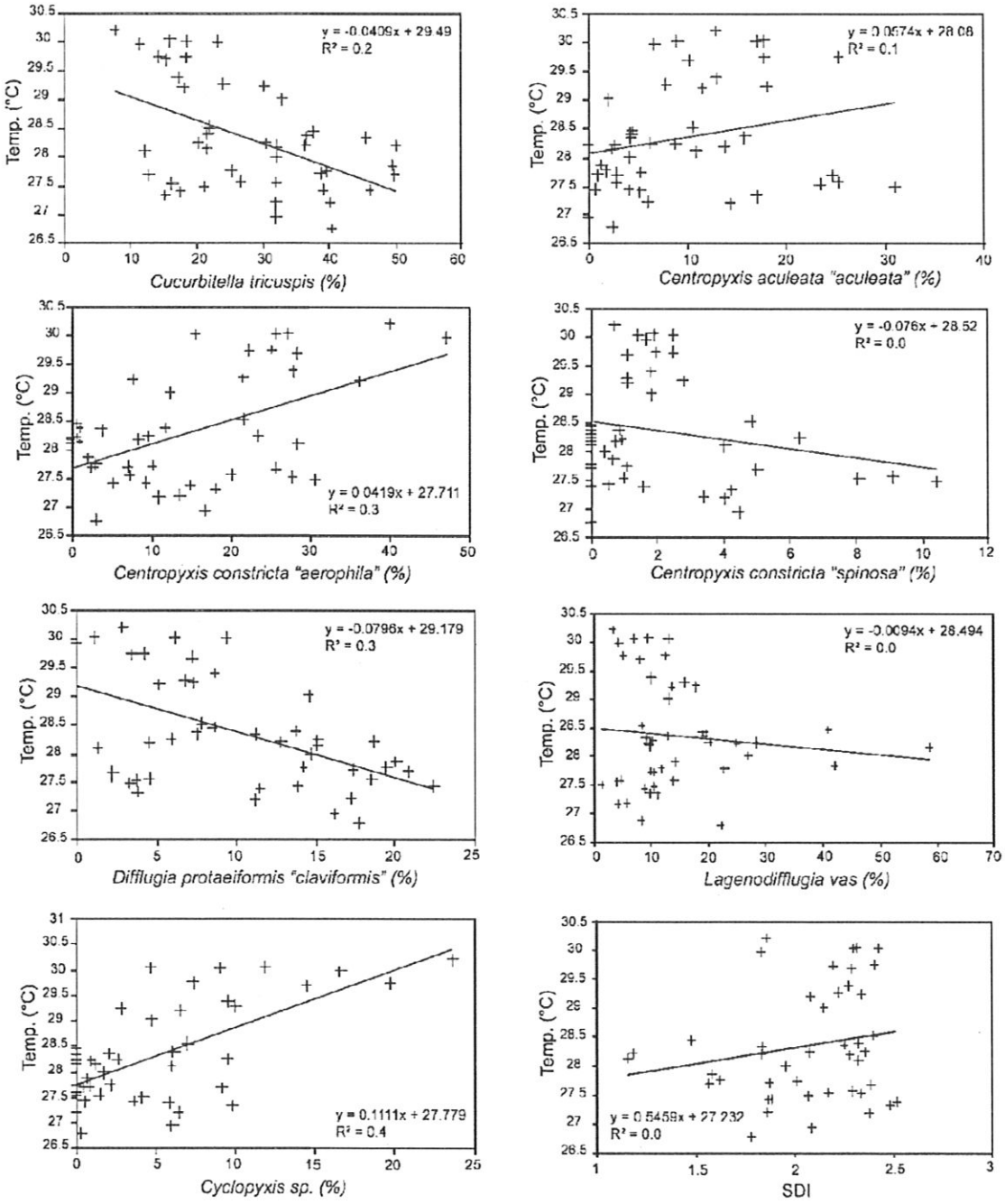


FIGURE 7. Relationship between Sp. Conductivity & taxa.



FIRGURE 8. Relationship between temperature & taxa.

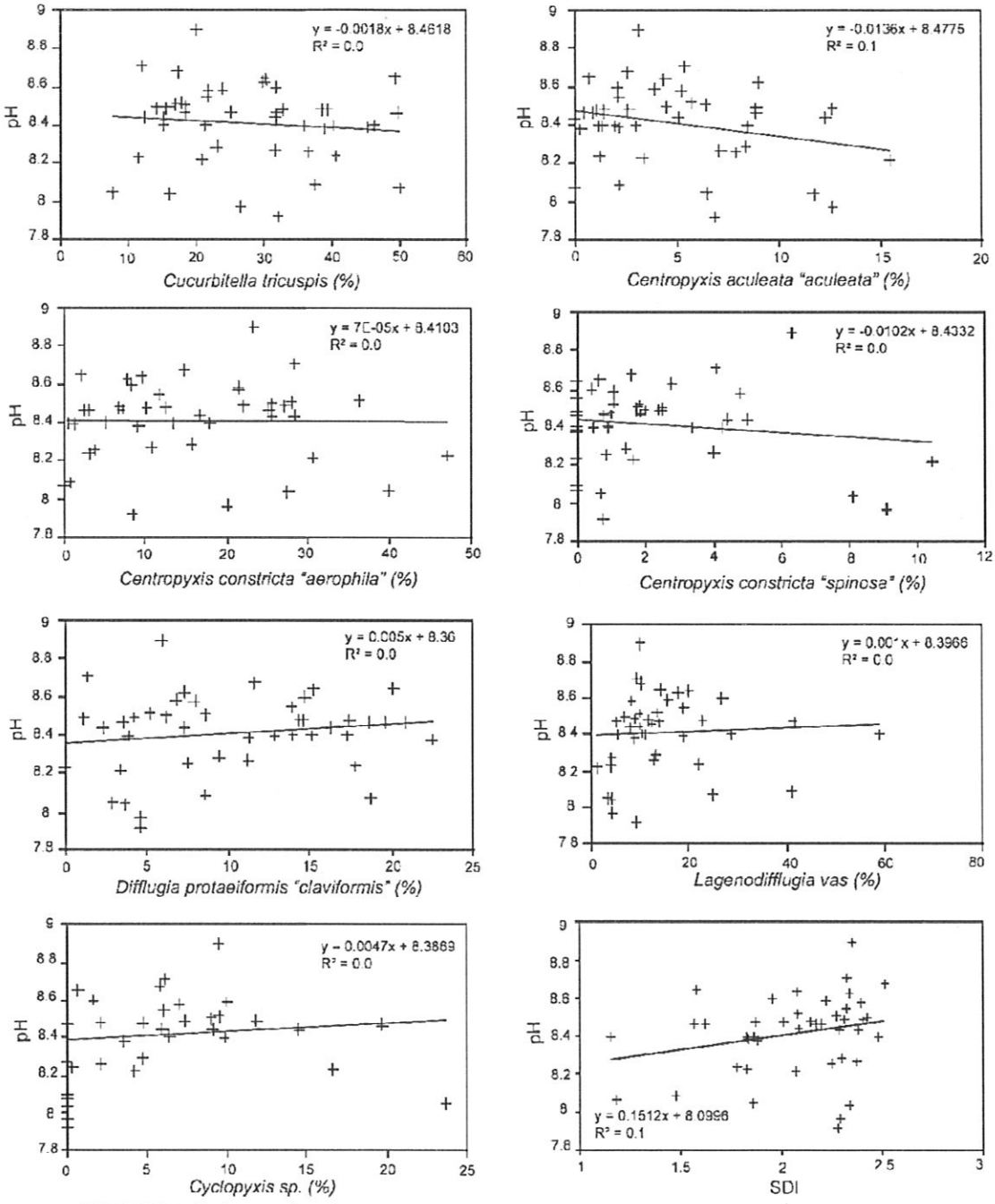


FIGURE 9. Relationship between pH & taxa.

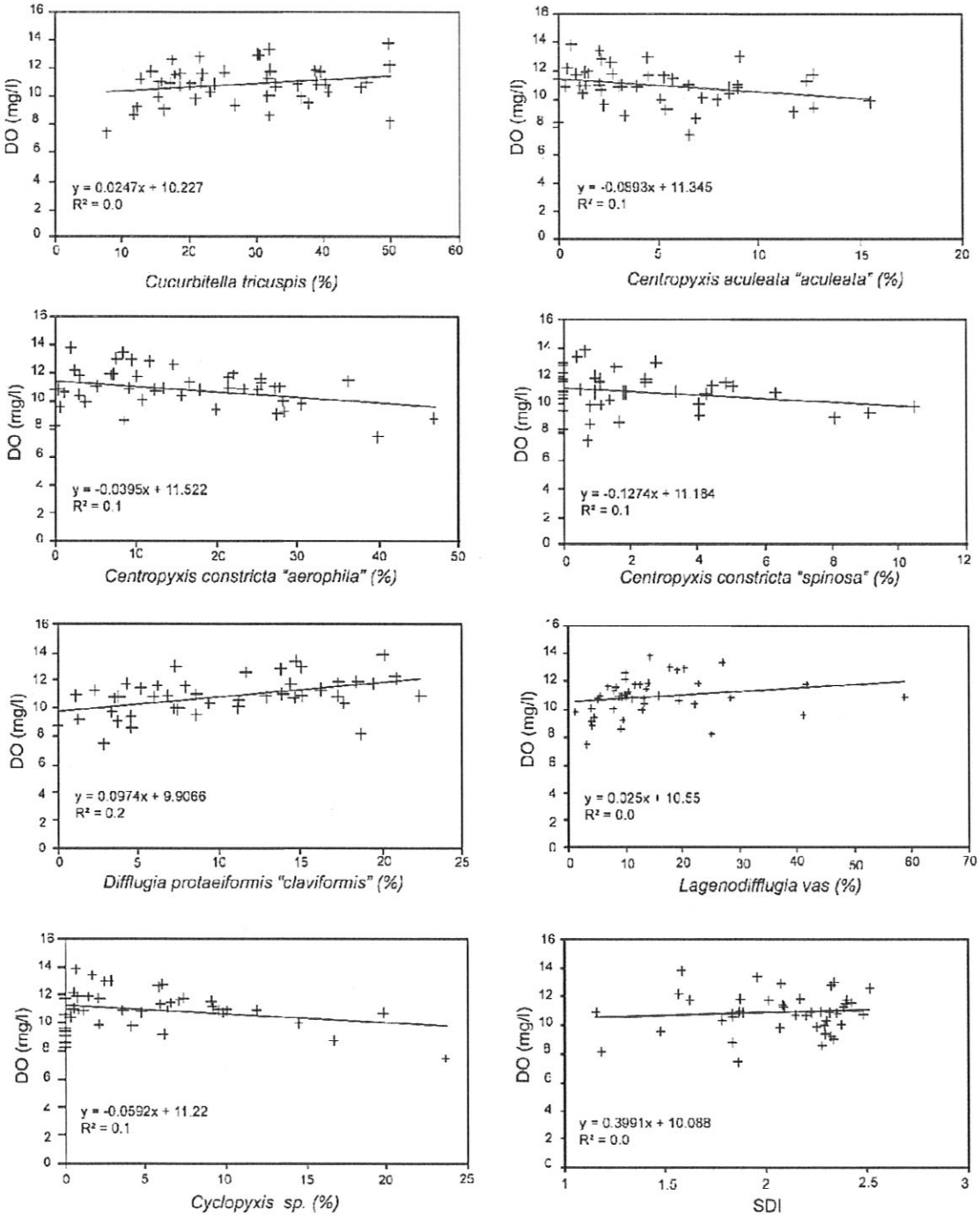


FIGURE 10. Relationship between DO & taxa.

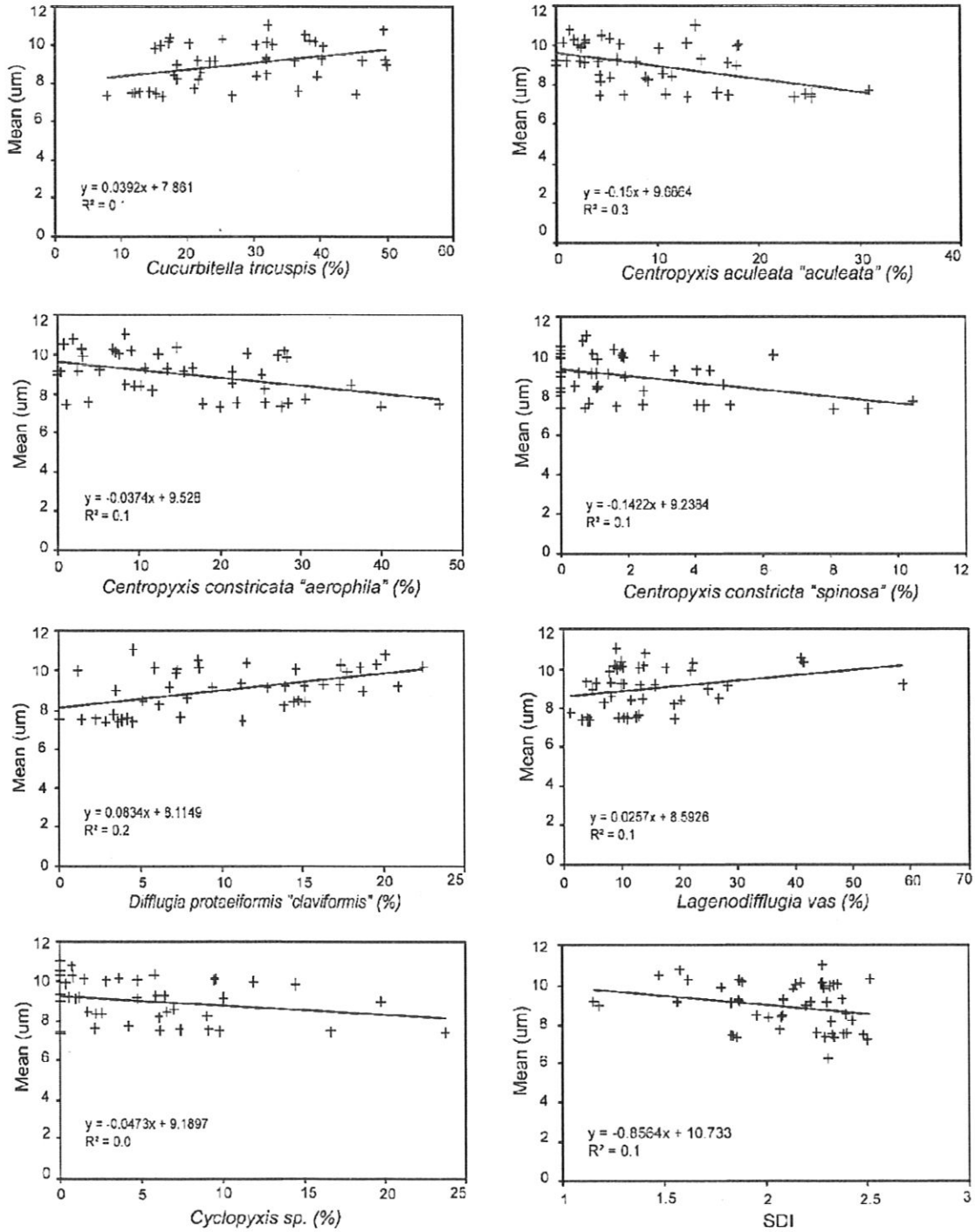


FIGURE 11. Relationship between mean & taxa.

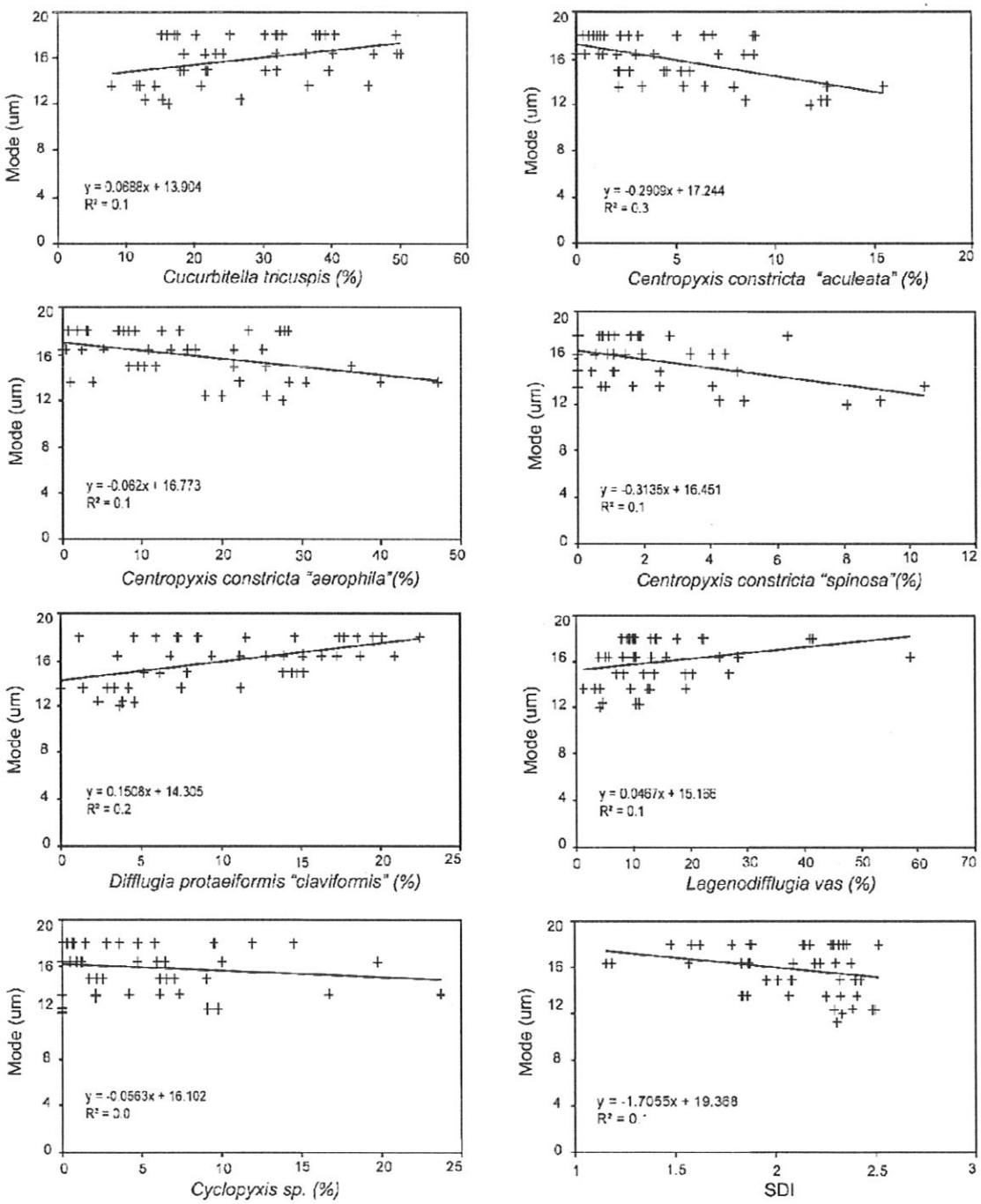


FIGURE 12. Relationship between mode & taxa.

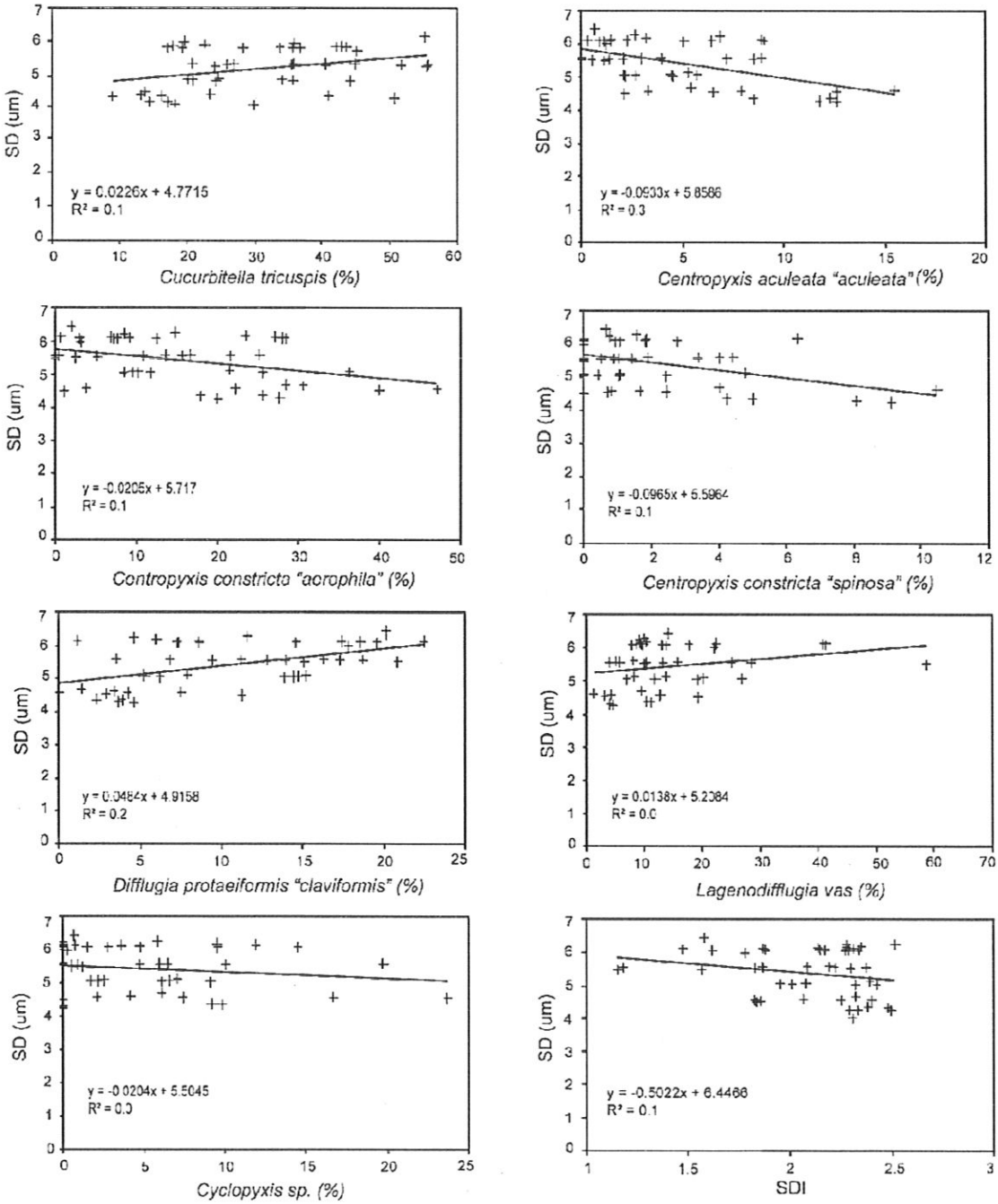


FIGURE 13. Relationship between SD & taxa.

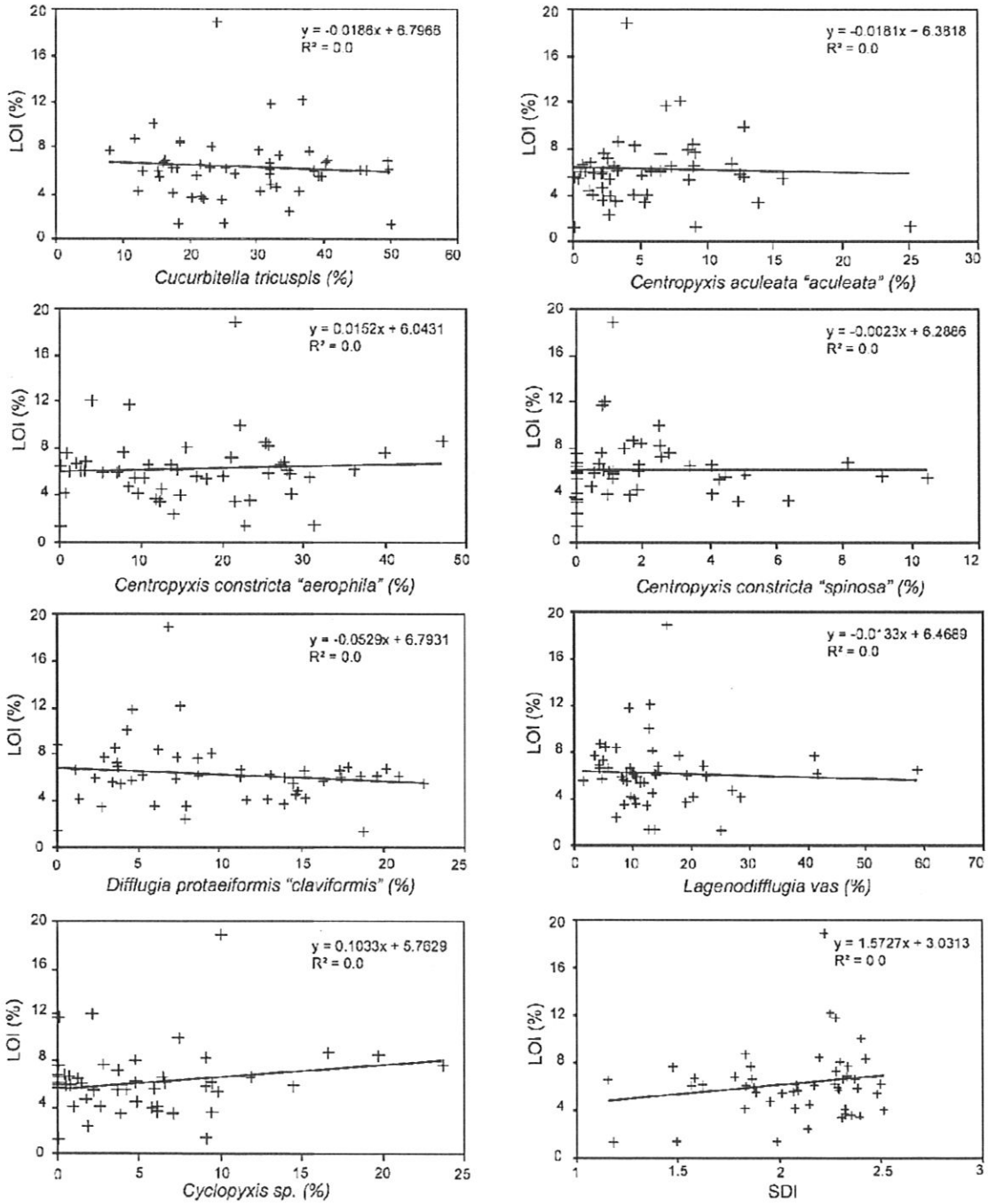


FIGURE 14. Relationship between LOI & taxa.

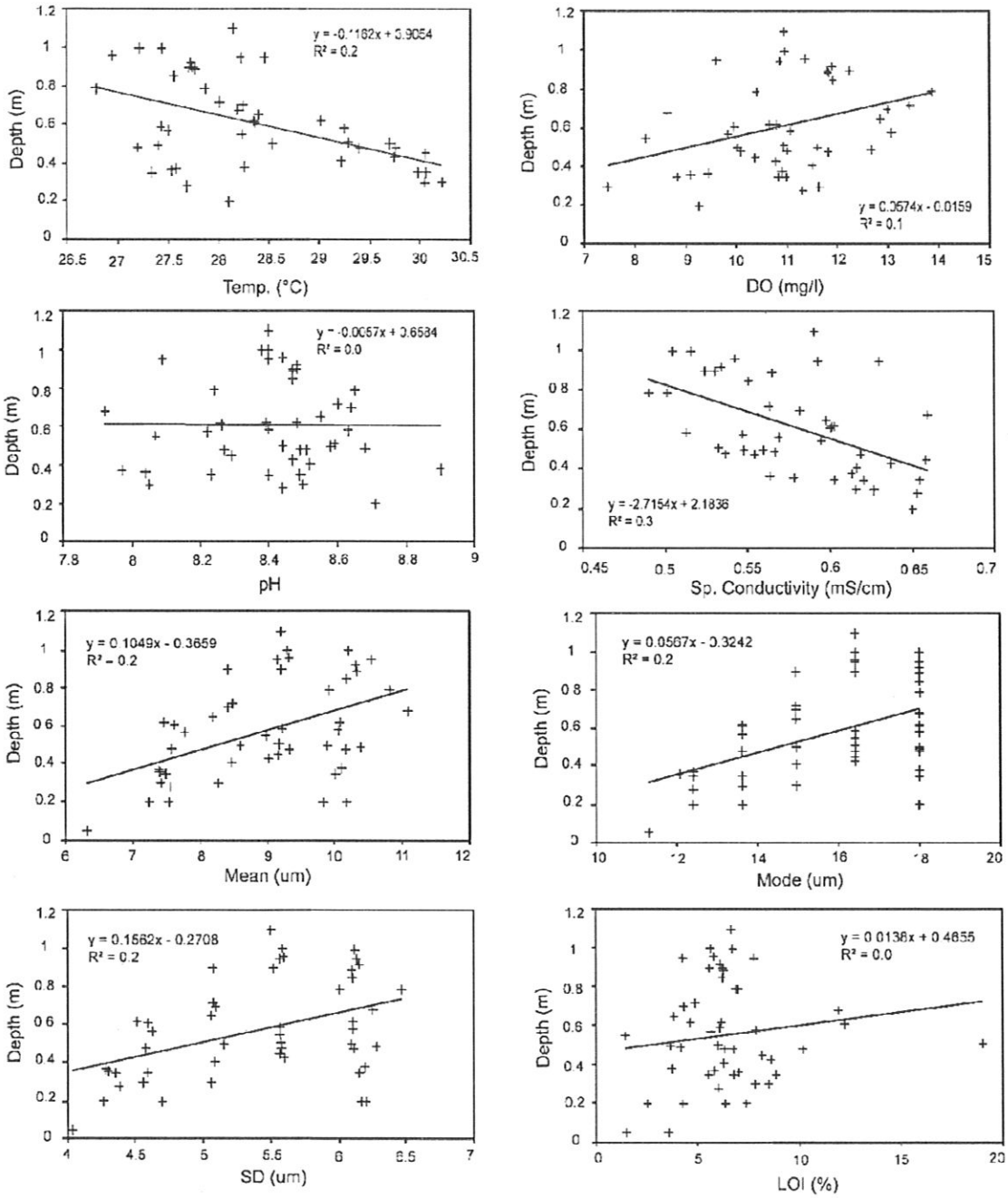


FIGURE 15. Relationship between depth and environmental parameters, Mean, Mode, SD and LOI.

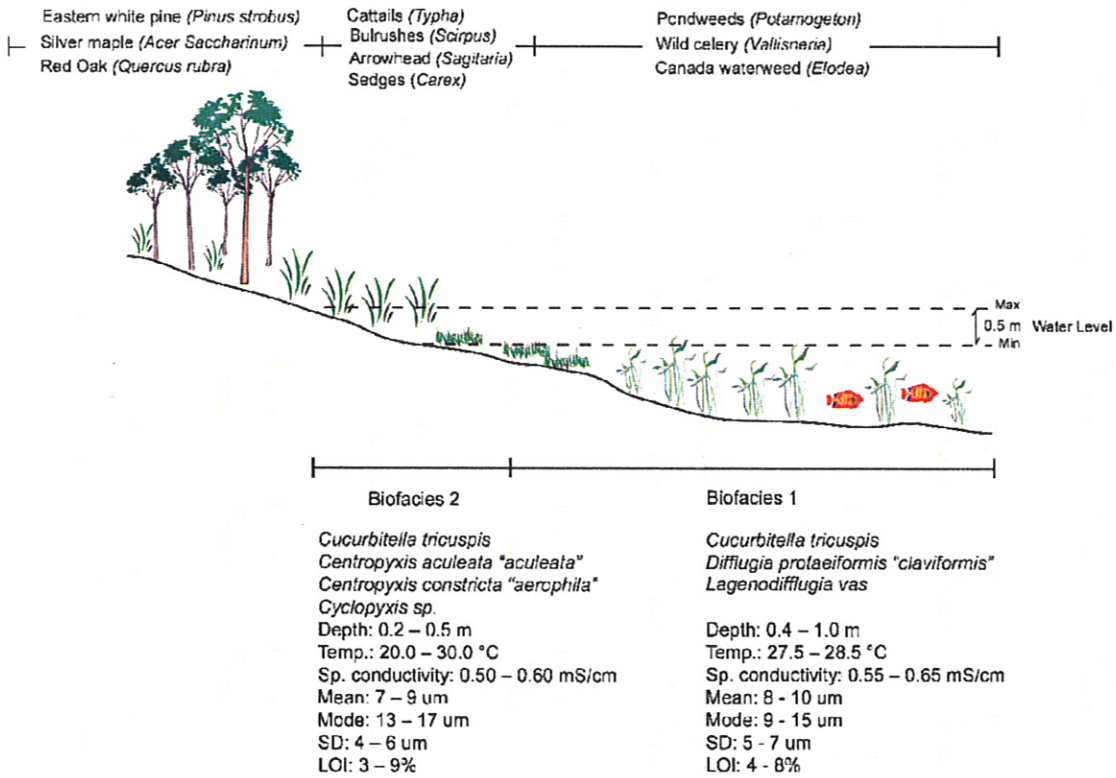


FIGURE 16. Summary of important biofacies characteristics in Cootes Paradise.

Source	Suspended Solids	Zn	Pb	Fe	Cu	Cr
Burlington WTP	1.4	2	0.5	1.1	4.1	1.7
All WTPs	12.1	13.7	3	8.1	21.6	13.5
Cootes Paradise	28.5	5.1	12.9	13.8	9.8	6.3
Steel mills	18.8	53.3	6.4	56.3	9.9	19.7
CSOs	19.2	11.1	49.3	12.6	20.5	8
Stream & urban runoff	13.4	10.8	20.9	6.7	5.7	6.3
Lake Ontario	8.1	6	7.5	2.4	32.6	46.3
Total loadings (kg/day)	44980	109	15.2	3156	14.1	31.8

TABLE 1. Percent contribution by source of suspended materials and selected contaminants to Hamilton Harbor (Canada – Ontario Agreement Review Board, 1992).

Biofacies	1	2
Shannon diversity index (SDI)	1.9 ± 0.3	2.2 ± 0.2
Arcella vulgaris	2 ± 2	3 ± 3
Centropyxis aculeata "aculeata"	3 ± 3	8 ± 5
Centropyxis aculeata "disciodes"	2 ± 2	4 ± 2
Centropyxis consticta "constricta"	1 ± 1	2 ± 2
Centropyxis constricta "spinosa"	1 ± 1	3 ± 3
Centropyxis constricta "aerophila"	7 ± 6	25 ± 8
Cucurbitella tricuspis	36 ± 8	18 ± 5
Difflugia oblonga " linearis"	1 ± 2	1 ± 1
Difflugia oblonga "bryophila"	1 ± 1	2 ± 3
Difflugia oblonga "lanceolata"	1 ± 2	1 ± 1
Difflugia oblonga "oblonga"	5 ± 2	4 ± 2
Difflugia oblonga "tenuis"	2 ± 2	3 ± 2
Difflugia protaeiformis "claviformis"	14 ± 6	5 ± 4
Cyclopyxis sp.	2 ± 2	9 ± 6
Lagenodifflugia vas	18 ± 13	9 ± 4

TABLE 2. Average Shannon diversity index and relative abundance for each taxon in the two biofacies (± 1 Std).

Biofacies	1	2
Depth (m)	0.70 ± 0.27	0.38 ± 0.15
pH	8.39 ± 0.18	8.44 ± 0.20
temp. (°C)	28.0 ± 0.6	29.0 ± 1.0
DO (mg/l)	11.0 ± 1.4	10.6 ± 1.3
Sp. Conductivity (mS/cm)	0.56 ± 0.04	0.6 ± 0.04
Salinity (ppt)	0.27 ± 0.02	0.29 ± 0.02
Mean (um)	9.0 ± 1.0	8.0 ± 1.0
Mode (um)	17.0 ± 2.0	15.0 ± 2.0
SD (um)	6.0 ± 1.0	5.0 ± 1.0
LOI (%)	6.0 ± 2.0	6.0 ± 3.0

TABLE 3. Average environmental variables for the two biofacies (± 1 Std).

Sample No.	Depth (m)	pH (± 0.2 units)	Temperature (± 0.10 ºC)	DO (± 0.01 - ± 0.02 mg/L)	Sp. Conductivity (± 0.001 mS/cm)	Redox (ORP) (± 20 mV)	Salinity (± 0.2 ppt)
CP1	0.48	8.27	27.2	10.07	0.5361	437	0.26
CP2	0.37	7.97	27.58	9.43	0.5632	452	0.27
CP3	0.36	8.04	27.54	9.09	0.5775	448	0.28
CP4	0.57	8.22	27.5	9.83	0.5684	447	0.28
CP5	0.96	8.44	26.95	11.35	0.542	434	0.24
CP6	1.0	8.4	27.22	10.94	0.5043	437	0.24
CP7	1.0	8.38	27.44	10.94	0.5153	438	0.25
CP8	0.59	8.4	27.43	11.05	0.5123	442	0.25
CP9	0.9	8.47	27.71	12.23	0.5236	444	0.25
CP10	0.79	8.65	27.87	13.85	0.5897	438	0.24
CP11	0.79	8.24	26.79	10.4	0.501	446	0.24
CP12	0.55	8.07	28.23	8.19	0.5945	449	0.29
CP13	0.85	8.47	27.56	11.91	0.5504	438	0.27
CP14	0.9	8.48	27.75	11.78	0.5299	437	0.26
CP15	0.92	8.48	27.73	11.89	0.5335	440	0.26
CP16	0.89	8.47	27.77	11.81	0.5644	441	0.27
CP17	0.95	8.4	28.22	10.85	0.5924	443	0.29
CP18	1.1	8.4	28.15	10.93	0.5902	443	0.28
CP19	0.61	8.26	28.36	9.95	0.5995	446	0.29
CP20	0.68	7.92	28.2	8.63	0.6592	455	0.32
CP21	0.95	8.09	28.46	9.6	0.6296	444	0.3
CP22	0.62	8.39	28.35	10.64	0.6005	444	0.29

Appendix 1. Environmental variables for each sample stations.

Appendix 1. Continued.

Sample No.	Depth (m)	pH (± 0.2 units)	Temperature (± 0.10 °C)	DO (± 0.01 - ± 0.02 mg/L)	Sp. Conductivity (± 0.001 mS/cm)	Redox (ORP) (± 20 mV)	Salinity (± 0.2 ppt)
CP23	0.72	8.6	28.01	13.42	0.5625	441	0.27
CP24	0.7	8.64	28.25	12.98	0.5815	441	0.28
CP25	0.65	8.55	28.4	12.83	0.5975	444	0.29
CP26	0.49	8.68	27.4	12.65	0.5659	439	0.27
CP27	0.35	8.4	27.34	10.81	0.6541	446	0.32
CP28	0.28	8.44	27.69	11.3	0.6528	446	0.32
CP29	0.2	8.71	28.11	9.25	0.65	450	0.32
CP30	0.38	8.9	28.26	10.88	0.6127	444	0.3
CP31	0.5	8.58	28.54	11.59	0.5474	441	0.26
CP32	0.62	8.48	29.02	10.78	0.6023	442	0.29
CP33	0.58	8.63	29.25	13.05	0.5469	437	0.26
CP34	0.41	8.52	29.22	11.48	0.6163	441	0.3
CP35	0.48	8.49	29.76	11.8	0.6182	439	0.3
CP36	0.48	8.51	29.4	10.99	0.5537	433	0.27
CP37	0.51	8.59	29.29	10.93	0.5322	430	0.26
CP38	0.5	8.44	29.7	10.02	0.5591	433	0.27
CP39	0.35	8.23	29.98	8.81	0.6202	439	0.3
CP40	0.3	8.05	30.22	7.46	0.6265	438	0.3
CP41	0.43	8.47	29.75	10.77	0.6369	432	0.31
CP42	0.45	8.29	30.05	10.36	0.6582	436	0.32
CP43	0.3	8.5	30.05	11.63	0.6157	433	0.3
CP44	0.35	8.49	30.06	10.96	0.6021	432	0.29

Samples	CP1	CP2	CP3	CP4	CP5	CP6	CP7
Water depth (m)	0.48	0.37	0.36	0.57	0.96	1.0	1.0
Total Counts	223	285	247	239	270	266	307
Specimens per cc	178	456	395	382	216	425	491
Shannon diversity index (SDI)	2.37	2.29	2.33	2.07	2.08	1.86	1.88
Arcella vulgaris	6.28	3.16	6.07	2.51	2.59	1.50	0.00
Std. Error ±	3.18	2.03	2.98	1.98	1.90	1.46	0.00
Centropyxis aculeata "aculeata"	7.17	12.63	11.74	15.48	0.00	3.01	0.33
Std. Error ±	3.39	3.86	4.01	4.59	0.00	2.05	0.64
Centropyxis aculeata "discoides"	5.38	5.61	4.86	4.60	4.07	3.76	2.28
Std. Error ±	2.96	2.67	2.68	2.66	2.36	2.29	1.67
Centropyxis consticta "constricta"	1.79	1.05	2.83	0.84	0.00	0.00	1.30
Std. Error ±	1.74	1.18	2.07	1.15	0.00	0.00	1.27
Centropyxis constricta "spinosa"	4.04	9.12	8.10	10.46	4.44	3.38	0.00
Std. Error ±	2.58	3.34	3.40	3.88	2.46	2.17	0.00
Centropyxis constricta "aerophila"	10.76	20.00	27.53	30.54	16.67	13.53	9.12
Std. Error ±	4.07	4.64	5.57	5.84	4.45	4.11	3.22
Cucurbitella tricuspis	31.84	26.67	16.19	20.92	31.85	40.23	39.09
Std. Error ±	6.11	5.13	4.59	5.16	5.56	5.89	5.46
Difflugia oblonga " linearis"	0.90	0.00	0.00	0.00	0.00	0.00	0.00
Std. Error ±	1.24	0.00	0.00	0.00	0.00	0.00	0.00
Difflugia oblonga "bryophila"	3.14	2.11	3.24	0.84	1.11	0.00	0.00
Std. Error ±	2.29	1.67	2.21	1.15	1.25	0.00	0.00
Difflugia oblonga "lanceolata"	0.00	1.05	0.40	0.00	0.00	0.00	0.65
Std. Error ±	0.00	1.18	0.79	0.00	0.00	0.00	0.90
Difflugia oblonga "oblonga"	4.48	2.46	5.26	1.26	4.44	4.51	5.86
Std. Error ±	2.72	1.80	2.78	1.41	2.46	2.49	2.63
Difflugia oblonga "tenuis"	3.14	1.40	1.21	0.42	0.00	0.00	0.98
Std. Error ±	2.29	1.37	1.37	0.82	0.00	0.00	1.10
Difflugia protaeiformis "claviformis"	11.21	4.56	3.64	3.35	16.30	17.29	22.48
Std. Error ±	4.14	2.42	2.34	2.28	4.41	4.54	4.67
Cyclopyxis sp.	0.00	0.00	0.00	4.18	5.93	6.39	3.58
Std. Error ±	0.00	0.00	0.00	2.54	2.82	2.94	2.08
Lagenodifflugia vas	4.04	4.56	4.05	1.26	8.15	5.64	8.79
Std. Error ±	2.58	2.42	2.46	1.41	3.26	2.77	3.17

Appendix 2. Relative abundance, water depth, standard error and Shannon diversity index for taxonomic units.

Appendix 2. Continued.

Samples	CP8	CP9	CP10	CP11	CP12	CP13	CP14
Water depth (m)	0.59	0.9	0.79	0.79	0.55	0.85	0.9
Total Counts	194	201	303	326	16	210	187
Specimens per cc	155	160	242	260	25	168	149
Shannon diversity index (SDI)	1.87	1.56	1.58	1.78	1.18	2.17	2.01
Arcella vulgaris	4.12	1.99	0.66	2.15	0.00	1.43	0.00
Std. Error ±	2.80	1.93	0.91	1.57	0.00	1.60	0.00
Centropyxis aculeata "aculeata"	2.06	0.50	0.66	1.23	0.00	1.43	2.67
Std. Error ±	2.00	0.97	0.91	1.20	0.00	1.60	2.31
Centropyxis aculeata "discoides"	1.03	1.00	0.66	0.00	0.00	2.38	1.60
Std. Error ±	1.42	1.37	0.91	0.00	0.00	2.06	1.80
Centropyxis consticta "constricta"	0.52	0.00	0.66	1.23	0.00	2.38	4.28
Std. Error ±	1.01	0.00	0.91	1.20	0.00	2.06	2.90
Centropyxis constricta "spinosa"	0.52	0.00	0.66	0.00	0.00	0.95	1.07
Std. Error ±	1.01	0.00	0.91	0.00	0.00	1.31	1.47
Centropyxis constricta "aerophila"	5.15	2.49	1.98	3.07	0.00	7.14	10.16
Std. Error ±	3.11	2.15	1.57	1.87	0.00	3.48	4.33
Cucurbitella tricuspis	46.39	49.75	49.50	40.49	50.00	31.90	39.57
Std. Error ±	7.02	6.91	5.63	5.33	24.50	6.30	7.01
Difflugia oblonga " linearis"	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Std. Error ±	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Difflugia oblonga "bryophila"	0.52	0.00	0.00	0.00	0.00	0.95	1.07
Std. Error ±	1.01	0.00	0.00	0.00	0.00	1.31	1.47
Difflugia oblonga "lanceolata"	0.00	3.98	0.00	0.00	0.00	0.00	0.00
Std. Error ±	0.00	2.70	0.00	0.00	0.00	0.00	0.00
Difflugia oblonga "oblonga"	7.22	6.97	6.27	3.99	6.25	9.05	5.35
Std. Error ±	3.64	3.52	2.73	2.12	11.86	3.88	3.22
Difflugia oblonga "tenuis"	0.00	0.00	0.00	0.92	0.00	1.43	0.00
Std. Error ±	0.00	0.00	0.00	1.04	0.00	1.60	0.00
Difflugia protaeiformis "claviformis"	13.92	20.90	20.13	17.79	18.75	18.57	14.44
Std. Error ±	4.87	5.62	4.52	4.15	19.13	5.26	5.04
Cyclopyxis sp.	0.52	0.50	0.66	0.31	0.00	1.43	2.14
Std. Error ±	1.01	0.97	0.91	0.60	0.00	1.60	2.07
Lagenodifflugia vas	10.31	9.95	14.19	22.09	25.00	13.81	11.76
Std. Error ±	4.28	4.14	3.93	4.50	21.22	4.67	4.62

Appendix 2. Continued.

Samples	CP15	CP16	CP17	CP18	CP19	CP20	CP21
Water depth (m)	0.92	0.89	0.95	1.1	0.61	0.68	0.95
Total Counts	276	337	218	172	240	131	268
Specimens per cc	220	269	348	137	192	104	428
Shannon diversity index (SDI)	1.87	1.62	1.83	1.15	2.25	2.27	1.47
Arcella vulgaris	0.72	0.30	0.92	0.58	4.17	3.05	0.00
Std. Error ±	1.00	0.58	1.27	1.14	2.53	2.95	0.00
Centropyxis aculeata "aculeata"	1.45	0.89	1.38	1.16	7.92	6.87	2.24
Std. Error ±	1.41	1.00	1.55	1.60	3.42	4.33	1.77
Centropyxis aculeata "discoides"	1.45	0.89	0.00	0.00	2.08	0.00	0.00
Std. Error ±	1.41	1.00	0.00	0.00	1.81	0.00	0.00
Centropyxis consticta "constricta"	0.72	0.30	0.46	0.00	1.67	0.00	0.00
Std. Error ±	1.00	0.58	0.90	0.00	1.62	0.00	0.00
Centropyxis constricta "spinosa"	0.00	0.00	0.92	0.00	0.83	0.76	0.00
Std. Error ±	0.00	0.00	1.27	0.00	1.15	1.49	0.00
Centropyxis constricta "aerophila"	6.88	2.97	0.46	0.00	3.75	8.40	0.75
Std. Error ±	2.99	1.81	0.90	0.00	2.40	4.75	1.03
Cucurbitella tricuspis	38.41	25.22	36.24	21.51	36.67	32.06	37.69
Std. Error ±	5.74	4.64	6.38	6.14	6.10	7.99	5.80
Difflugia oblonga " linearis"	2.17	1.19	5.50	0.58	3.33	6.87	0.00
Std. Error ±	1.72	1.16	3.03	1.14	2.27	4.33	0.00
Difflugia oblonga "bryophila"	0.36	0.30	0.00	0.00	2.92	3.82	0.75
Std. Error ±	0.71	0.58	0.00	0.00	2.13	3.28	1.03
Difflugia oblonga "lanceolata"	0.36	0.00	0.00	0.00	0.00	8.40	1.12
Std. Error ±	0.71	0.00	0.00	0.00	0.00	4.75	1.26
Difflugia oblonga "oblonga"	1.81	2.08	3.21	0.58	6.25	8.40	2.61
Std. Error ±	1.57	1.52	2.34	1.14	3.06	4.75	1.91
Difflugia oblonga "tenuis"	0.72	0.89	3.67	0.00	2.50	1.53	0.75
Std. Error ±	1.00	1.00	2.50	0.00	1.98	2.10	1.03
Difflugia protaeiformis "claviformis"	17.39	19.58	12.84	15.12	7.50	4.58	8.58
Std. Error ±	4.47	4.24	4.44	5.35	3.33	3.58	3.35
Cyclopyxis sp.	0.72	0.00	0.92	1.16	2.08	0.00	0.00
Std. Error ±	1.00	0.00	1.27	1.60	1.81	0.00	0.00
Lagenodifflugia vas	22.46	41.54	28.44	58.72	12.92	9.16	41.04
Std. Error ±	4.92	5.26	5.99	7.36	4.24	4.94	5.89

Appendix 2. Continued.

Samples	CP22	CP23	CP24	CP25	CP26	CP27	CP28
Water depth (m)	0.62	0.72	0.7	0.65	0.49	0.35	0.28
Total Counts	187	238	158	231	190	235	219
Specimens per cc	299	380	126	369	152	626	175
Shannon diversity index (SDI)	1.83	1.95	2.07	2.32	2.51	2.48	2.38
Arcella vulgaris	1.07	1.68	0.00	0.87	1.58	2.13	3.65
Std. Error ±	1.47	1.63	0.00	1.19	1.77	1.85	2.48
Centropyxis aculeata "aculeata"	2.14	2.10	4.43	2.16	2.63	8.51	12.33
Std. Error ±	2.07	1.82	3.21	1.88	2.28	3.57	4.35
Centropyxis aculeata "discoides"	1.60	0.42	1.27	0.00	2.63	2.13	2.74
Std. Error ±	1.80	0.82	1.74	0.00	2.28	1.85	2.16
Centropyxis consticta "constricta"	1.60	1.26	1.27	3.46	3.68	5.11	1.83
Std. Error ±	1.80	1.42	1.74	2.36	2.68	2.81	1.77
Centropyxis constricta "spinosa"	0.00	0.42	0.00	0.00	1.58	4.26	5.02
Std. Error ±	0.00	0.82	0.00	0.00	1.77	2.58	2.89
Centropyxis constricta "aerophila"	1.07	8.40	9.49	11.69	14.74	17.87	25.57
Std. Error ±	1.47	3.52	4.57	4.14	5.04	4.90	5.78
Cucurbitella tricuspis	45.45	31.93	30.38	21.65	17.37	15.32	12.79
Std. Error ±	7.14	5.92	7.17	5.31	5.39	4.61	4.42
Difflugia oblonga " linearis"	2.67	3.36	2.53	3.03	2.63	0.00	0.00
Std. Error ±	2.31	2.29	2.45	2.21	2.28	0.00	0.00
Difflugia oblonga "bryophila"	2.14	0.84	3.16	3.46	11.05	8.51	4.11
Std. Error ±	2.07	1.16	2.73	2.36	4.46	3.57	2.63
Difflugia oblonga "lanceolata"	0.53	0.00	0.00	1.30	1.05	0.85	2.28
Std. Error ±	1.05	0.00	0.00	1.46	1.45	1.17	1.98
Difflugia oblonga "oblonga"	5.88	1.68	4.43	3.46	4.21	5.96	3.20
Std. Error ±	3.37	1.63	3.21	2.36	2.86	3.03	2.33
Difflugia oblonga "tenuis"	2.14	1.26	3.16	5.19	5.26	2.13	1.83
Std. Error ±	2.07	1.42	2.73	2.86	3.18	1.85	1.77
Difflugia protaeiformis "claviformis"	11.23	14.71	15.19	13.85	11.58	3.83	2.28
Std. Error ±	4.53	4.50	5.60	4.45	4.55	2.45	1.98
Cyclopyxis sp.	0.00	1.68	2.53	6.06	5.79	9.79	9.13
Std. Error ±	0.00	1.63	2.45	3.08	3.32	3.80	3.82
Lagenodifflugia vas	19.25	26.89	20.25	19.05	10.00	11.06	10.50
Std. Error ±	5.65	5.63	6.27	5.06	4.27	4.01	4.06

Appendix 2. Continued.

Samples	CP29	CP30	CP31	CP32	CP33	CP34	CP35
Water depth (m)	0.2	0.38	0.5	0.62	0.58	0.41	0.48
Total Counts	148	253	228	274	288	367	285
Specimens per cc	59	202	91	438	230	587	228
Shannon diversity index (SDI)	2.32	2.35	2.39	2.14	2.33	2.08	2.40
Arcella vulgaris	2.03	2.77	1.75	0.73	1.74	2.18	3.16
Std. Error ±	2.27	2.02	1.70	1.01	1.51	1.49	2.03
Centropyxis aculeata "aculeata"	5.41	3.16	5.26	1.09	9.03	5.72	12.63
Std. Error ±	3.64	2.16	2.90	1.23	3.31	2.38	3.86
Centropyxis aculeata "discoides"	0.00	1.98	4.39	5.47	4.86	2.45	3.51
Std. Error ±	0.00	1.72	2.66	2.69	2.48	1.58	2.14
Centropyxis consticta "constricta"	9.46	3.56	1.75	0.73	2.43	1.36	1.40
Std. Error ±	4.71	2.28	1.70	1.01	1.78	1.19	1.37
Centropyxis constricta "spinosa"	4.05	6.32	4.82	1.82	2.78	1.09	2.46
Std. Error ±	3.18	3.00	2.78	1.58	1.90	1.06	1.80
Centropyxis constricta "aerophila"	28.38	23.32	21.49	12.41	7.64	36.24	22.11
Std. Error ±	7.26	5.21	5.33	3.90	3.07	4.92	4.82
Cucurbitella tricuspis	12.16	20.16	21.93	32.85	30.21	17.98	14.39
Std. Error ±	5.27	4.94	5.37	5.56	5.30	3.93	4.07
Difflugia oblonga " linearis"	0.00	1.19	1.32	1.09	1.39	1.09	1.75
Std. Error ±	0.00	1.33	1.48	1.23	1.35	1.06	1.52
Difflugia oblonga "bryophila"	8.11	2.77	2.63	0.00	1.39	0.82	1.75
Std. Error ±	4.40	2.02	2.08	0.00	1.35	0.92	1.52
Difflugia oblonga "lanceolata"	0.00	0.00	0.00	0.36	2.08	0.54	3.51
Std. Error ±	0.00	0.00	0.00	0.71	1.65	0.75	2.14
Difflugia oblonga "oblonga"	5.41	4.35	4.82	4.74	3.82	1.91	4.21
Std. Error ±	3.64	2.51	2.78	2.52	2.21	1.40	2.33
Difflugia oblonga "tenuis"	3.38	1.98	3.51	3.28	0.69	1.09	4.21
Std. Error ±	2.91	1.72	2.39	2.11	0.96	1.06	2.33
Difflugia protaeiformis "claviformis"	1.35	5.93	7.89	14.60	7.29	5.18	4.21
Std. Error ±	1.86	2.91	3.50	4.18	3.00	2.27	2.33
Cyclopyxis sp.	6.08	9.49	7.02	4.74	2.78	6.54	7.37
Std. Error ±	3.85	3.61	3.32	2.52	1.90	2.53	3.03
Lagenodifflugia vas	9.46	10.28	8.33	13.14	17.71	13.62	12.63
Std. Error ±	4.71	3.74	3.59	4.00	4.41	3.51	3.86

Appendix 2. Continued.

Samples	CP36	CP37	CP38	CP39	CP40	CP41	CP42
Water depth (m)	0.48	0.51	0.5	0.35	0.3	0.43	0.45
Total Counts	326	279	276	240	278	314	212
Specimens per cc	521	446	441	384	444	502	169
Shannon diversity index (SDI)	2.27	2.22	2.29	1.83	1.86	2.19	2.30
Arcella vulgaris	0.00	1.43	4.71	2.92	6.12	4.46	0.47
Std. Error ±	0.00	1.39	2.50	2.13	2.82	2.28	0.92
Centropyxis aculeata "aculeata"	6.44	3.94	5.07	3.33	6.47	8.92	8.49
Std. Error ±	2.66	2.28	2.59	2.27	2.89	3.15	3.75
Centropyxis aculeata "discoides"	3.99	3.94	2.90	2.92	3.96	3.18	1.89
Std. Error ±	2.12	2.28	1.98	2.13	2.29	1.94	1.83
Centropyxis consticta "constricta"	1.53	1.08	0.72	1.25	0.00	0.64	3.30
Std. Error ±	1.33	1.21	1.00	1.41	0.00	0.88	2.41
Centropyxis constricta "spinosa"	1.84	1.08	1.09	1.67	0.72	1.91	1.42
Std. Error ±	1.46	1.21	1.22	1.62	0.99	1.51	1.59
Centropyxis constricta "aerophila"	27.91	21.51	28.26	47.08	39.93	25.16	15.57
Std. Error ±	4.87	4.82	5.31	6.32	5.76	4.80	4.88
Cucurbitella tricuspis	17.18	24.01	15.22	11.67	7.91	18.47	23.11
Std. Error ±	4.09	5.01	4.24	4.06	3.17	4.29	5.67
Difflugia oblonga " linearis"	0.61	0.72	1.09	0.42	0.00	1.27	0.00
Std. Error ±	0.85	0.99	1.22	0.81	0.00	1.24	0.00
Difflugia oblonga "bryophila"	1.23	1.08	0.72	1.25	0.00	0.64	0.00
Std. Error ±	1.20	1.21	1.00	1.41	0.00	0.88	0.00
Difflugia oblonga "lanceolata"	0.92	0.36	0.72	0.83	1.08	0.96	1.89
Std. Error ±	1.04	0.70	1.00	1.15	1.21	1.08	1.83
Difflugia oblonga "oblonga"	3.99	2.87	5.07	3.33	1.80	3.50	8.49
Std. Error ±	2.12	1.96	2.59	2.27	1.56	2.03	3.75
Difflugia oblonga "tenuis"	3.99	2.87	2.17	0.83	1.44	1.27	5.66
Std. Error ±	2.12	1.96	1.72	1.15	1.40	1.24	3.11
Difflugia protaeiformis "claviformis"	8.59	6.81	7.25	0.00	2.88	3.50	9.43
Std. Error ±	3.04	2.96	3.06	0.00	1.97	2.03	3.93
Cyclopyxis sp.	9.51	10.04	14.49	16.67	23.74	19.75	4.72
Std. Error ±	3.18	3.53	4.15	4.72	5.00	4.40	2.85
Lagenodifflugia vas	9.82	15.77	7.97	4.17	3.24	5.10	13.21
Std. Error ±	3.23	4.28	3.20	2.53	2.08	2.43	4.56

Appendix 2. Continued.

Samples	CP43	CP44	CPS1c	CPS1d	CPS2c	CPS2d	CPS3c	CPS3d
Water depth (m)	0.3	0.35	0.05	0.2	0.05	0.2	0.05	0.2
Total Counts	243	269	263	277	16	115	44	253
Specimens per cc	194	215	17	55	3	23	8	50
Shannon diversity index (SDI)	2.42	2.31	2.30	2.28	1.49	2.13	1.98	2.49
Arcella vulgaris	3.29	1.86	6.46	2.89	0.00	4.35	13.64	4.35
Std. Error ±	2.24	1.61	2.97	1.97	0.00	3.73	10.14	2.51
Centropyxis aculeata "aculeata"	4.53	8.92	13.69	2.53	25.00	2.61	9.09	3.16
Std. Error ±	2.61	3.41	4.15	1.85	21.22	2.91	8.49	2.16
Centropyxis aculeata "discoides"	2.88	4.83	5.70	6.14	6.25	6.09	9.09	4.35
Std. Error ±	2.10	2.56	2.80	2.83	11.86	4.37	8.49	2.51
Centropyxis consticta "constricta"	0.82	1.86	0.00	1.44	0.00	0.00	0.00	3.16
Std. Error ±	1.14	1.61	0.00	1.40	0.00	0.00	0.00	2.16
Centropyxis constricta "spinosa"	2.47	1.86	0.00	2.53	0.00	0.00	0.00	0.79
Std. Error ±	1.95	1.61	0.00	1.85	0.00	0.00	0.00	1.09
Centropyxis constricta "aerophila"	25.51	27.14	12.17	20.94	31.25	13.91	22.73	14.23
Std. Error ±	5.48	5.31	3.95	4.79	22.71	6.33	12.38	4.30
Cucurbitella tricuspis	18.52	15.99	24.71	33.21	25.00	34.78	18.18	22.92
Std. Error ±	4.88	4.38	5.21	5.55	21.22	8.71	11.40	5.18
Difflugia oblonga " linearis"	2.06	1.12	0.76	2.89	0.00	2.61	0.00	1.58
Std. Error ±	1.78	1.25	1.05	1.97	0.00	2.91	0.00	1.54
Difflugia oblonga "bryophila"	0.41	0.00	2.28	1.08	0.00	3.48	0.00	0.40
Std. Error ±	0.80	0.00	1.80	1.22	0.00	3.35	0.00	0.77
Difflugia oblonga "lanceolata"	1.65	1.49	0.00	0.00	0.00	0.00	0.00	0.00
Std. Error ±	1.60	1.45	0.00	0.00	0.00	0.00	0.00	0.00
Difflugia oblonga "oblonga"	6.58	6.69	6.08	2.89	0.00	6.09	0.00	4.35
Std. Error ±	3.12	2.99	2.89	1.97	0.00	4.37	0.00	2.51
Difflugia oblonga "tenuis"	2.88	2.60	2.66	5.78	0.00	8.70	4.55	5.53
Std. Error ±	2.10	1.90	1.95	2.75	0.00	5.15	6.15	2.82
Difflugia protaeiformis "claviformis"	6.17	1.12	2.66	3.61	0.00	7.83	0.00	13.04
Std. Error ±	3.03	1.25	1.95	2.20	0.00	4.91	0.00	4.15
Cyclopyxis sp.	9.05	11.90	3.80	3.61	0.00	1.74	9.09	4.74
Std. Error ±	3.61	3.87	2.31	2.20	0.00	2.39	8.49	2.62
Lagenodifflugia vas	7.00	9.29	12.17	4.69	12.50	6.96	13.64	8.70
Std. Error ±	3.21	3.47	3.95	2.49	16.21	4.65	10.14	3.47

Sample No.	Crucible Wt. (g)	Crucible + Wet Sample(Wwet)	Wet sample Wt. (g)	Crucible + Dry Sample (Wdry)	Dry sample Wt. (g)	Moist Content (%)	Crucible + Burnt Sample (W550 °C)	Sample Wt. aftre burnt (550 °C) (g)	LOI (550 °C) (%)
CP1	16.6048	17.9815	1.3767	17.0677	0.4629	66.38	17.0367	0.4319	6.70
CP2	17.5801	19.5998	2.0197	18.2668	0.6867	66.00	18.2271	0.647	5.78
CP3	19.1121	20.4359	1.3238	19.7104	0.5983	54.80	19.6688	0.5567	6.95
CP4	17.3113	18.7563	1.445	17.8774	0.5661	60.82	17.8456	0.5343	5.62
CP5	22.224	23.7418	1.5178	22.7765	0.5525	63.60	22.7448	0.5208	5.74
CP6	18.9152	20.5052	1.59	19.6897	0.7745	51.29	19.6378	0.7226	6.70
CP7	19.4377	20.8794	1.4417	19.9626	0.5249	63.59	19.9333	0.4956	5.58
CP8	16.611	18.2682	1.6572	17.1292	0.5182	68.73	17.0979	0.4869	6.04
CP9	17.798	19.1184	1.3204	18.1893	0.3913	70.37	18.1651	0.3671	6.18
CP10	17.2485	18.9511	1.7026	17.7062	0.4577	73.12	17.6749	0.4264	6.84
CP11	16.4999	17.9925	1.4926	16.9392	0.4393	70.57	16.9087	0.4088	6.94
CP12	18.2607	19.9224	1.6617	19.4326	1.1719	29.48	19.4163	1.1556	1.39
CP13	18.7948	20.3373	1.5425	19.3368	0.542	64.86	19.3033	0.5085	6.18
CP14	17.2054	18.6656	1.4602	17.7174	0.512	64.94	17.689	0.4836	5.55
CP15	17.765	19.5609	1.7959	18.3742	0.6092	66.08	18.3374	0.5724	6.04
CP16	20.1466	21.6796	1.533	20.6541	0.5075	66.89	20.6225	0.4759	6.23
CP17	16.6083	18.3315	1.7232	17.4676	0.8593	50.13	17.4313	0.823	4.22
CP18	18.7175	20.4561	1.7386	19.2394	0.5219	69.98	19.2049	0.4874	6.61
CP19	18.2351	20.5267	2.2916	18.7844	0.5493	76.03	18.7174	0.4823	12.20
CP20	18.2787	20.2255	1.9468	18.6684	0.3897	79.98	18.622	0.3433	11.91
CP21	20.0746	22.0064	1.9318	20.6008	0.5262	72.76	20.5601	0.4855	7.73
CP22	18.0772	19.567	1.4898	18.5982	0.521	65.03	18.5663	0.4891	6.12
CP23	18.7463	20.2948	1.5485	19.3576	0.6113	60.52	19.328	0.5817	4.84
CP24	19.9691	21.7331	1.764	20.7408	0.7717	56.25	20.708	0.7389	4.25
CP25	18.5424	20.0863	1.5439	19.4045	0.8621	44.16	19.372	0.8296	3.77

Appendix 3. LOI data.

Appendix 3. Continued.

Sample No.	Crucible Wt. (g)	Crucible + Wet Sample(Wwet)	Wet sample Wt. (g)	Crucible + Dry Sample (Wdry)	Dry sample Wt. (g)	Moist Content (%)	Crucible + Burnt Sample (W550 °C)	Sample Wt. aftre burnt (550 °C) (g)	LOI (550 °C) (%)
CP26	19.6841	21.122	1.4379	20.5471	0.863	39.98	20.5117	0.8276	4.10
CP27	17.6953	19.4629	1.7676	18.5051	0.8098	54.19	18.4606	0.7653	5.50
CP28	19.7806	21.8186	2.038	20.7294	0.9488	53.44	20.6727	0.8921	5.98
CP29	18.4321	20.9287	2.4966	19.5977	1.1656	53.31	19.5484	1.1163	4.23
CP30	16.7725	18.6834	1.9109	17.6868	0.9143	52.15	17.6532	0.8807	3.67
CP31	21.0289	22.353	1.3241	21.6928	0.6639	49.86	21.6689	0.64	3.60
CP32	22.3383	23.7616	1.4233	22.8602	0.5219	63.33	22.8362	0.4979	4.60
CP33	18.207	19.9244	1.7174	18.5531	0.3461	79.85	18.5261	0.3191	7.80
CP34	18.7867	20.1189	1.3322	19.1933	0.4066	69.48	19.1679	0.3812	6.25
CP35	17.6967	19.1299	1.4332	17.9536	0.2569	82.08	17.9276	0.2309	10.12
CP36	18.0498	19.1974	1.1476	18.4842	0.4344	62.15	18.457	0.4072	6.26
CP37	18.48	19.8187	1.3387	18.659	0.179	86.63	18.625	0.145	18.99
CP38	19.7306	21.1845	1.4539	20.2925	0.5619	61.35	20.259	0.5284	5.96
CP39	17.6035	19.0834	1.4799	18.027	0.4235	71.38	17.9897	0.3862	8.81
CP40	18.5843	19.9494	1.3651	18.9762	0.3919	71.29	18.9457	0.3614	7.78
CP41	16.5225	17.7638	1.2413	16.9746	0.4521	63.58	16.9359	0.4134	8.56
CP42	16.3449	18.2806	1.9357	16.7536	0.4087	78.89	16.7203	0.3754	8.15
CP43	18.1982	19.5211	1.3229	18.5221	0.3239	75.52	18.4948	0.2966	8.43
CP44	18.8115	20.2192	1.4077	19.2646	0.4531	67.81	19.2342	0.4227	6.71
CPS1 c	16.893	18.4963	1.6033	18.0995	1.2065	24.75	18.0572	1.1642	3.51
CPS1 d	20.4032	21.218	0.8148	20.7049	0.3017	62.97	20.6827	0.2795	7.36
CPS2 c	19.3755	20.5829	1.2074	20.259	0.8835	26.83	20.2461	0.8706	1.46
CPS2 d	18.6892	19.9991	1.3099	19.5335	0.8443	35.54	19.5124	0.8232	2.50
CPS3 c	18.6469	20.1783	1.5314	19.834	1.1871	22.48	19.8168	1.1699	1.45
CPS3 d	17.6562	18.9196	1.2634	18.2019	0.5457	56.81	18.1674	0.5112	6.32

Sample No.	Depth (m)	Mean (um)	mm	phi	Median (um)	mm	phi	Mode (um)	mm
CP1	0.48	9.31	0.00931	6.7470031	8.875	0.008875	6.8160372	16.4	0.0164
CP2	0.37	7.377	0.007377	7.08275	7.359	0.007359	7.0862746	12.4	0.0124
CP3	0.36	7.383	0.007383	7.0815771	7.369	0.007369	7.0843154	12.04	0.01204
CP4	0.57	7.762	0.007762	7.0093559	7.487	0.007487	7.0613965	13.61	0.01361
CP5	0.96	9.308	0.009308	6.7473131	8.885	0.008885	6.8144125	16.4	0.0164
CP6	1.0	9.284	0.009284	6.7510378	8.832	0.008832	6.8230441	16.4	0.0164
CP7	1.0	10.19	0.01019	6.6167021	9.766	0.009766	6.6780165	18	0.018
CP8	0.59	9.207	0.009207	6.7630531	8.732	0.008732	6.8394722	16.4	0.0164
CP9	0.9	9.197	0.009197	6.7646209	8.714	0.008714	6.8424492	16.4	0.0164
CP10	0.79	10.8	0.0108	6.5328249	10.3	0.0103	6.6012119	18	0.018
CP11	0.79	9.907	0.009907	6.657336	9.336	0.009336	6.7429797	18	0.018
CP12	0.55	8.96	0.00896	6.8022856	8.373	0.008373	6.9000397	16.4	0.0164
CP13	0.85	10.16	0.01016	6.6209558	9.713	0.009713	6.6858673	18	0.018
CP14	0.9	8.4	0.0084	6.895395	8	0.008	6.9657843	14.94	0.01494
CP15	0.92	10.3	0.0103	6.6012119	9.919	0.009919	6.6555896	18	0.018
CP16	0.89	10.31	0.01031	6.5998119	9.954	0.009954	6.6505079	18	0.018
CP17	0.95	9.135	0.009135	6.7743796	8.596	0.008596	6.8621188	16.4	0.0164
CP18	1.1	9.191	0.009191	6.7655624	8.704	0.008704	6.8441057	16.4	0.0164
CP19	0.61	7.604	0.007604	7.0390258	7.233	0.007233	7.1111901	13.61	0.01361
CP20	0.68	11.07	0.01107	6.497201	11.18	0.01118	6.482936	18	0.018
CP21	0.95	10.53	0.01053	6.5693508	9.974	0.009974	6.6476121	18	0.018
CP22	0.62	7.455	0.007455	7.0675759	7.039	0.007039	7.1504138	13.61	0.01361
CP23	0.72	8.48	0.00848	6.88172	8.073	0.008073	6.9526794	14.94	0.01494
CP24	0.7	8.409	0.008409	6.89385	8.004	0.008004	6.9650631	14.94	0.01494
CP25	0.65	8.185	0.008185	6.9328019	7.652	0.007652	7.0299474	14.94	0.01494
CP26	0.49	10.37	0.01037	6.5914403	10.07	0.01007	6.6337925	18	0.018
CP27	0.35	7.489	0.007489	7.0610112	7.583	0.007583	7.0430156	12.4	0.0124
CP28	0.28	7.544	0.007544	7.0504546	7.695	0.007695	7.021863	12.4	0.0124
CP29	0.2	7.518	0.007518	7.0554354	7.142	0.007142	7.1294562	13.61	0.01361
CP30	0.38	10.09	0.01009	6.63093	9.592	0.009592	6.7039526	18	0.018
CP31	0.5	8.577	0.008577	6.8653112	8.265	0.008265	6.9187695	14.94	0.01494
CP32	0.62	10.06	0.01006	6.6352259	9.541	0.009541	6.7116438	18	0.018
CP33	0.58	10.04	0.01004	6.6380969	9.521	0.009521	6.7146712	18	0.018
CP34	0.41	8.457	0.008457	6.8856383	8.069	0.008069	6.9533944	14.94	0.01494
CP35	0.48	7.564	0.007564	7.0466349	7.173	0.007173	7.1232077	13.61	0.01361
CP36	0.48	10.15	0.01015	6.6223765	9.688	0.009688	6.6895854	18	0.018
CP37	0.51	9.169	0.009169	6.7690199	8.661	0.008661	6.8512507	16.4	0.0164
CP38	0.5	9.877	0.009877	6.6617114	9.259	0.009259	6.7549279	18	0.018
CP39	0.35	7.471	0.007471	7.0644829	7.04	0.00704	7.1502089	13.61	0.01361

Appendix 4. Particle size analysis data.

Appendix 4. Continued.

Sample No.	Depth (m)	Mean (um)	mm	phi	Median (um)	mm	phi	Mode (um)	mm
CP40	0.3	7.402	0.007402	7.0778691	6.942	0.006942	7.1704329	13.61	0.01361
CP41	0.43	9.003	0.009003	6.7953785	8.388	0.008388	6.8974574	16.4	0.0164
CP42	0.45	9.147	0.009147	6.7724856	8.626	0.008626	6.8570926	16.4	0.0164
CP43	0.3	8.257	0.008257	6.9201666	7.76	0.00776	7.0097276	14.94	0.01494
CP44	0.35	9.996	0.009996	6.6444334	9.435	0.009435	6.7277618	18	0.018
CPS1 c	0.05	6.309	0.006309	7.3083729	5.729	0.005729	7.4475009	11.29	0.01129
CPS1 d	0.2	10.16	0.01016	6.6209558	9.706	0.009706	6.6869074	18	0.018
CPS2 c	0.05	340.6	0.3406	1.5538497	290.5	0.2905	1.7833899	356.1	0.3561
CPS2 d	0.2	9.814	0.009814	6.670943	9.121	0.009121	6.7765923	18	0.018
CPS3 c	0.05	490.5	0.4905	1.027675	381.2	0.3812	1.39138	429.2	0.4292
CPS3 d	0.2	7.235	0.007235	7.1107913	7.128	0.007128	7.1322869	12.4	0.0124

Appendix 4. Continued.

Sample No.	Depth (m)	phi	SD (um)	mm	phi	Skewness		Kurtosis	
CP1	0.48	5.9301604	5.577	0.005577	7.486295	0.218	right	-1.02	Platykurtic
CP2	0.37	6.3335161	4.282	0.004282	7.8674995	0.082	right	-1.13	Platykurtic
CP3	0.36	6.3760208	4.297	0.004297	7.8624545	0.08	right	-1.139	Platykurtic
CP4	0.57	6.1991891	4.621	0.004621	7.7575792	0.202	right	-1.007	Platykurtic
CP5	0.96	5.9301604	5.593	0.005593	7.482162	0.214	right	-1.027	Platykurtic
CP6	1.0	5.9301604	5.585	0.005585	7.484227	0.225	right	-1.023	Platykurtic
CP7	1.0	5.7958593	6.117	0.006117	7.35296	0.196	right	-1.049	Platykurtic
CP8	0.59	5.9301604	5.566	0.005566	7.4891434	0.239	right	-1.007	Platykurtic
CP9	0.9	5.9301604	5.518	0.005518	7.5016388	0.246	right	-0.985	Platykurtic
CP10	0.79	5.7958593	6.46	0.00646	7.2742501	0.26	right	-0.91	Platykurtic
CP11	0.79	5.7958593	6.007	0.006007	7.3791396	0.262	right	-0.983	Platykurtic
CP12	0.55	5.9301604	5.566	0.005566	7.4891434	0.29	right	-0.983	Platykurtic
CP13	0.85	5.7958593	6.098	0.006098	7.3574481	0.204	right	-1.04	Platykurtic
CP14	0.9	6.064676	5.068	0.005068	7.6243678	0.238	right	-0.991	Platykurtic
CP15	0.92	5.7958593	6.148	0.006148	7.3456671	0.173	right	-1.081	Platykurtic
CP16	0.89	5.7958593	6.092	0.006092	7.3588683	0.168	right	-1.056	Platykurtic
CP17	0.95	5.9301604	5.564	0.005564	7.4896619	0.263	right	-1.001	Platykurtic
CP18	1.1	5.9301604	5.502	0.005502	7.5058281	0.25	right	-0.977	Platykurtic
CP19	0.61	6.1991891	4.592	0.004592	7.7666616	0.254	right	-0.98	Platykurtic
CP20	0.68	5.7958593	6.249	0.006249	7.3221589	0.011	right	-1.138	Platykurtic
CP21	0.95	5.7958593	6.128	0.006128	7.350368	0.171	right	-1.078	Platykurtic
CP22	0.62	6.1991891	4.514	0.004514	7.7913779	20.37	right	-0.919	Platykurtic
CP23	0.72	6.064676	5.073	0.005073	7.6229451	0.227	right	-0.998	Platykurtic
CP24	0.7	6.064676	5.092	0.005092	7.6175519	0.237	right	-1.005	Platykurtic
CP25	0.65	6.064676	5.059	0.005059	7.626932	0.3	right	-0.963	Platykurtic
CP26	0.49	5.7958593	6.277	0.006277	7.3157091	0.147	right	-1.123	Platykurtic
CP27	0.35	6.3335161	4.357	0.004357	7.8424492	0.037	right	-1.171	Platykurtic
CP28	0.28	6.3335161	4.382	0.004382	7.8341948	0.015	right	-1.183	Platykurtic
CP29	0.2	6.1991891	4.701	0.004701	7.7328166	0.255	right	-1.03	Platykurtic
CP30	0.38	5.7958593	6.191	0.006191	7.3356118	0.215	right	-1.073	Platykurtic
CP31	0.5	6.064676	5.152	0.005152	7.6006517	0.19	right	-1.046	Platykurtic
CP32	0.62	5.7958593	6.102	0.006102	7.3565021	0.228	right	-1.034	Platykurtic
CP33	0.58	5.7958593	6.1	0.0061	7.356975	0.231	right	-1.033	Platykurtic
CP34	0.41	6.064676	5.087	0.005087	7.6189692	0.228	right	-1.007	Platykurtic
CP35	0.48	6.1991891	4.576	0.004576	7.7716972	0.266	right	-0.967	Platykurtic
CP36	0.48	5.7958593	6.106	0.006106	7.3555567	0.21	right	-1.044	Platykurtic
CP37	0.51	5.9301604	5.572	0.005572	7.487589	0.251	right	-1.0006	Platykurtic
CP38	0.5	5.7958593	6.091	0.006091	7.3591052	0.268	right	-1.017	Platykurtic
CP39	0.35	6.1991891	4.59	0.00459	7.7672901	0.289	right	-0.964	Platykurtic

Appendix 4. Continued.

Sample No.	Depth (m)	phi	SD (um)	mm	phi	Skewness		Kurtosis	
CP40	0.3	6.1991891	4.554	0.004554	7.77865	0.31	right	-0.935	Platykurtic
CP41	0.43	5.9301604	5.598	0.005598	7.4808728	0.291	right	-1.002	Platykurtic
CP42	0.45	5.9301604	5.563	0.005563	7.4899212	0.257	right	-1	Platykurtic
CP43	0.3	6.064676	5.059	0.005059	7.626932	0.283	right	-0.947	Platykurtic
CP44	0.35	5.7958593	6.151	0.006151	7.3449633	0.24	right	-1.053	Platykurtic
CPS1 c	0.05	6.4688107	4.04	0.00404	7.951429	0.423	right	-0.849	Platykurtic
CPS1 d	0.2	5.7958593	6.196	0.006196	7.3344471	0.199	right	-1.083	Platykurtic
CPS2 c	0.05	1.4896457	274.3	0.2743	1.8661735	1.37	right	2.605	Leptokerttic
CPS2 d	0.2	5.7958593	6.164	0.006164	7.3419174	0.276	right	-1.053	Platykurtic
CPS3 c	0.05	1.220278	429.5	0.4295	1.21927	1.039	right	0.44	Leptokerttic
CPS3 d	0.2	6.3335161	4.266	0.004266	7.8729003	0.126	right	-1.116	Platykurtic

8. Conclusions

Based on the research presented here, it appears that sampling bias maybe hindering the research trying to link thecamoebians with their environmental control. The taxonomy as discussed in the introduction is no doubt problematic and requires more research to refine it. However, our results show that high density sampling with site specific environmental measurements may provide more success in the future. It is surprising that the trends with water depth have not been reported previously, and at this stage their reproducibility in other wetlands is unknown. However, similar studies with higher density sampling and over shallow water depths may provide more data that may allow thecamoebians to be used as a water-level indicator in wetlands paralleling the success of foraminifera in marine marshes (Scott et al., 2001).