



Testate amoebae as paleoindicators of surface-moisture changes on Michigan peatlands: modern ecology and hydrological calibration

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Abstract

Peatland testate amoebae are sensitive indicators of local hydrology and have been used as proxies for surface moisture conditions in fossil studies. However, few regional calibration datasets exist in North America, and knowledge of testate amoeba ecology and distribution patterns are limited. The objectives of this study were to (1) investigate the relationship between testate amoebae, environment, and *Sphagnum* species in Michigan peatlands; (2) generate transfer functions from this dataset that can be applied to fossil data; and (3) describe vertical variation of testate amoebae inhabiting *Sphagnum* moss. Testate amoeba assemblages from 139 microsites within 11 peatlands in Michigan were compared to assess variability between and within peatlands. Most peatlands contained similar testate amoeba assemblages, although within individual peatlands the amount of assemblage variability is correlated to the amount of environmental heterogeneity. Of the measured environmental variables, depth to water table showed the strongest relationship with testate amoebae. Depth to water table can be reconstructed from fossil data with a mean error of ± 7.5 cm, although predictive ability deteriorates in extremely dry environments (>30 cm water table depth). Vertical variation in testate amoeba assemblages was investigated at 89 *Sphagnum*-dominated microsites by directly comparing amoeba assemblages and the abundance and frequency of common taxa in upper and lower portions of the *Sphagnum* stem. Except for extremely dry microsites, considerable vertical variation in assemblage composition exists. Many agglutinate taxa are more abundant on lower portions of the *Sphagnum* stem, and taxa containing symbiotic zoochlorellae are more abundant on upper portions. Refinements in sampling procedures and analysis may improve the predictive ability of transfer functions.

Introduction

Peatland stratigraphy, particularly the stratigraphy of bogs, has been used as a source of paleoclimate data for over a century. Early paleoclimate work on peatlands was focused on documenting times of high-magnitude hydrological change, typically at millennial timescales (e.g., Sernander (1908)). Concerns about the integrity of the stratigraphic record, the extent of autogenic influences (i.e., non-climatic), and a now-refuted paradigm of bog development (Barber 1981; Backeus 1990), all hampered investigations of lower magnitude and higher frequency climatic changes until the late 1970's (e.g., Aaby (1976)). Although the relative importance of autogenic and

allogenic factors in controlling stratigraphic changes in different peatland types and at differing stages of peatland development has not been completely resolved (e.g., Tolonen et al. (1985), Foster and Wright (1990), Almquist-Jacobson and Foster (1995), Hu and Davis (1995), Hendon et al. (2001)), a growing body of evidence documents high-frequency, low-magnitude climatic changes from bog stratigraphy (e.g., Charman et al. (1999), Mauquoy and Barber (1999), Barber et al. (2000), Hendon et al. (2001)). A few studies also suggest that sensitive paleoclimate records may even be obtained from peatlands that are not truly ombrotrophic (Anderson 1998; Hendon et al. 2001), although more research is needed to assess the climatic sensitivity of these systems. During the past

decade techniques have been developed and refined for quantitative inference of past climatic changes using a variety of climate proxies, including plant macrofossils (e.g., Barber et al. (2000)), testate amoebae (e.g., Woodland et al. (1998)), and measurements of humification (Blackford and Chambers 1993). However, especially in the case of testate amoeba analysis, further development of biological proxies requires a better knowledge of species-environment relationships, and the collection of calibration datasets from a variety of geographic regions and peatland types.

Testate amoebae (Protozoa: Rhizopoda) inhabit moist soils, wetlands, and lakes. Fossil tests can usually be identified to species and recovered from sediments in quantities sufficiently large to permit estimation of relative abundance (Warner 1990). The dominant control on taxa distribution patterns in wetland habitats is substrate moisture (Tolonen 1986; Warner 1987, 1990), although trophic status (Tolonen et al. 1992), pH (e.g., Charman and Warner (1992), Mitchell et al. (1999), Booth (2001)), and other aspects of water chemistry (Woodland et al. 1998; Mitchell et al. 2000a) are also important factors. Many taxa are cosmopolitan in distribution, although comparisons of species' hydrological preferences are difficult because most studies have only measured hydrological data at a single time during the year. However, qualitative comparisons of the hydrologic preferences of taxa along relative moisture gradients in North America, Europe, and New Zealand suggest that many have similar preferences among locations (Charman et al. 2000; Booth 2001). Transfer functions to infer moisture conditions from testate amoeba assemblages have been developed from regions in North America (Charman and Warner 1992, 1997; Warner and Charman 1994; Booth 2001), Europe (Tolonen et al. 1992; Woodland et al. 1998; Mitchell et al. 1999), Russia (Bobrov et al. 1999), and New Zealand (Charman 1997). Transfer functions based on testate amoebae have been used to infer centennial-scale climate patterns in England for the last 4500 years (Charman and Hendon 2000; Hendon et al. 2001), although they have not yet been extensively used for high-resolution paleoclimate reconstructions in other regions.

Testate amoebae are especially abundant and diverse in *Sphagnum*-dominated habitats. Modern testate amoeba samples are usually collected from the brown *Sphagnum* directly below the green portion of the stems, because it has been suggested that samples

from this horizon best represent what would be found in the fossil record (Warner 1987; Tolonen et al. 1992). Stratification of testate amoeba taxa has been observed vertically along the *Sphagnum* stem, and this spatial variation has been attributed primarily to gradients of light, temperature, food, and oxygen (Heal 1962; Meisterfeld 1977). Vertical variation along the *Sphagnum* stem is also a gradient from mostly alive testate amoebae in the aerobic, upper portions and mostly empty tests in the more anaerobic, lower portions (Schönborn 1963). However, quantitative comparisons between amoeba assemblages found on the upper, chlorophyllous portion of the *Sphagnum* stem and those characterizing the lower, non-chlorophyllous portion of the *Sphagnum* stem are needed. These comparisons can provide insights into the taphonomy and ecology of testate amoebae, and may guide the collection of more representative calibration datasets. In this paper, I refer to the testate amoeba assemblages found on the upper and lower portions of the stem as upper and lower assemblages.

In this study I investigate the relationship among testate amoebae, vegetation, pH, water table depth, substrate percent moisture, depth of the living moss, and bulk density from 139 samples collected from 11 peatlands in Michigan, USA. I investigate controls on testate amoeba distribution between and within these peatlands, and develop transfer functions from this dataset to infer water table depth from fossil data. I also compare testate amoeba assemblages inhabiting upper and lower portions of the *Sphagnum* stem at 89 microsites within eight of these peatlands. I attempt to quantify and describe differences in these assemblages and develop hypotheses to explain these differences.

Study sites

Eleven peatlands in Michigan were sampled for modern testate amoebae (Figure 1). The peatlands span the entire gradient from ombrotrophic to minerotrophic, and the amount of environmental variation within each peatland varies considerably (Table 1). However, because fossil records from oligotrophic, *Sphagnum*-dominated peatlands (i.e., poor fens and bogs) would be expected to yield more climatically sensitive records than those from strongly minerotrophic peatlands, I focused primarily on *Sphagnum*-dominated sites.

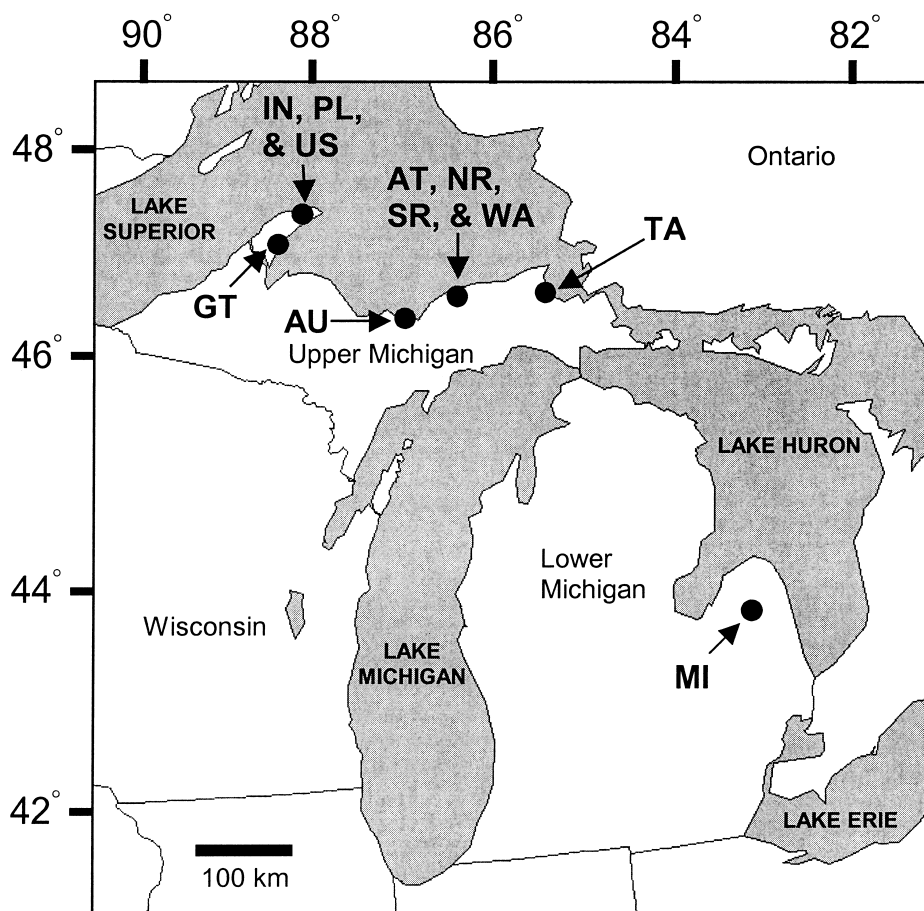


Figure 1. Location of the studied peatlands. Abbreviations for the peatlands are as follows: IN – Independence; PL – Partridge Lake; US – Uncle Sam; GT – Grand Traverse; AU – Au Train; AT – Adams Trail; NR – North Rhody; SR – South Rhody; WA – West Adams Trail; TA – Tahquamenon; MI – Minden. Specific locations and characteristics of each peatland are shown in Table 1.

Methods

Field methods

Field sampling at all sites was conducted in a 4-week period from late June until late July 2000. The sampling procedure was similar to previous investigations (Charman and Warner 1992, 1997; Warner and Charman 1994; Mitchell et al. 1999; Booth 2001). Microsites within each peatland were sampled in an attempt to represent the full range of surface-moisture variability (e.g., hummocks, hollows, pools) of each peatland. At each microsite, a peat sample was collected by pushing a cylinder (7.5-cm diameter, 11-cm length) with one open end into the peat until it was level with the surface of the wetland. A knife was used to cut the peat around the cylinder as the can was pushed into the peat. A small hole in the closed end of

the cylinder allowed air to escape and prevented peat compaction. The peat was held in the cylinder with one hand and pulled up carefully. Peat sticking out of the bottom of the cylinder was cut off, and the cylinder was sealed. These peat samples were used to calculate percent moisture and bulk density. From the edge of the hole left by the peat sample, a $\sim 10\text{-cm}^3$ sample was collected from the brown *Sphagnum* directly below the green portion of the stems. The entire green portion of the *Sphagnum* directly above this lower sample was also collected at 89 microsites. Samples from this upper portion of the moss ranged from $\sim 10\text{ cm}^3$ to $\sim 30\text{ cm}^3$ and represent the upper assemblage. *Sphagnum* from around the hole was also collected for species-level identification. Vascular plant species growing within 50 cm of each microsite were also recorded. At microsites lacking *Sphagnum*, a $\sim 10\text{-cm}^3$ sample of peat was collected from the

Table 1. Site characteristics, including the range of environmental variables measured (%M – percent moisture; WTD – water table depth; BD – bulk density), number of testate amoeba microsites sampled (n), *Sphagnum* spp. present, and a general description of each peatland. Negative values of water table depth refer to sites with standing water. Dominant vascular plant species include those that had a percent cover of greater than 20% within 50 cm of a testate amoeba sample.

Site name and location	pH	%M	WTD (cm)	BD (g/cm ³)	n	<i>Sphagnum</i> spp.	General description and dominant vascular plant species
Adams Trail Bog (AT) (46° 32'52"N, 86° 3'32"W)	3.7–4.0	72.5–87.8	12 to –2	0.13–0.15	10	<i>S. papillosum</i> , <i>S. magellanicum</i> , <i>S. capillifolium</i> , <i>S. angustifolium</i> , <i>S. rubellum</i> , <i>S. alba</i>	Open, poor fen dominated by <i>Carex oligosperma</i> , <i>Carex</i> spp., <i>Scheuchzeria palustris</i> , and <i>Rhynchospora alba</i>
Au Train Bay (AU) (46° 25'48"N, 86° 51'36"W)	3.5–6.4	64.1–86.3	29 to –27	0.13–0.17	22	<i>viridum</i> , <i>S. isoviitae</i> , <i>S. papillosum</i> , <i>S. fallax</i> , <i>S. magellanicum</i> , <i>S. majus</i> ssp. <i>norvegicum</i> , <i>S. angustifolium</i> , <i>S. pulchrum</i>	Microsites ranging from rich to poor fens. Dominant plant species included <i>Carex lasiocarpa</i> , <i>Carex oligosperma</i> , <i>Menyanthes trifoliata</i> , <i>Rhynchospora alba</i> , <i>Scheuchzeria palustris</i> , <i>Chamaedaphne calyculata</i> , <i>Myrica gale</i> , <i>Vaccinium oxycoccos</i> , and <i>Typha latifolia</i>
Grand Traverse Bay (GT) (47° 9'56"N, 88° 15'17"W)	3.7–5.2	40.0–80.8	45 to 5	0.13–0.52	8	<i>S. pulchrum</i> , <i>S. papillosum</i> , <i>S. fallax</i> , <i>S. angustifolium</i> , <i>S. majus</i> ssp. <i>majus</i>	Microsites ranging from moderate to poor fen. Dominant plant species include <i>Carex oligosperma</i> , <i>Chamaedaphne calyculata</i> , and <i>Andromeda glaucophylla</i>
Independence Bog (IN) (47° 25'9"N, 88° 0'37"W)	3.7–4.4	56.6–85.9	36 to –14	0.12–0.22	29	<i>S. viridum</i> , <i>S. majus</i> ssp. <i>norvegicum</i> , <i>S. magellanicum</i> , <i>S. fuscum</i> , <i>S. papillosum</i> , <i>S. angustifolium</i> , <i>S. majus</i> ssp. <i>majus</i>	Microsites ranging from moderate to poor fen, with scattered hummocks containing <i>Picea mariana</i> , <i>Larix laricina</i> , and <i>Chamaedaphne calyculata</i> . Hollows and pools characterized by <i>Nymphaea odorata</i> , <i>Menyanthes trifoliata</i> , <i>Scheuchzeria palustris</i> , <i>Rhynchospora alba</i> , and <i>Rhynchospora lasca</i>
Minden Bog (MI) (43° 36'43"N, 82° 50'10"W)	3.4–3.6	38.5–78.9	48 to 12	0.13–0.23	14	<i>S. palustre</i> , <i>S. russowii</i> , <i>S. papillosum</i> , <i>S. isoviitae</i> , <i>S. fallax</i> , <i>S. magellanicum</i> , <i>S. bartlettianum</i> , <i>S. angustifolium</i> , <i>S. capillifolium</i>	Raised bog with scattered <i>Picea mariana</i> . <i>Kalmia polifolia</i> and <i>Chamaedaphne calyculata</i> are dominant.
North Rhody Bog (NR) (46° 34'57"N, 86° 4'20"W)	3.8–4.3	60.7–76.3	27 to 13	0.13–0.18	12	<i>S. papillosum</i> , <i>S. magellanicum</i> , <i>S. angustifolium</i>	Open, poor fen with <i>Chamaedaphne calyculata</i> , <i>Carex oligosperma</i> , and <i>Carex limosa</i>
Partridge Lake Bog (PL) (47° 25'11"N, 88° 0'27"W)	3.5–5.4	63.3–85.0	26 to –10	0.11–0.17	20	<i>S. angustifolium</i> , <i>S. magellanicum</i> , <i>S. fuscum</i> , <i>S. isoviitae</i> , <i>S. russowii</i> , <i>S. majus</i> ssp.	Rich to poor fen (floating mat) with <i>Chamaedaphne calyculata</i> , <i>Rhynchospora alba</i> , <i>Vaccinium oxycoccos</i> , <i>Ledum groenlandicum</i> , and <i>Scheuchzeria palustris</i>
South Rhody Bog (SR) (46° 33'57"N, 86° 4'30"W)	3.7–4.2	76.3–82.0	25 to 2	0.14–0.18	16	<i>S. magellanicum</i> , <i>S. majus</i> ssp. <i>majus</i> , <i>S. cuspidatum</i> , <i>S. viridum</i> , <i>S. fallax</i> , <i>S. angustifolium</i> , <i>S. papillosum</i> , <i>S. majus</i> ssp. <i>norvegicum</i>	Open, poor fen with <i>Carex oligosperma</i> , <i>Carex limosa</i> , <i>Chamaedaphne calyculata</i> , and <i>Vaccinium oxycoccos</i>
Tahquamenon Bay (TA) (46° 28'31"N, 84° 59'41"W)	4.2–5.1	67.3–78.7	50 to 9	0.16–0.17	4	<i>S. centrale</i> , <i>S. papillosum</i> , <i>S. magellanicum</i> , <i>S. fallax</i> , <i>S. russowii</i>	Poor to moderately rich fen, with some microsites open and others forested. Dominant species include <i>Carex trispirma</i> , <i>Smilacina racemosa</i> , and other <i>Carex</i> spp.
Uncle Sam Bog (US) (47° 25'9"N, 88° 0'59"W)	3.3–3.7	50.5–62.6	50 to 48	0.15–0.17	8	<i>S. magellanicum</i> , <i>S. angustifolium</i> , <i>S. russowii</i> , <i>S. girgensohnii</i>	Forested bog dominated by <i>Picea mariana</i> . Some <i>Pinus strobus</i> , <i>Larix laricina</i> , and <i>Betula alleghaniensis</i> . Understorey dominated by <i>Ledum groenlandicum</i> , <i>Vaccinium myrtilloides</i> , and <i>Chamaedaphne calyculata</i>
West Adams Trail Bog (WA) (46° 33'12"N, 86° 6'17"W)	3.9–3.9	74.4–80.1	19 to 19	0.16–0.18	4	<i>S. magellanicum</i> , <i>S. fallax</i> , <i>S. papillosum</i> , <i>S. angustifolium</i>	Open, poor fen with <i>Chamaedaphne calyculata</i> , <i>Andromeda glaucophylla</i> , and <i>Carex</i> spp.

surface of the peatland. The depth to the water table and pH were measured at each microsite. pH was measured as close to the amoeba sample as possible, typically by squeezing the water from the adjacent *Sphagnum* or peat. At sites with standing water, depth to water table was recorded as a negative value (i.e., distance from the peat surface to the water surface).

Dataset descriptions

The calibration dataset consists of all the lower assemblages and the samples collected from substrates other than *Sphagnum*. These samples are comparable to datasets collected for the development of transfer functions in other studies (e.g., Charman and Warner (1992, 1997), Charman (1997), Warner and Charman (1994), Woodland et al. (1998), Mitchell et al. (1999), Booth (2001)). The upper and lower assemblage dataset consists of pairs of upper and lower assemblages, with each pair corresponding to one microsite.

Laboratory methods

Testate amoebae were isolated from peat using the method of Hendon and Charman (1997). Samples were boiled in distilled water for 10 min and washed through 355 and 15- μm sieves. The material caught in the 15- μm sieve was stained with two drops of safranin and stored in glycerol. Slides were prepared, and all testate amoebae were identified and counted until a total of at least 150 was reached. Taxonomy follows Charman et al. (2000), except as indicated in Table 2. The relative abundance of each taxon was calculated as a percent of the total number of testate amoebae counted. *Habrotricha angusticollis*, a commonly fossilized rotifer (Warner 1988), was included in the analysis and count total.

Analytical techniques

Outlier analysis was performed with the software PC-ORD (McCune and Mefford 1999). Five samples from the calibration dataset had species or environmental data more than 2 S.D. from the mean and were identified as outliers. These samples were removed from further analysis. The resulting calibration dataset included 139 assemblages, of which 89 samples collected from *Sphagnum* had associated upper assemblages. Taxa encountered in less than five samples (*Cryptodiffugia sacculus* Penard, *Lesquereusia modesta* (Ehrenberg) Bütschli, *Nebela barbata* Leidy,

Nebela vitraea Penard, *Nebela flabellulum* Leidy, *Nebela tubulosa* Penard, *Quadruella symmetrica* (Wallich) Schulze) were eliminated from the dataset prior to the calculation of relative abundances. Taxa included in the analyses, including taxonomic authorities, are listed in Table 2. *Sphagnum* species present at less than three sites (*S. bartlettianum* Warnstorf, *S. centrale* C. Jensen, *S. cuspidatum* Hoffman, *S. fuscum* (Schimp.) Klinggraff, *S. girgensohnii* Russov, *S. palustre* Linnaeus, *S. pulchrum* (Braithw.) Warnstorf) were not included in the analyses.

Direct and indirect gradient analysis techniques were used to investigate relationships among sites, environment, *Sphagnum* species, and testate amoebae in the calibration dataset. Four models were used to develop transfer functions using the program CALIBRATE (Juggins 1998). These models included weighted averaging (WA), tolerance downweighted averaging (WA-Tol), partial least squares (PLS), and weighted averaging partial least squares (WA-PLS) (Birks 1995, 1998). These models were chosen because they have been used successfully to develop transfer functions from testate amoeba data in other studies (e.g., Woodland et al. (1998), Charman (1997), Charman and Warner (1992), Booth (2001)).

Indirect and direct gradient analysis techniques were also used to investigate relationships among environment, *Sphagnum* species, and upper and lower assemblages. A community coefficient was also calculated between each pair of upper and lower assemblages using Sørensen's index of taxonomic similarity and this index of similarity was compared to environmental variables using linear correlation. Species richness, and the abundance and frequency of common taxa (those present in at least 25 upper-lower sample pairs), were compared between upper and lower assemblages. Regression was used to compare the abundance of each taxon in upper and lower assemblages. Chi-Square tests and odds ratios were used to assess differences in the frequency of taxa in the two groups.

Results and discussion

Variability among sites in the calibration dataset

Detrended correspondence analysis (DCA) was used to investigate the similarity of the 139 assemblages at the different study sites. Samples from most sites overlap a great deal in ordination space (Figure 2).

Table 2. Testate amoeba taxa encountered in this study and abbreviations used on figures. Taxonomy follows Charman et al. (2000) except where noted by asterisks.

Taxon	Abbreviation
<i>Amphitrema flavum</i> (Archer) Penard	amp fla
<i>Amphitrema wrightianum</i> Archer	amp wri
<i>Arcella artocrea</i> Leidy type ¹	arc art
<i>Arcella crenulata</i> Deflandre** ²	arc cre
<i>Arcella discoides</i> Ehrenberg type	arc dis
<i>Arcella hemisphaerica</i> Perty	arc hem
<i>Arcella vulgaris</i> Ehrenberg	arc vul
<i>Assulina muscorum</i> Greeff	ass mus
<i>Assulina seminulum</i> (Ehrenberg) Leidy	ass sem
<i>Bullinularia indica</i> Penard	bul ind
<i>Centropyxis platystoma</i> (Penard) Deflandre type	cen pla
<i>Centropyxis cassis</i> (Wallich) Deflandre type	cen cas
<i>Centropyxis ecornis</i> Ehrenberg type** ³	cen eco
<i>Centropyxis aculeata</i> (Ehrenberg) von Stein type	cen acu
<i>Corythion-Trinema</i> type	cor tri
<i>Cyclopyxis arcelloides</i> (Penard) Deflandre type ⁴	cyc arc
<i>Diffflugia acuminata</i> Ehrenberg type	dif acu
<i>Diffflugia bacillifera</i> Penard	dif bac
<i>Diffflugia globulosa</i> Dujardin type ⁴	dif glo
<i>Diffflugia leidyi</i> Wailes	dif lei
<i>Diffflugia oblonga</i> Ehrenberg type	dif obl
<i>Diffflugia oviformis</i> Cash type** ⁵	dif ovi
<i>Diffflugia pulex</i> Penard	dif pul
<i>Diffflugia urceolata</i> Carter*	dif urc
<i>Euglypha rotunda</i> Wailes and Penard type	eug rot
<i>Euglypha strigosa</i> (Ehrenberg) Leidy type	eug str
<i>Euglypha tuberculata</i> Dujardin type	eug tub
<i>Habrotricha angusticollis</i> Murray***	hab ang
<i>Heleopera petricola</i> Leidy	hel pet
<i>Heleopera rosea</i> Penard	hel ros
<i>Heleopera sphagni</i> (Leidy) Cash and Hopkinson	hel sph
<i>Heleopera sylvatica</i> Penard	hel syl
<i>Hyalosphenia elegans</i> Leidy	hya ele
<i>Hyalosphenia papilio</i> Leidy	pap
<i>Hyalosphenia subflava</i> Cash and Hopkinson	hya sub
<i>Nebela carinata</i> (Archer) Leidy	neb car
<i>Nebela collaris</i> (Ehrenberg) Leidy	neb col
<i>Nebela galeata</i> Penard*	neb gal
<i>Nebela griseola</i> Penard	neb gri
<i>Nebela marginata</i> Penard	neb mar
<i>Nebela militaris</i> Penard	neb mil
<i>Nebela parvula</i> Cash and Hopkinson	neb par
<i>Nebela penardiana</i> Deflandre*	neb pen
<i>Nebela tincta</i> (Leidy) Awerintzew	neb tin
<i>Placocista spinosa</i> (Carter) Leidy	pla spi
<i>Phryganella acropodia</i> (Hertwig & Lesser) type ⁴	phy acr
<i>Pontigulasia</i> sp.**	pon sp.
<i>Pseudodiffflugia fascicularis</i> Penard	pse fas
<i>Pyxidicula operculata</i> (Agardh) Ehrenberg****	pyx ope
<i>Sphenoderia lenta</i> Schlumberger	sph len
<i>Tracheuglypha dentata</i> Moniez*	tra den
<i>Trigonopyxis arcula</i> (Leidy) Penard	tri arc
<i>Trigonopyxis minuta</i> Schönborn and Peschke	tri min

*Taxonomy follows Ogden and Hedley (1980), Corbet (1973). **Taxonomy follows Ogden and Hedley (1980). ***Taxonomy follows Warner (1988). ****Taxonomy follows Grospietsch (1958), Meisterfeld (2000). ¹Probably includes *Arcella catinus* and *Arcella artocrea*.

²Differentiated from *Arcella vulgaris* by its denticular aperture margin. ³Differentiated from *Centropyxis cassis* type and *Centropyxis platystoma* type by its circular to sub-circular outline, circular and more medially positioned aperture, and large test diameter (> 150 µm).

⁴Like *Cyclopyxis arcelloides* type *sensu* Charman et al. (2000). However, three morphotypes were differentiated in this study: *Cyclopyxis arcelloides* type (test diameter < 70 µm with aperture diameter more than three-quarters the diameter of the test), *Diffflugia globulosa* type (test diameter > 70 µm with aperture diameter usually more than half the test), *Phryganella acropodia* type (test diameter < 70 µm with aperture diameter less than three-quarters of the test, often with fungal hyphae incorporated into the test). ⁵Specimens with 4–5 lobes surrounding the aperture.

Even the most geographically separate peatlands, Minden Bog and Uncle Sam Bog, are similar to each other in testate amoeba composition. Minden Bog and Uncle Sam Bog were also the driest of the eleven peatlands studied (Table 1). Testate amoeba assemblages are also similar to those found at other sites in North America (Charman and Warner 1992, 1997; Warner and Charman 1994). The similarity in testate amoeba assemblage composition between the sites from this study, and to other sites in North America is not surprising given the cosmopolitan distribution of many taxa. The high degree of between-site and between-region similarity also suggests that this dataset could aid interpretation of fossil data not only from Michigan, but throughout the western Great Lakes region and perhaps other regions of North America.

Although many of the sites overlap in ordination space, the amount of assemblage variation represented by the samples of each site (i.e., the amount of multivariate space occupied by each site) varies considerably. This assemblage variation is attributable to the amount of environmental variation sampled in each peatland. For example, direct comparison of a measure of testate amoeba assemblage variability (i.e., the range of DCA axis 1 scores of each peatland) and the range of water table depths measured at each

peatland indicates that sites with highly variable environmental conditions show more variability in testate amoeba assemblages (Figure 3). Minden Bog appears to be somewhat of an exception, as the assemblage variation is relatively low (i.e., small range of DCA axis 1 scores) but the range of water depths is large (Figure 3). Minden Bog is the only truly ombrotrophic peatland in this study, and is therefore fundamentally different hydrologically than the other sites.

Relationships among testate amoebae, environment, and Sphagnum species in the calibration dataset

Sphagnum species were distributed primarily along environmental gradients of moisture and pH (Figure 4), and the species are generally organized along these gradients according to their known ecology (Andrus 1980, 1986), although exceptions exist. For example, *Sphagnum russowii* was only encountered at dry and relatively acid microsites, although in other regions it is more minerotrophic (Andrus 1980). Sites without *Sphagnum* were at the extreme end of the pH and moisture gradient, corresponding to high pH and very wet conditions.

Substrate moisture has been identified as the dominant control on the distribution of testate amoeba taxa

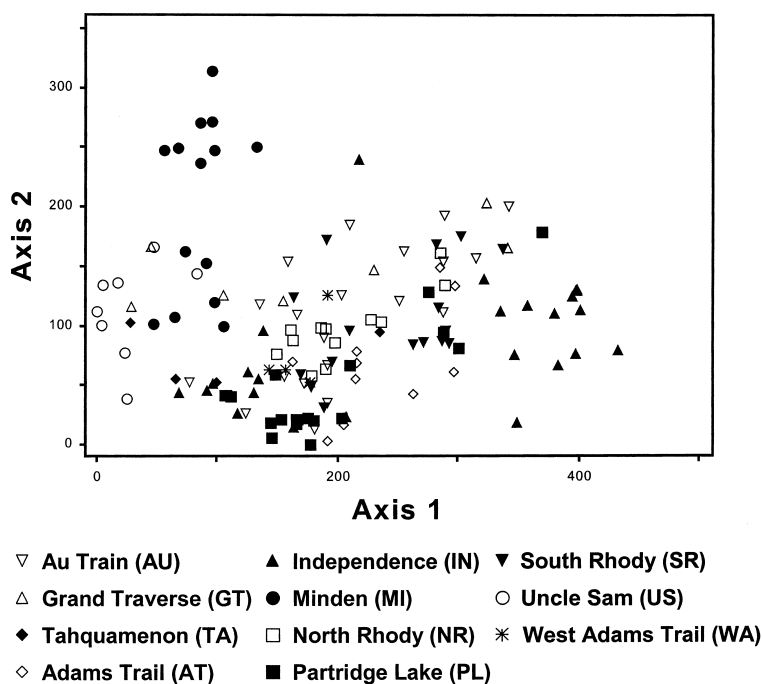


Figure 2. DCA of the 139 samples in the calibration dataset. Samples are coded by site.

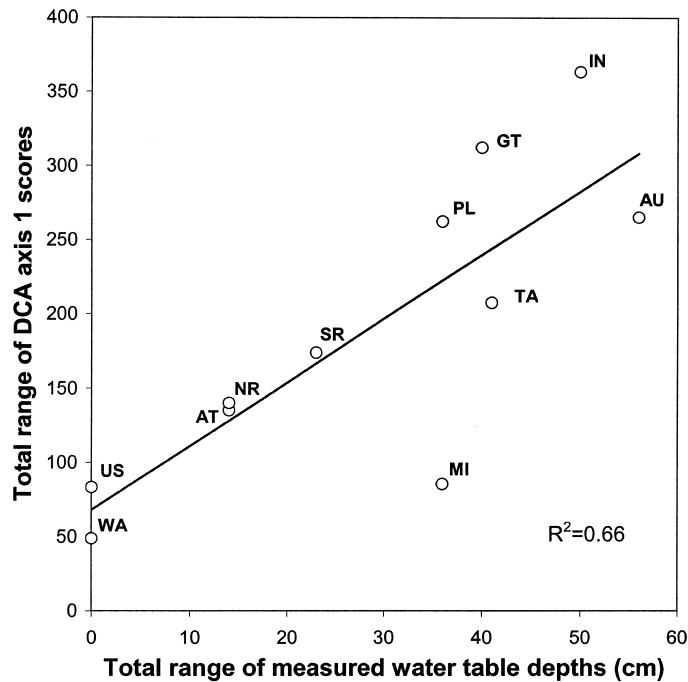


Figure 3. Total range of DCA axis 1 sample scores for each site (a measure of amoeba assemblage variation) plotted against the total range of water table depths for each site. Abbreviations for sites are the same as Figure 1.

in peatlands throughout the world (e.g., Tolonen (1986), Warner (1987, 1990), Charman and Warner (1992), Mitchell et al. (1999, 2000a), Woodland et al. (1998)). CCA shows that the composition of testate amoeba assemblages in this study is also primarily controlled by substrate moisture conditions (Figure 5, Table 3). Most peatland species inhabit the water film on *Sphagnum* mosses, and CCA indicates that testate amoeba taxa and *Sphagnum* species are arranged along similar hydrological gradients (Figure 5). Testate amoeba taxa and *Sphagnum* species abundant in drier, and generally more acidic conditions, are positioned on the right side of axis 1, while those taxa abundant in wetter, generally higher pH habitats, are to the left (Figure 5). Microsites without *Sphagnum* occur on the far left side of axis 1 (Figure 5); these represent the wettest and highest pH habitats sampled (Figure 4).

Although the variance explained by the CCA is relatively low (Table 3), it is an improvement over the only other study of testate amoeba ecology in Michigan (Booth 2001) and is similar to studies from other regions (e.g., Charman (1997), Woodland et al. (1998)). Several potential explanations exist for the low amount of variance explained. Spatial and temporal scale differences certainly exist between the en-

vironment as experienced by the testate amoebae and the environmental parameters I measured. For example, horizontal and vertical variation in amoeba assemblages is known to occur on scales ranging from centimeters or smaller (Heal 1962; Schönborn 1963; Meisterfeld 1977; Mitchell et al. 2000b) and the environmental measurements are at a much coarser scale. Even in relatively small areas (0.25 m²) that look homogeneous from a macroscopic perspective, testate amoeba assemblages are spatially heterogeneous (Mitchell et al. 2000b). Also, environmental variables were only measured on the day of sample collection and do not take seasonal fluctuations into account, and testate amoebae certainly respond to these temporal fluctuations (Heal 1964). Seasonal variability has not been a major problem in other studies (e.g., Warner (1987), Charman and Warner (1992), Tolonen et al. (1992), Charman (1997)), apparently because species and environmental data have been collected during one field season and thus the measurements fall along relative environmental gradients. However, improvements can be made using annual hydrologic data (Woodland et al. 1998).

The amoeba assemblages inhabiting the water film of *Sphagnum* moss are quite different from those inhabiting sites without *Sphagnum*, and pH may be

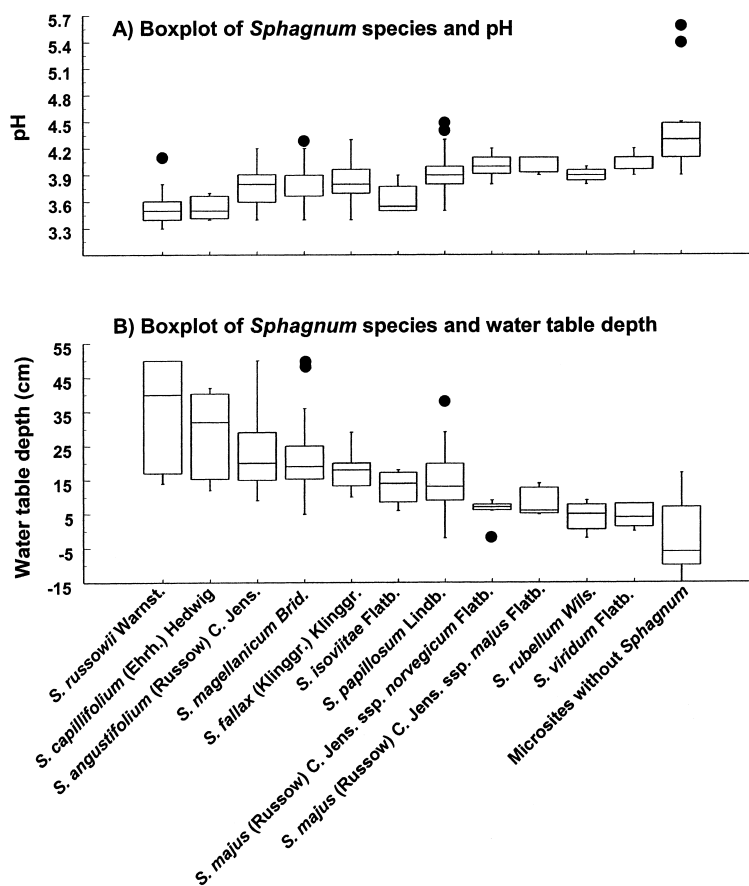


Figure 4. Boxplots showing the relationship of *Sphagnum* species to (A) pH and (B) water table depth, as measured in this study. *Sphagnum* species are organized along the x-axis according to increasing preferences for wetter habitats. The boxplots show the median, interquartile range, adjacent values (lines), and outside values (dots) of the distribution.

more important in controlling distribution patterns within habitats lacking *Sphagnum* (Booth 2001). In this study, microsites without *Sphagnum* were usually characterized by standing water, or at least evidence of standing water earlier in the season. Taxa more abundant in these habitats included *Pontigulasia* sp., *Pyxidicula operculata*, *Arcella vulgaris* type, *Diffugia globulosa* type, and *Diffugia oviformis* type. However, *Diffugia oviformis* type was only encountered in samples from Independence Bog so its ecological range may not have been fully sampled. Taxa abundant in wet *Sphagnum*-dominated habitats included *Amphitrema* spp., *Nebela carinata*, *Nebela marginata*, *Nebela galeata*, and *Heleopera sphagni*. *Sphagnum* species commonly inhabited by these testate amoebae included the more minerotrophic species *S. viridum*, *S. rubellum*, *S. majus* spp. *majus*, and *S. majus* spp. *norvegicum*. Testate amoebae characteristic of drier habitats include *Trigonopyxis* spp., *Hy-*

alospheia subflava, *Bullinularia indica*, *Nebela tinctoria*, *Nebela militaris*, *Heleopera sylvatica*, and *Cyclopyxis arcelloides* type. *Sphagnum* mosses most common in these drier habitats include *S. russowii*, *S. capillifolium*, and *S. angustifolium*.

Development of transfer functions from the calibration dataset

Because water table depth showed the strongest relationship with testate amoeba assemblages (Figure 5, Table 3), I used it to develop a transfer function from the dataset. Water table depth has also been extensively used as the inferred variable in other testate amoeba calibration studies (e.g., Charman (1997), Woodland et al. (1998)). Of the four transfer functions developed, WA-PLS, PLS, and WA-Tol performed the best (Figure 6), although all four models performed better than the only previously developed transfer

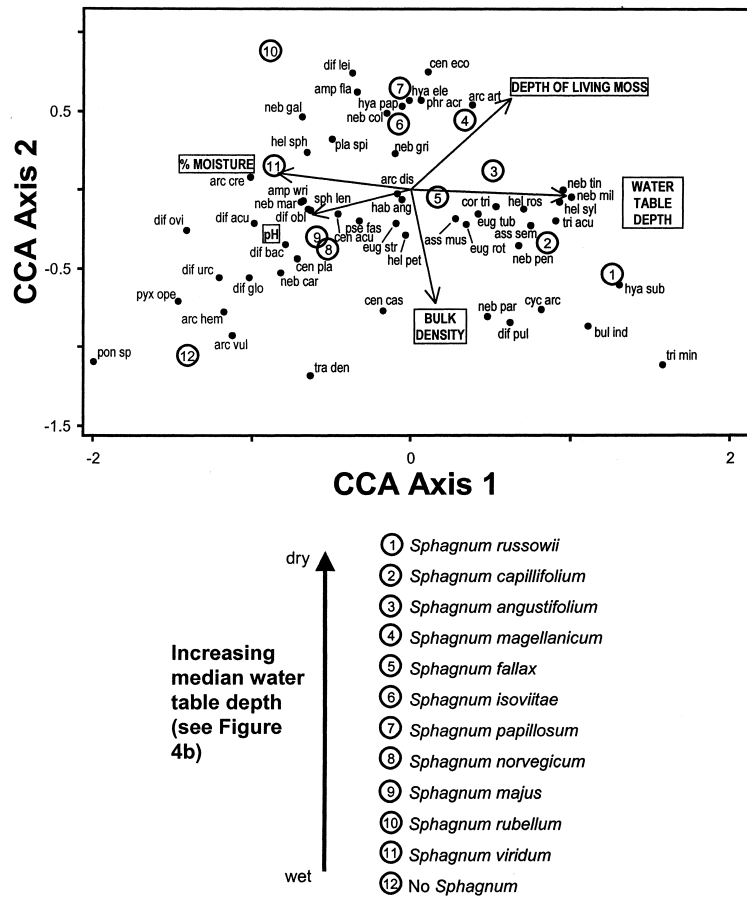


Figure 5. CCA ordination of testate amoebae and environmental variables in the calibration dataset (n = 139). Abbreviations for testate amoebae are shown in Table 2. Numbers refer to the position of *Sphagnum* species, which were included as passive variables. The numbering of the *Sphagnum* species corresponds to the position of the *Sphagnum* species along a water table depth gradient (Figure 4b). Axis characteristics are shown in Table 3.

functions from Michigan, at least in terms of r^2 (Booth 2001). All four models lose predictive ability on the extreme dry side of the gradient (Figure 6), and this has also been observed in other regions (Charman and Warner 1997; Mitchell et al. 1999). In this study,

Table 3. Characteristics of CCA axes 1 and 2 and correlation coefficients for the analysis of the calibration dataset (n = 139)

	Axis 1	Axis 2
Axes characteristics		
Eigenvalue	0.446	0.191
% Variance explained	11.7	5.0
Cumulative % explained	11.7	16.7
Correlation coefficients		
Water table depth	0.868	-0.025
% Moisture	-0.749	0.068
pH	-0.551	-0.105
Bulk density	0.137	-0.484
Depth of living <i>Sphagnum</i>	0.561	0.386

microsites with water table depths greater than ca. 30-cm deep appear to be characterized by a similar group of xerophytic taxa, and deeper water tables within the range of sampling do not appear to significantly affect testate amoeba assemblages. Assuming good modern analogues, water-table depth can be predicted from fossil data with a mean error of ± 7.5 cm (Table 3). However, because this dataset is based on instantaneous measurements of water table depth (i.e., water table depth only measured at the time of sampling), fossil reconstructions should be interpreted within the context of a relative moisture gradient.

Comparison between upper and lower assemblages

Considerable variation in the vertical distance spanned by the upper and lower assemblages existed

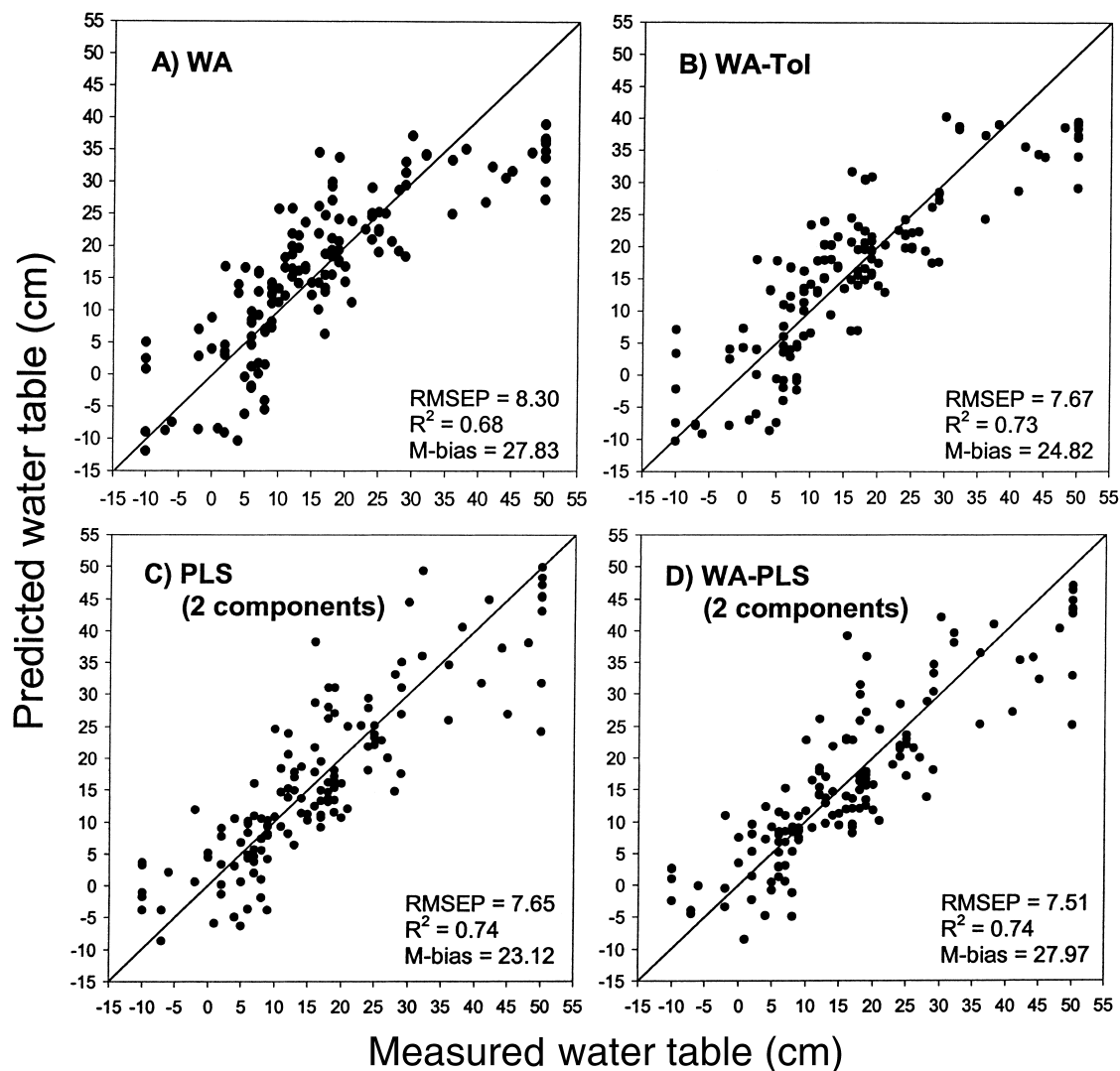


Figure 6. Observed and predicted water table depth values for the 139 samples based on jack-knifed estimates from (A) WA, (B) WA-Tol, (C) PLS, (D) WA-PLS. Performance of the four transfer function models at predicting water table depth from testate amoeba assemblages is assessed by jack-knifed scores (RMSEP), correlation coefficients (r^2), and maximum bias (M-bias). The optimum number of components were used for PLS and WA-PLS, up to a maximum of six.

between microsites, which is probably a result of differences in growth form and species of *Sphagnum*. The upper assemblages were composed of the testate amoebae inhabiting the green portion of the *Sphagnum* stem, which in this study corresponded to the upper 1.5–12 cm of the *Sphagnum* plant. This vertical distance is somewhat longer than some other studies (e.g., Charman and Warner (1992)). However, the average distance spanned by the upper assemblages was 4.5 cm, and only 6 upper assemblages were collected from more than the upper 9 cm. Samples that spanned more than 9 cm were collected from sites with very loose-growing *Sphagnum*, and those span-

ning less than 2 cm were collected from sites with very compact-growing *Sphagnum*. Lower assemblages typically spanned a vertical distance of 2–5 cm, and were collected from directly below the upper assemblage yet above the compacted peat.

Lower assemblages had significantly higher taxonomic richness than upper assemblages (Figure 7). Several factors probably contribute to this. Some taxa occur predominantly on upper portions of the *Sphagnum*, and these taxa eventually become incorporated into the lower assemblage by vertical transport and growth/senescence of the moss. The lower assemblages also represent the accumulation of tests over a

longer time period, and may contain taxa that occur in the upper assemblages at different seasons and in other years. Therefore, the testate amoebae in the lower assemblage may integrate the full range of environmental conditions over the course of a year or multiple years. Also, although lower assemblages contain a larger proportion of dead individuals, many taxa are restricted to living in this portion of the *Sphagnum* (Heal 1962; Meisterfeld 1977). In fact, several taxa were only found in the lower assemblages, including *Nebela penardiana*, *Pseudodiffugia fascicularis*, *Sphenoderia lenta*, and *Tracheuglypha dentata*. However, all taxa encountered in the upper assemblages were also found in the lower assemblages, suggesting that decomposition at this temporal and spatial scale does not completely eliminate any taxa and supporting the hypotheses stated above.

Substrate moisture and pH are dominant controls on distribution patterns in both the upper and lower assemblages, and the total variance explained by each CCA is similar (Figure 8, Table 4). Testate amoeba taxa and *Sphagnum* species are also distributed similarly along environmental gradients in both assemblage types (Figure 8). However, a few exceptions do exist. For example, *Centropyxis cassis* type and *Centropyxis aculeata* type, two taxonomic groups consisting of multiple species, are associated with moister substrates in the lower assemblages. Different species within these groups may inhabit the upper and/or lower assemblages, with more xerophytic species inhabiting the upper portions of the moss. Increased taxonomic resolution within these groups in

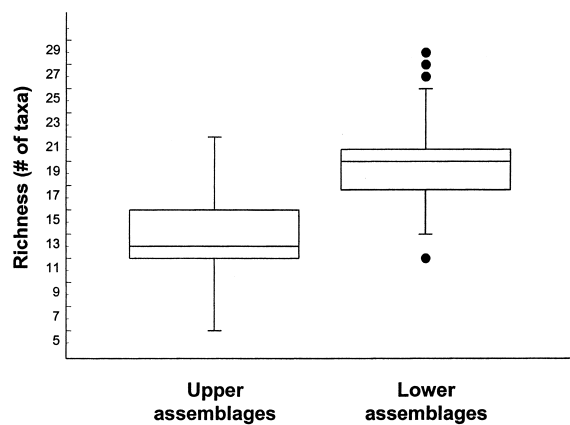


Figure 7. Boxplot showing taxa richness in upper and lower assemblages. The difference between the two groups is statistically significant (paired t-test, $p < 0.001$). Symbols are as in Figure 4.

paleoecological investigations might allow more precise environmental inferences.

Even though most taxa inhabit lower portions of the *Sphagnum* stem, the larger proportion of dead individuals in this region (Heal 1962; Schönborn 1963) suggests that upper assemblages should be more representative of the environmental conditions at the time of sample collection. However, the environmental variables explain the variability of amoeba assemblages on upper and lower portions of the *Sphagnum* stem equally well (Figure 8, Table 4). This suggests that living individuals on lower portions of the *Sphagnum* stem are a large proportion of the total number of tests, and/or the greater species richness present in the lower region increases the overall variance explained by the environmental variables.

The difference between the composition of the upper and lower assemblages appears to be partially related to microsite conditions. DCA of lower and upper assemblages indicates that separation in ordination space between corresponding upper and lower assemblages is smaller in extremely dry environments (Figure 9). Correlation between water table depth and a measure of community similarity (Sørensen's community coefficient) was also significant, although the variance explained was small (Table 5). No significant correlations were found between upper/lower similarity coefficients and any of the other measured environmental parameters (Table 5). The small variance explained by the correlation between similarity and water table depth (Table 5), and the clustering of the more similar upper and lower assemblages in Figure 9, suggests that the relationship between moisture and vertical variation is not linear. Instead, upper and lower assemblages become extremely similar above some threshold of dryness. Alternatively, upper and lower assemblages are more similar because the green portion of the *Sphagnum* stem may correspond to a smaller amount of time in dry versus wet environments. However, the transfer functions based on the calibration dataset have decreased predictive ability in dry environments (Figure 6), suggesting that once a dryness threshold is reached (water table depth of ~30 cm in this study) the same taxa are found between microsites, and vertical differences along the *Sphagnum* stem are likely minimized.

Additional patterns are revealed by directly comparing the abundance and frequency of taxa in the upper and lower assemblages. Although several taxa were more frequent in the lower assemblages, no taxa were more frequent in the upper assemblages (Table

Table 4. Characteristics of CCA axes 1 and 2 and correlation coefficients for the analyses of upper and lower assemblages

Variable	Lower assemblages (n = 89)		Upper assemblages (n = 89)	
	Axis 1	Axis 2	Axis 1	Axis 2
Axes characteristics				
Eigenvalue	0.414	0.139	0.448	0.161
% Variance explained	13.8	4.6	13.6	4.9
Cummulative % explained	13.8	18.5	13.6	18.5
Correlation coefficients				
Water table depth	-0.892	0.064	-0.913	-0.045
% moisture	0.774	0.040	0.743	0.132
pH	0.727	-0.004	0.700	0.093
Bulk density	-0.006	0.504	-0.219	0.498
Depth of living <i>Sphagnum</i>	-0.435	-0.353	-0.308	-0.424

abundant in the lower assemblages include many agglutinate taxa that incorporate foreign material, such as sand grains or fungal hyphae, into their tests. Examples include *Amphitrema wrightianum*, *Centropyxis aculeata* type, *Centropyxis cassis* type, *Cyclopyxis arcelloides* type, *Diffflugia acuminata*, *Diffflugia leidyi*, *Phryganella acropodia* type, and *Trigonopyxis arcula* (Table 6, Figure 10). These taxa

are probably more common in the lower assemblages because of the increased availability of materials to incorporate into their tests and increased food availability (Heal 1962). *Phryganella acropodia*, a mycophagous species (Coûteaux 1985), is probably more common in lower regions of the *Sphagnum* because of the increased availability of fungal material. Only one non-agglutinate taxon, *Arcella discoides*

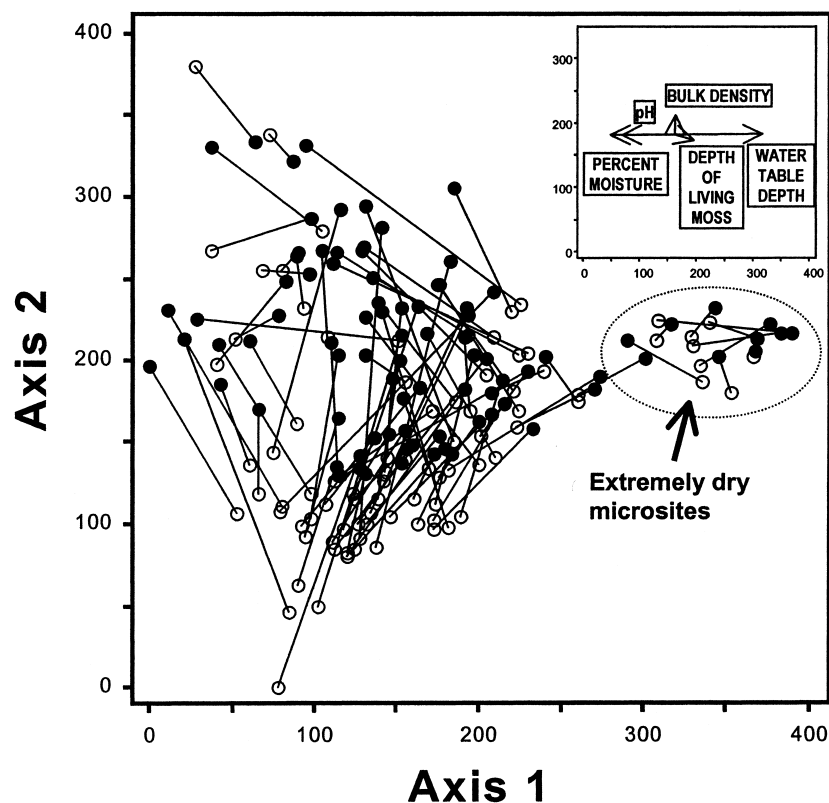


Figure 9. DCA of upper and lower testate amoeba assemblages. Vectors connect each pair of upper and lower assemblages, and thus the length of each vector represents the degree of similarity between each pair of upper and lower assemblages. Inset shows axis correlations with environmental parameters, with the length of the arrows corresponding to the strength of the correlation.

Table 5. Linear correlations of Sorenson's measure of community similarity of the upper and lower assemblages and measured environmental parameters. Significance denoted with an asterisk ($p < 0.05$)

Variable	Correlation (r) with Sorenson's community coefficient
Water table depth	0.22*
% Moisture	0.14
pH	0.03
Depth of living moss	0.14
Bulk density	0.03

type, was more frequent and more abundant in the lower assemblages. *Arcella discoides* type has a proteinaceous test and is common in very wet habitats. Its preference for lower portions of the *Sphagnum* plant may be related to increased moisture and food availability.

Materials for the construction of siliceous tests are

probably also more abundant in lower regions of the *Sphagnum* stems. *Euglypha strigosa* type and *Euglypha tuberculata* type, taxa composed of many siliceous plates, are more frequent in lower assemblages, although only *Euglypha strigosa* type was more abundant in lower assemblages (Figure 10). Spineless forms of *Euglypha* are more common in drier habitats (Bobrov et al. 1999), and vertical moisture gradients may explain the increased abundance of the spined taxon, *Euglypha strigosa* type, on lower, wetter portions of the *Sphagnum* stem. Most of the *Nebela* species were also more frequent and/or more abundant in the lower assemblages. Members of the *Nebela* genus commonly ingest other testate amoebae (primarily *Euglypha* and *Assulina* spp.) and diatoms, incorporating the silica plates and frustules of the ingested organisms into their own tests. However, *Nebela tincta* was more abundant in upper assem-

Table 6. Comparisons of the frequency of testate amoebae in lower and upper assemblages

Taxon	Frequency %			X ²	Odds ratio
	N	Lower	Upper		
<i>Amphitrema flavum</i>	54	83.33	79.63	0.25	1.28
<i>Amphitrema wrightianum</i>	25	95.65	52.17	11.28**	20.17
<i>Arcella artocrea</i>	76	86.84	81.58	0.79	1.49
<i>Arcella discoides</i> type	63	98.41	38.10	52.89**	100.75
<i>Assulina muscorum</i>	89	98.88	94.38	2.81	5.30
<i>Assulina seminulum</i>	65	87.69	70.77	5.66*	2.94
<i>Bullinularia indica</i>	31	64.52	67.74	0.07	0.87
<i>Centropyxis aculeata</i> type	66	87.88	63.64	10.56**	4.14
<i>Centropyxis cassis</i> type	32	84.38	37.50	14.77**	9.00
<i>Centropyxis ecornis</i> type	53	88.68	77.36	2.41	2.29
<i>Corythion dubium</i> type	37	78.38	51.35	5.93*	3.43
<i>Cyclopyxis arcelloides</i> type	32	96.88	6.25	52.61**	465.00
<i>Diffflugia acuminata</i>	30	86.67	46.67	10.80**	7.43
<i>Diffflugia globulosa</i>	36	94.44	66.67	8.87*	8.50
<i>Diffflugia leidy</i>	27	81.48	37.04	11.05**	7.48
<i>Euglypha strigosa</i> type	42	92.86	23.81	41.19**	41.60
<i>Euglypha tuberculata</i> type	79	91.14	67.09	13.83**	5.05
<i>Habrotricha angusticollis</i>	52	94.23	36.54	38.34**	28.37
<i>Heleopera sphagni</i>	68	89.71	94.12	0.89	0.55
<i>Heleopera sylvatica</i>	52	96.15	55.77	23.22**	19.82
<i>Hyalosphenia elegans</i>	74	94.59	85.14	3.64	3.06
<i>Hyalosphenia papilio</i>	78	98.72	98.72	0.00	1.00
<i>Nebela carinata</i>	25	95.45	68.18	5.50*	9.80
<i>Nebela collaris</i>	65	84.62	70.77	3.60	2.72
<i>Nebela griseola</i>	78	98.72	77.92	16.11**	21.53
<i>Nebela militaris</i>	45	97.78	46.67	29.30**	50.29
<i>Nebela tincta</i>	58	91.38	72.41	7.04**	4.04
<i>Phryganella acropodia</i>	75	97.33	74.67	16.00**	12.38
<i>Trigonopyxis arcuata</i>	65	93.85	41.54	40.66**	21.46

Taxa not encountered in at least 25 samples were excluded from the analysis, and comparisons were only made using sample pairs (i.e., lower and upper) in which the taxon was present in one of the samples. The frequency of each taxon is shown as the percentage of samples in which the taxon was present. Chi-square tests assess significant differences between upper and lower assemblages (* $p < 0.05$; ** $p < 0.01$). Odds ratios describe the increased probability of finding a taxon in upper or lower assemblages.

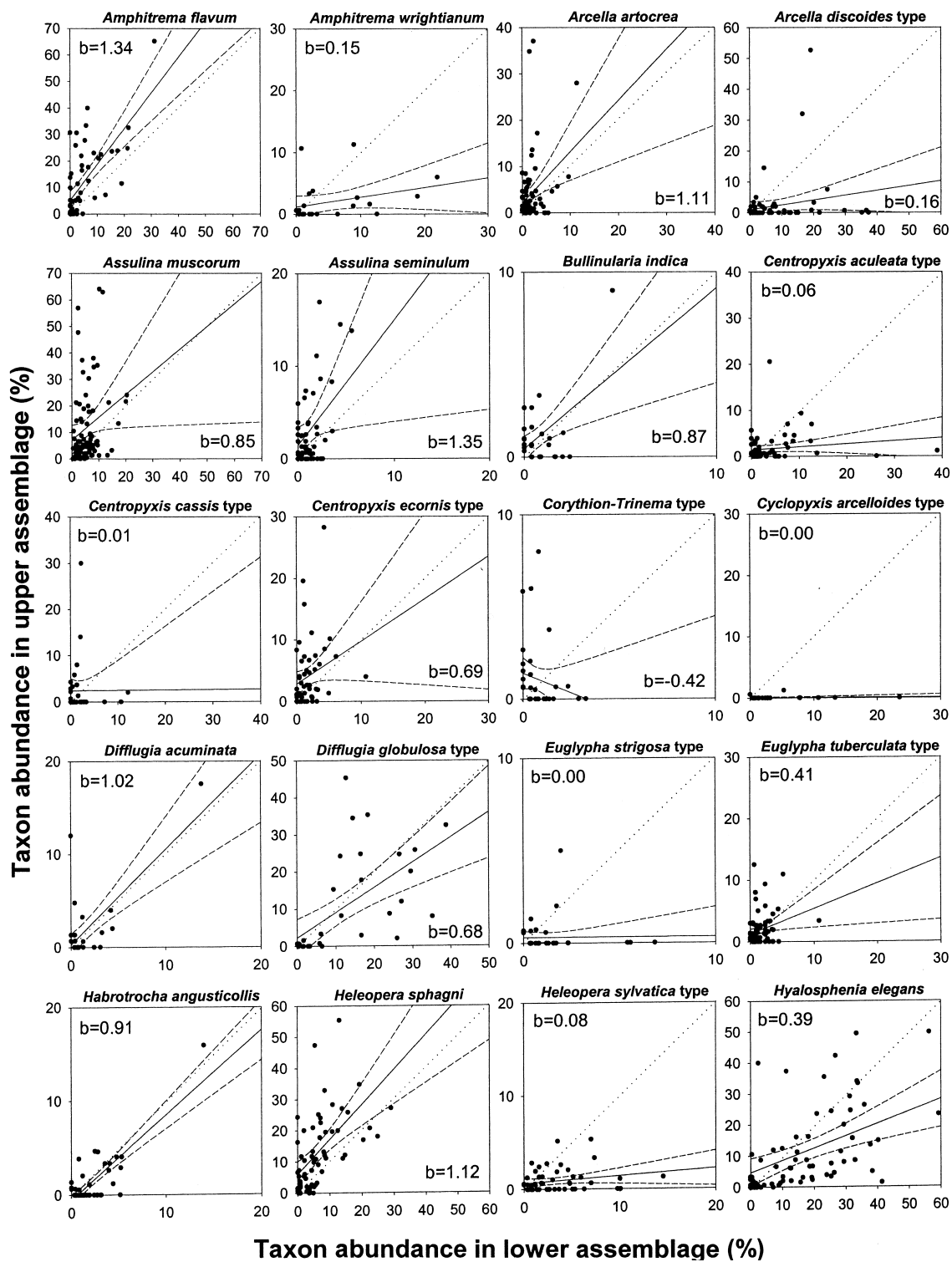


Figure 10. Comparison of the abundance of common taxa (present in at least 25 sample pairs) in upper and lower assemblages. The slope of the regression line (b) is indicated for each comparison

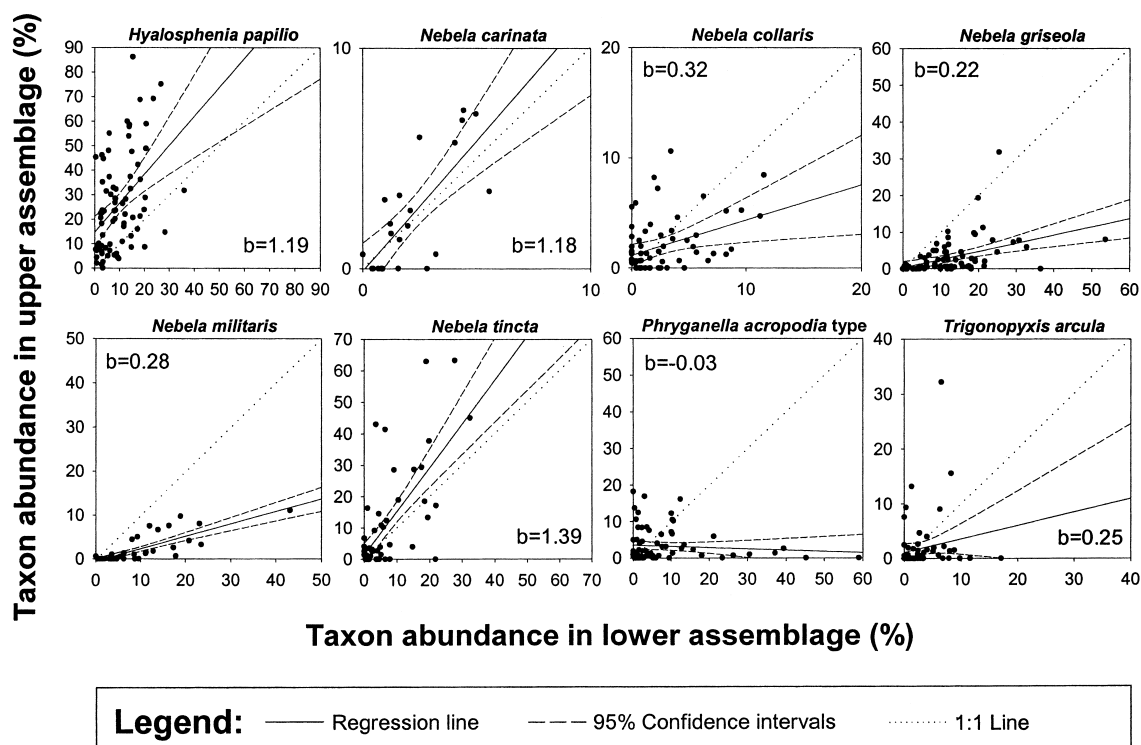


Figure 10. continued

blages and perhaps has more alternate food sources than other *Nebela* species. Individuals of *N. tinctoria* in upper assemblages also appear to incorporate fewer silica plates into their tests than those encountered in lower assemblages, although this was not quantified. However, experimental studies have shown that *N. collaris* produces proteinaceous shells lacking shell plates when cultured in the absence of other small testate amoebae (MacKinlay 1936). Heal (1963) found that *N. tinctoria* was the most abundant of the *N. tinctoria-collaris-bohemica* group at the tops of hummocks, and this suggests that it may be more tolerant of the drier conditions at the top of the *Sphagnum* stem.

In addition to *Nebela tinctoria*, several other taxa were more abundant in the upper assemblages (Figure 10), although many of these taxa were equally frequent in upper and lower assemblages (Table 6) (e.g., *Amphitrema flavum*, *Heleopera sphagni*, and *Hyalosphenia papilio*). *Amphitrema flavum*, *Heleopera sphagni*, and *Hyalosphenia papilio* possess symbiotic zoochlorellae, and their increased abundance in upper portions

of *Sphagnum* where light is more available has been observed in other studies (Heal 1962; Schönborn 1963; Meisterfeld 1977). In addition to taxa containing symbiotic zoochlorellae and *Nebela tinctoria*, other taxa more common in upper assemblages include *Arcella artocrea* type and *Assulina seminulum*.

The vertical distribution patterns of testate amoebae in this study are similar to studies from Europe (Heal 1962; Schönborn 1963; Meisterfeld 1977; Buttler et al. 1996). Agglutinate taxa are more common on lower portions of the *Sphagnum* stem, and taxa with symbiotic zoochlorellae, *Amphitrema flavum*, *Hyalosphenia papilio*, and *Heleopera sphagni*, are consistently found in higher numbers at the top of the *Sphagnum* stem (Heal 1962; Schönborn 1963; Meisterfeld 1977). *Amphitrema wrightianum* also contains symbiotic zoochlorellae, but in this study was not found to be higher in abundance or frequency on the upper portions of the stem. However, *Amphitrema wrightianum* has been shown to occur lower than the related, although non-agglutinate species, *Amphitrema flavum* (Heal 1962). The vertical distribution

pattern of *Amphitrema wrightianum* has been interpreted as a compromise between the availability of light and particles for test construction (Heal 1962).

Implications of vertical variation to the collection of calibration datasets

Previous calibration datasets derived from *Sphagnum*-dominated peatlands have used assemblages collected from the lower portions of the *Sphagnum* stem, because assemblages from this horizon are thought to best represent what is found in the fossil record (Warner 1987; Tolonen et al. 1992). This study indicates that taxon richness is higher on lower portions of the *Sphagnum* stem (Figure 7), all taxa present in upper assemblages are also present in lower assemblages (Table 6), and significant differences exist in the abundance of some taxa between upper and lower portions of the *Sphagnum* stem (Figure 10). However, the relationships between environmental variables and testate amoebae are just as strong in both assemblage types (Figure 8, Table 4), probably because living individuals occur on both portions of the *Sphagnum* stem (Heal 1962; Schönborn 1963). Since a greater percentage of living individuals probably comprise upper assemblages, and living individuals should better represent the environmental conditions at the time of sampling, combination of the two assemblage types might show stronger relationships with the environmental variables than either assemblage type alone. To test this hypothesis I combined the raw data from each upper and lower assemblage pair and repeated the CCA. The analysis of the combined dataset indicates that relationships with the environmental variables improve, and the total variance explained increases (Table 7) compared to separate analyses of the two assemblage types (Table 4).

Table 7. Characteristics of CCA axes 1 and 2 and correlation coefficients for the analysis of the combined upper and lower assemblage dataset (n = 89)

	Axis 1	Axis 2
Axes characteristics		
Eigenvalue	0.408	0.141
% Variance explained	16.7	5.8
Cumulative % explained	16.7	22.5
Correlation coefficients		
Water table depth	-0.929	-0.017
% Moisture	0.782	0.094
pH	0.737	0.064
Bulk density	-0.120	0.523
Depth of living <i>Sphagnum</i>	-0.382	-0.416

The increased variance explained by combining lower and upper assemblages (Table 7) suggests that improvements in the predictive ability of transfer functions may be possible. Sampling the entire vertical region of *Sphagnum* where living testate amoebae occur may result in more accurate depictions of species-environment relationships. However, more research is needed on the effects of sampling larger vertical regions of *Sphagnum* and determining the vertical extent of the zone where living testate amoebae occur. Identification and quantification of living individuals would also improve the correlation between amoeba assemblages and hydrology, especially when comparisons are made with instantaneous measurements of hydrologic conditions. Although living and dead individuals are easily distinguished in some taxa, in others it is difficult or impossible (Heal 1964), and some living individuals can remain encysted for long periods of time.

Conclusions

Testate amoeba assemblages are primarily controlled by local hydrology in Michigan peatlands, similar to other regions. Particularly in *Sphagnum*-dominated peatlands, testate amoebae have considerable potential in the reconstruction of past surface-moisture variability. Testate amoebae have been underused in North American paleoclimate studies, although their sensitivity to low-magnitude and high-frequency hydrologic fluctuations offers advantages over other climate proxies (Charman et al. 1999). The calibration dataset presented in this study can be used to quantitatively infer past surface-moisture conditions from fossil testate amoeba data in the western Great Lakes region and perhaps other regions in North America. Water table depth can be reconstructed from fossil data with a mean error of ± 7.5 cm, although reconstructed water table values should be interpreted along a relative moisture gradient.

Vertical variation of amoeba assemblages along the stems of *Sphagnum* moss is considerable, although this variation appears to be reduced in dry environments. Taxa with agglutinate or siliceous shells are generally more abundant lower on the *Sphagnum* stem whereas taxa with symbiotic zoochlorellae are generally found higher on the stem. In addition to light and the availability of materials for test construction, vertical variation is probably related to gradients of moisture and food. Assemblages characterizing the

upper portions of the *Sphagnum* stem are not more representative of environmental conditions than those inhabiting lower portions of the *Sphagnum* stem, although taxon richness is higher on lower portions. Further developments in sampling methodology, and perhaps the identification and quantification of living testate amoebae, may facilitate the collection of more representative calibration datasets.

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