

Ecology and paleoenvironmental application of testate amoebae in peatlands of the high-elevation Colombian páramo

Bing Liu^{a,b}, Robert K. Booth^{a*}, Jaime Escobar^{c,d}, Zhiqiang Wei^{a,b}, Broxton W. Bird^e, Andres Pardo^f, Jason H. Curtis^g, Jun Ouyang^b

^aEarth & Environmental Science Department, Lehigh University, Bethlehem, Pennsylvania 18015, USA

^bSchool of Geography, South China Normal University, Guangzhou, GD, China.

^cDepartamento de Ingeniería Civil y Ambiental, Universidad del Norte, Km 5 Vía Puerto Colombia, Colombia

^dCenter for Tropical Paleocology and Archaeology, Smithsonian Tropical Research Institute, Balboa, Panama

^eDepartment of Earth Sciences, Indiana University–Purdue University, Indianapolis, Indiana, USA

^fInstituto de Investigaciones en Estratigrafía, Universidad de Caldas, Manizales, Colombia

^gDepartment of Geological Sciences, University of Florida, Gainesville, Florida 32611, USA

*Corresponding author e-mail address: rkb205@lehigh.edu

(RECEIVED April 5, 2018; ACCEPTED November 13, 2018)

Abstract

We investigated the ecology and paleoecology of testate amoebae in peatlands of the Colombian páramo to assess the use of testate amoebae as paleoenvironmental indicators. Objectives were to (1) identify environmental controls on testate amoebae, (2) develop transfer functions for paleoenvironmental inference, and (3) examine testate amoebae in a Holocene peat core and compare our findings with other proxy records. Results from 96 modern samples indicate that testate amoebae are sensitive to pH and surface moisture, and cross-validation of transfer functions indicates potential for paleoenvironmental applications. Testate amoebae from the Triunfo Peatland in the Central Cordillera provided a proxy record of pH and water-table depth for the late Holocene, and inferred changes were correlated with peat C/N measurements during most of the record. Comparison with a lake-level reconstruction suggests that at least the major testate amoeba-inferred changes were driven by climate. Our work indicates that testate amoebae are useful paleoenvironmental indicators in high-elevation tropical peatlands.

Keywords: Testate amoebae; Paleoenvironment; Hydrology; Bioindicators; Páramo ecosystem; Tropical peatland; Holocene

INTRODUCTION

Holocene paleoclimatic and paleoecological records have been developed from many natural archives and collectively highlight the tremendous spatiotemporal variability in the climate of the Earth system (e.g., Chen et al., 2008; Wanner et al., 2008; Baker and Fritz, 2015; Flantua et al., 2016; Marlon et al., 2017). Furthermore, these records demonstrate that climatic variability has been sufficient in magnitude and rate to drive rapid changes in disturbance regimes and ecosystems (e.g., Marchant et al., 2001; Marlon et al., 2013; Clifford and Booth, 2015; Flantua et al., 2016; Calder and Shuman, 2017). However, the climatic and ecological history of some critically important regions and ecosystems is incompletely described, and this is particularly true in tropical regions,

where much of the planet's biodiversity and nearly 3 billion people reside (Cincotta et al., 2000; Myers et al., 2000; Kummu and Varis, 2010). One tropical ecosystem type of considerable importance from a hydrologic and biodiversity perspective is the páramo, an ecosystem located above the upper forest line and below the permanent snowline in the Andes of tropical South America and the highlands of Costa Rica (Baruch, 1984). Up to 60% of its plant species are endemic, and together with the surrounding Andean forest, the region may be home to 50% of the plant diversity found in mountain ecosystems (e.g., Cleef, 1981; Vuilleumier and Monasterio, 1986; Luteyn, 1999; Sklenář et al., 2005). In addition to its high conservation value, the páramo and its watersheds store and supply critical water resources for major Andean rivers and cities (Buytaert et al., 2006, 2011). High-altitude tropical ecosystems like the páramo are expected to experience increasing temperature in the coming decades, with considerable spatial variability in anticipated precipitation changes (Buytaert et al., 2011). Such projections highlight the need for a dense spatial network of paleoclimatic

Cite this article: Liu, B., Booth, R.K., Escobar, J., Wei, Z., Bird, B.W., Pardo, A., Curtis, J.H., Ouyang, J. 2019. Ecology and paleoenvironmental application of testate amoebae in peatlands of the high-elevation Colombian páramo. *Quaternary Research* 92, 14–32.

and paleoecological records that can capture the spatial complexity of climatic and ecological variability at a range of temporal scales to better assess model projections, anticipate climate-induced ecological and hydrologic impacts, and assist in management efforts (e.g., Marchant et al., 2001).

Pollen studies from the high-elevation Andes have provided detailed records of vegetation history and centennial- to millennial-scale temperature change (e.g., Bosman et al., 1994; Marchant et al., 2001; Bakker et al., 2008; Bogota-A et al., 2011; González-Carranza et al., 2012; Velásquez and Hooghiemstra, 2013; Giraldo-Giraldo et al., 2017; Munoz et al., 2017). Paleoenvironmental reconstructions using multiple proxies, however, provide richer information on past environments and potentially greater sensitivity to decadal- and multidecadal-scale climatic and ecological changes (Marlon et al., 2017). Furthermore, vegetation-independent proxies for past hydrology and/or temperature have proved extremely valuable for directly assessing vegetation sensitivity to past climatic variability and examining vegetation responses to abrupt climate change (e.g., Booth et al., 2012). For example, testate amoebae, a group of protozoa that produce decay-resistant shells, have been widely applied as paleoenvironmental proxies in north-temperate and boreal regions (e.g., Woodland et al., 1998; Lamentowicz and Mitchell, 2005; Booth, 2008; Payne et al., 2008; Swindles et al., 2009; Markel et al., 2010; Lamarre et al., 2013; Amesbury et al., 2016) and are a potential source of information on past hydrology and ecology that could be developed from the many peatlands and lakes of the páramo. However, applying testate amoebae and similar approaches is contingent on adequate ecological understanding and calibration of paleoenvironmental proxies, and little work on the calibration of paleoenvironmental indicators other than pollen has been undertaken in the páramo to date.

Testate amoebae occur in a range of environments, including lakes and peatlands, where they have been used for paleoenvironmental reconstruction (e.g., Charman, 2001; Mitchell et al., 2008). Studies on peatland-inhabiting testate amoebae have demonstrated that community composition in these habitats is primarily controlled by environmental conditions at the surface of the peatland, particularly substrate moisture and pH (Mitchell et al., 2008). Well-defined ecological niches with respect to substrate moisture and pH have been described empirically and used to develop transfer functions for paleohydrologic reconstruction in many temperate and boreal regions, including portions of Europe (e.g., Woodland et al., 1998; Lamentowicz and Mitchell, 2005; Payne et al., 2008; Swindles et al., 2009; Amesbury et al., 2016), North America (e.g., Booth, 2008; Markel et al., 2010; Lamarre et al., 2013; Swindles et al., 2015), South America (e.g., Van Bellen et al., 2014), New Zealand (Charman, 1997; Wilmschurst et al., 2003), and Asia (Qin et al., 2013; Song et al., 2014; Li et al., 2015). However, very few modern calibration studies have focused on tropical ecosystems (Swindles et al., 2014), and although testate amoebae have been described from the páramo (Grabandt, 1990; Montoya et al., 2010), species–environment relationships have not been quantitatively explored for transfer function development.

In this study, we investigated the modern ecology of testate amoebae in peatlands of the Colombian páramo to assess their potential use as paleoenvironmental indicators in these ecosystems. Our specific objectives were to (1) assess potential environmental controls on testate amoeba communities in páramo peatlands, (2) develop transfer functions for paleoenvironmental inference, and (3) examine testate amoebae in a peat core spanning the late Holocene. Finally, we use our modern calibration data set to infer past environmental changes from our subfossil data set and compare our inferences to geochemical and pollen records from the same core and a recently published lake sediment record from the region.

STUDY SITES

Colombia has three major orogenic belts that support páramo ecosystems: the Western, Central, and Eastern Cordillera. Our study sites for modern sampling were located in the Central and Eastern Cordillera, with four peatlands located near the city of Manizales just north of the Nevado del Ruiz Volcano in the Central Cordillera and one peatland in the Eastern Cordillera near the city of Sogamoso (Fig. 1, Table 1). A peat core was analyzed from the Triunfo peatland, one of our Central Cordillera sites, and was described in more detail by Giraldo-Giraldo et al. (2017).

The Eastern Cordillera of Colombia is a northeast-trending inverted basin where more than 6 km of sediment accumulated during Cretaceous times (Kerr and Tarney, 2005; Spikings et al., 2015). These sediments include thick sequences of black shales and nearly pure quartz arenites derived from the Guiana craton to the east. Near Lake Tota, a singular, Miocene volcanic complex is present, but represents an anomalous occurrence in the deformed, mostly sedimentary pile of the Eastern Cordillera. The Central Cordillera near the Ruiz volcano is composed of a mostly Paleozoic–Mesozoic plutonic–metamorphic complex of continental affinities to the east and mostly Mesozoic–Cenozoic oceanic–affinity volcanic and sedimentary sequences to the west (Gonzalez, 2001). These sequences are separated by a north- to northeast-trending suture. The Ruiz volcano and other volcanoes in the Central Cordillera are Pliocene and younger, the result of subduction of the Nazca plate under northwestern South America (Kerr and Tarney, 2005; Spikings et al., 2015).

In the Colombian Andes the seasonal hydrologic cycle is controlled by the interhemispheric movement of the intertropical convergence zone (ITCZ), superimposed on more local patterns caused by orographic relief, evapotranspiration in the Amazon basin, continent–atmosphere–ocean interactions (i.e., El Niño–Southern Oscillation [ENSO]), and dynamics of the western Colombian wind currents (Mesa et al., 1997; Poveda et al., 2001; Poveda and Mesa, 2004; Flantua et al., 2016). Due to the double annual passage of the ITCZ over the Colombian Andes, the Central Cordillera experiences a bimodal annual cycle with wet seasons in April–May and September–November (Poveda et al., 2011). Precipitation exhibits a unimodal peak from June to August on the eastern slopes of the Eastern Cordillera as a result of the convection

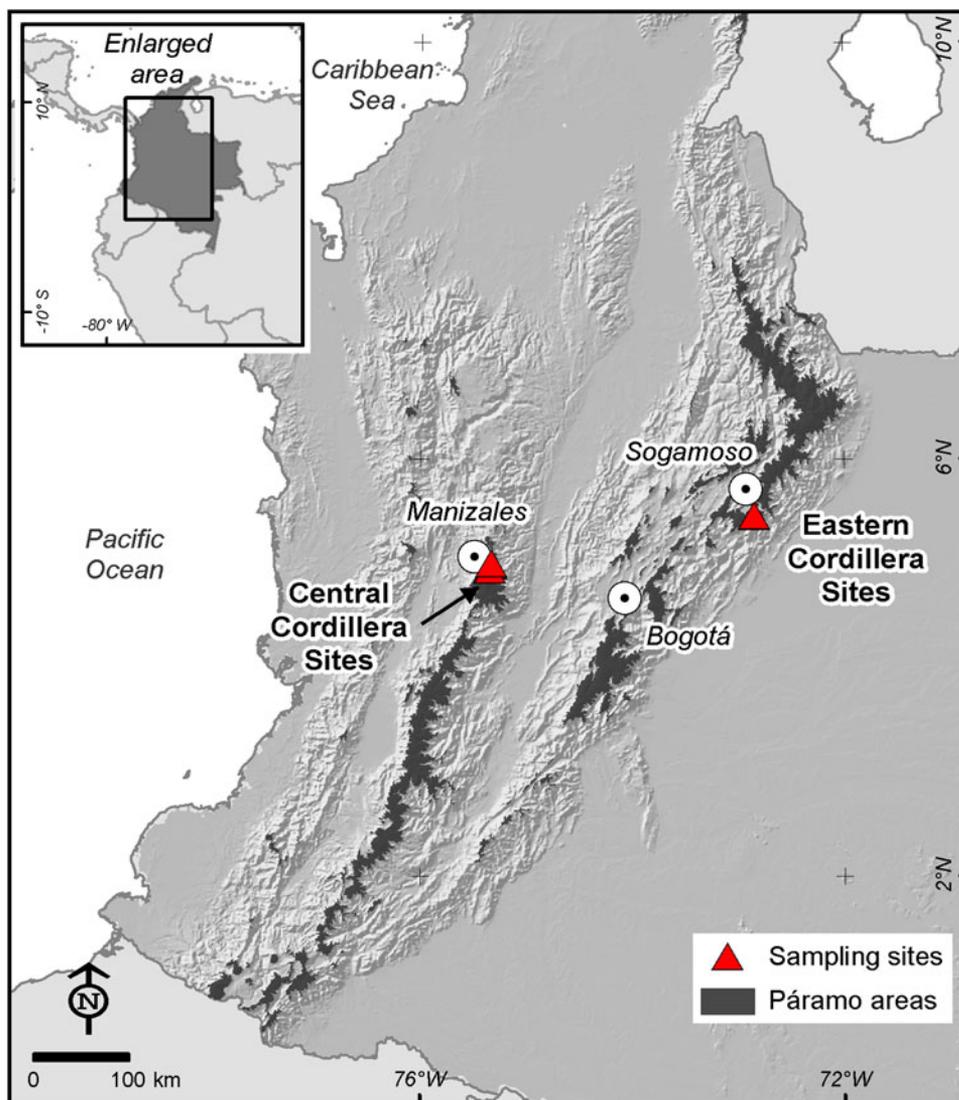


Figure 1. (color online) Location of the sampled peatlands in the Central and Eastern Cordillera of Colombia, showing the nearest major cities to our sampling locations and the distribution of páramo in Colombia (data for páramo distribution from Instituto Alexander Von Humboldt [2012]; <http://www.siac.gov.co/catalogo-de-mapas>).

of humid air masses from the Amazon basin into the Andes (Poveda et al., 2011). Over interannual and decadal times scales, ENSO and other ocean–atmosphere phenomena such as the Pacific Decadal Oscillation, North Atlantic Oscillation, and Atlantic Multidecadal Oscillation influence the climate of the region (Poveda et al., 2002, Martínez et al.,

2015; Flantua et al., 2016). In the Colombian Andes in particular, ENSO has a strong effect on precipitation, river discharge, and soil moisture (Poveda et al., 2001, 2002). El Niño events (warm phase) are associated with a decrease in precipitation, a decrease in soil moisture and evapotranspiration, and, consequently, a decrease in riverine discharge in the

Table 1. Studied peatlands and site-specific data.

Site	Samples (n)	Longitude	Latitude	Elevation	Water-table depth range	pH range
Triunfo (Central Cordillera)	34	−75.331	4.990	3825 m	−11 to 40 cm	3.89 to 6.79
Casa Blanca (Central Cordillera)	20	−75.333	4.997	4050 m	−3 to 67 cm	No data
Nevada del Ruiz (Central Cordillera)	5	−75.344	4.939	4050 m	−2 to 14 cm	No data
Laguna Negra (Central Cordillera)	20	−75.337	4.980	3850 m	−12 to 51 cm	3.90 to 5.51
Aquitania (Eastern Cordillera)	28	−72.849	5.456	3775 m	−8 to 50 cm	No data

central region of Colombia (Poveda et al., 2001). La Nina events (cold phase) bring the opposite pattern, with intense and abundant rainfall and increased river discharge (Mesa et al., 1997; Poveda et al., 2001).

All modern sampling sites for testate amoebae were located between 3750 and 4100 m in elevation and were surrounded by páramo vegetation (Table 1). Peatlands were bryophyte dominated, including abundant mosses likely representing species in the genus *Breutelia*, although many other similar mosses were likely also present. *Sphagnum* mosses and cushion plants (*Plantago rigida*) were locally abundant within several of the peatlands. A diversity of other vascular plants were also present, including species of Cyperaceae, Asteraceae, Apiaceae, Rosaceae, and Gunneraceae, although these species rarely dominated the sampling locations. A more detailed vegetation and geologic description of the Triunfo peatland where the peat core was collected can be found in Giraldo-Giraldo et al. (2017).

METHODS

Field methods

Modern testate amoeba sampling

At each of the five studied peatlands, we collected surface substrate samples for testate amoebae in an effort to capture the full range of moisture and vegetation variability of the site. At locations where moss or moss mixed with low-growing vascular plants occurred, we collected approximately 10 cm³ of the upper 5–7 cm of the moss and associated vegetation. At locations with standing water that lacked vegetation, we collected approximately 10 cm³ of the upper 1–2 cm of the loose substrate. Each sampling location was photographed, and these photographs were later used to quantify the presence or absence of various plant groups at the surface where testate amoebae live as an additional descriptor of the microhabitat. Microhabitat groups included *Sphagnum* moss, *Disterigma* sp., cf. *Breutelia*, *Plantago rigida*, other mosses, and standing water.

At each sampling location at Triunfo and Laguna Negra, water-table depth, pH, and conductivity were measured. However, our pH meter broke after sampling these two peatlands, and for the remaining sites we could only obtain water-table depth measurements for sampling locations. Water-table depth was measured as distance from the peatland surface to the water table, by digging a small hole and waiting for the water table within the hole to equilibrate with the surrounding peat. Positive values were used to denote water-table depths below the surface of the peat, whereas standing water was recorded as negative values (i.e., higher values indicate drier conditions). Conductivity and pH were measured with an Oakton multiparameter meter, with measurements made on water squeezed from surface moss/substrate for locations without standing water, and measurements taken directly from standing water where present. Measurements were made in this way to record the

water-chemistry environment experienced most directly by testate amoebae, which live on the moss water film and on the surface substrate.

Peat-core collection

A 7-m-long peat core was collected with a Russian corer from near the center of the Triunfo peatland in 2012. A pollen record from the upper portion of the core has been previously published (Giraldo-Giraldo et al., 2017). The peatland is located in the Central Cordillera of Colombia at about 3800 m above sea level (m asl); it lies in a depression created by late Quaternary glacial activity and is surrounded by escarpments of andesitic and basaltic volcanic deposits (Herd, 1982). The peatland is fed hydrologically by direct precipitation and runoff from the surrounding volcanic deposits. The drainage basin is characterized by páramo vegetation, although livestock grazing and other human activities have modified the natural vegetation patterns. The peat core was collected from Triunfo peatland in 50-cm-long increments, and individual core drives were wrapped in the field and sent to the Institute for Stratigraphic Research at Universidad de Caldas for cold storage and subsampling.

Laboratory methods

Testate amoebae were isolated from both modern and subfossil samples using the method of Booth et al. (2010). For peat-core analyses, approximately 1 cm³ of peat was collected from 1-cm spanning peat slices at approximately 4–6 cm increments along the core, except within thick tephra layers lacking in organic matter. Both modern and subfossil samples were boiled in distilled water for 10 minutes and washed through 355- μ m- and 15- μ m-diameter sieves, and the material caught in the 15- μ m sieve was stored in glycerol. Microscopic slides were prepared from these residues, and all testate amoebae were identified and tallied under a light microscope at 400 \times magnification until a minimum of 200 or 100 individuals was obtained for the modern and subfossil samples, respectively. Identifications were made using various guides and literature sources (Table 2). For a few fossil samples, count totals of 100 individuals were not obtainable, and only fossil samples with at least 50 tallied testate amoebae were included in our interpretation and analyses. The relative abundance of each taxon was calculated as a percent of the total number of testate amoeba counted in the sample.

Sediment samples for geochemical analyses were dried and crushed with a mortar and pestle before geochemical analysis. Geochemical analyses, including total carbon, total inorganic carbon, total nitrogen, $\delta^{15}\text{N}$, and $\delta^{13}\text{C}$, were performed on core samples at the Light Stable Isotope Mass Spectrometer Laboratory in the Department of Geological Sciences at the University of Florida. Total carbon and total nitrogen were measured using a Carlo Erba NA 1500 CNS elemental analyzer. Inorganic carbon was determined by acidification followed by coulometric titration using an AutoMate Prep Device coupled with a UIC 5014 CO₂

Table 2. Testate amoeba taxa identified in modern and subfossil samples from the studied páramo peatlands, with information on taxonomy and identification. Ranking along the nonmetric multidimensional scaling (NMDS) axis 1 gradient is also shown, with higher numbers generally indicating greater abundance in drier, more acidic habitats and lower numbers generally indicating higher abundance in wetter, less acidic habitats. Taxa without a ranking occurred in fewer than four samples and were excluded from analyses.

Taxon	NMDS axis 1 ranking on Figure 3	Identification source and/or description
<i>Amphitrema wrightianum/stenostoma</i> type	17	Charman et al. (2000)
<i>Archerella flavum</i>	54	Charman et al. (2000)
<i>Apodera vas</i> type		Loeblich and Tappan (1961)
<i>Arcella catinus</i> type	29	Charman et al. (2000)
<i>Arcella crenulata</i> type	1	Likely includes <i>Arcella mitrata</i> ; Leidy (1879), Deflandre (1928).
<i>Arcella discoides</i> type	11	Charman et al. (2000)
<i>Arcella hemisphaerica</i> type	7	Charman et al. (2000)
<i>Arcella vulgaris</i> type	19	Charman et al. (2000)
<i>Argynnia caudata</i> type	57	Horn number varied from 2 to 4; Leidy (1879).
<i>Assulina muscorum</i> type	48	Includes the typical brown-colored individuals as well as clear individuals (<i>Valkanovia elegans</i>); Schönborn and Peschke (1990), Charman et al. (2000)
<i>Centropyxis aculeata</i> type	9	All <i>Centropyxis</i> species with spines were included; Charman et al. (2000)
<i>Centropyxis cassis</i> type	32	Charman et al. (2000)
<i>Centropyxis platystoma</i> type	35	Charman et al. (2000)
<i>Certesella martiali</i>	42	Loeblich and Tappan (1961)
<i>Corythion</i> spp.	53	Charman et al. (2000)
<i>Cryptodiffugia crenulata</i> type	5	Playfair (1917)
<i>Cryptodiffugia oviformis</i> type	12	Penard (1902)
<i>Cyclopyxis arcelloides</i> type	40	Charman et al. (2000), as modified by Booth (2008)
<i>Cyphoderia ampulla</i>	14	Charman et al. (2000)
<i>Diffugia acuminata</i> type	4	Charman et al. (2000)
<i>Diffugia globulosa</i> type	25	Booth (2008)
<i>Diffugia lucida</i> type	10	Charman et al. (2000)
<i>Diffugia oblonga</i> type	15	Charman et al. (2000)
<i>Diffugia pristis</i> type	21	Charman et al. (2000)
<i>Diffugia pulex</i> type	33	Charman et al. (2000); also includes individuals similar to <i>Diffugia pulex</i> , but about twice as long.
<i>Diffugia rubescens</i> type	3	Charman et al. (2000)
<i>Diffugia</i> small spherical type	2	Individuals somewhat similar to <i>Phryganella dissimulatoris</i> (Chardez, 1969); small (20–30 µm), spherical, with diatoms and mineral grains.
<i>Diffugia</i> undifferentiated	24	Undifferentiated <i>Diffugia</i>
<i>Euglypha cristata</i> type	45	Charman et al. (2000)
<i>Euglypha rotunda</i> type	44	Charman et al. (2000)
<i>Euglypha strigosa</i> type	51	Charman et al. (2000)
<i>Euglypha tuberculata</i> type	43	Charman et al. (2000)
<i>Heleopera rosea</i> type	28	Charman et al. (2000)
<i>Heleopera sphagni-petricola</i> type	30	Charman et al. (2000)
<i>Heleopera sylvatica</i>	52	Charman et al. (2000)
<i>Lesquereusia spiralis</i> type	8	Ehrenberg (1840)
<i>Nebela collaris-bohemica</i> type	50	Charman et al. (2000)
<i>Nebela galeata-carinata</i> type	18	Individuals with keels of variable thickness were encountered, so these were combined into one group; Charman et al. (2000)
<i>Nebela militaris</i>	58	Charman et al. (2000)
<i>Nebela penardiana</i> type	26	Charman et al. (2000)
<i>Nebela</i> type		Individuals with circular or hexagonal-like plates, laterally compressed;
<i>Nebela tincta-parvula</i> type	46	Charman et al. (2000)
<i>Nebela vitraea</i> type	36	Charman et al. (2000)
<i>Nebela wailesi</i> type	38	Charman et al. (2000)
<i>Netzelia oviformis</i> type		Ogden (1979)
<i>Netzelia wailesi</i> type	6	Ogden (1980)

Table 2. Continued.

Taxon	NMDS axis 1 ranking on Figure 3	Identification source and/or description
<i>Paraquadrula irregularis</i> type	13	Deflandre (1932)
<i>Phryganella acropodia</i> type	27	Charman et al. (2000), as modified by Booth (2008)
<i>Plagiopyxis labiata</i> type	55	Penard (1910)
<i>Placocista spinosa</i>	56	Charman et al. (2000)
<i>Pseudodiffugia fulva</i> type	23	Charman et al. (2000)
<i>Pyxidicula cymbalum</i> type	20	Penard (1902)
<i>Quadrueella-Apodera</i> type	22	Individuals consistent with <i>Apodera</i> , but with square plates.
<i>Quadrueella symmetrica</i> type	31	Charman et al. (2000)
<i>Quadrueella wailesi</i> type	16	Individuals with secreted square plates similar to <i>Quadrueella symmetrica</i> type, but test shape similar to <i>Nebela wailesi</i> type.
<i>Sphenoderia lenta</i> type	47	Charman et al. (2000)
<i>Tracheleuglypha dentata</i> type	34	Charman et al. (2000)
<i>Trinema enchelys</i> type	41	Charman et al. (2000)
<i>Trinema lineare</i> type	39	Charman et al. (2000)

coulometer. Percent organic carbon was calculated by subtraction of inorganic carbon from total carbon. Samples for carbon stable-isotope analysis of organic matter were treated with 2N HCl to remove any carbonate and then washed with distilled water to remove chloride. Approximately 50 mg of carbonate-free bulk sediment was loaded into tin sample capsules for carbon isotopes and placed in a 50-position automated carousel on the elemental analyzer. Nonacidified samples were used for nitrogen isotopes. Combustion gases were carried in a helium stream through a ConFlo II interface to a Thermo Electron DeltaV Advantage isotope ratio mass spectrometer. All carbon and nitrogen isotope results are reported in per mil (‰) and expressed in standard delta notation. Carbon isotopes are reported relative to VPDB while nitrogen isotopes are reported relative to AIR.

Analytical methods

To describe patterns of variation in testate amoeba communities and explore relationships between community composition and environmental variables, we used nonmetric multidimensional scaling (NMDS) (Kruskal, 1964; McCune et al., 2002). NMDS does not make assumptions regarding underlying species distributions along compositional gradients, making it particularly well suited to ecological community data. Outliers and rare taxa can have a large influence on ordination techniques, and to reduce the effect of rare taxa that are likely not well ecologically described in our data set, we excluded those taxa present in less than four modern samples from all analyses. Community data were square-root transformed before ordination analyses, and all analyses were performed using PC-ORD software (McCune and Mefford, 2011)

Transfer functions were developed using a variety of commonly used models, including weighted averaging (weighted average [WA]), partial least squares (PLS), weighted-averaging partial least squares (WA-PLS), and modern analog technique

(MAT). 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; Booth, 2008; Lamentowicz et al., 2008; Amesbury et al., 2016; Swindles et al., 2016). Assessment of transfer function performance was done using standard cross-validation approaches, including bootstrapping and leave-one-site-out procedures, and all transfer function analyses were carried out in the software package C2 (Juggins, 2003).

Standard methods were used to radiocarbon date and examine the paleoecological and geochemical record from the peat core. An age-depth model was built using a Bayesian approach, using 11 radiocarbon dates obtained on bulk sediment samples, the top of the core, and the default priors of Bacon software (Blaauw and Christen, 2011). The composition of subfossil testate amoeba assemblages was described using cluster analysis approaches and the CONISS software package (Grimm, 1987).

RESULTS

Modern ecology of testate amoebae

A total of 59 testate amoeba taxa were identified from 96 modern samples collected within the five peatlands. The most abundant taxa included *Assulina muscorum*, *Euglypha tuberculata* type, *Corythion* spp., *Euglypha strigosa* type, *Pseudodiffugia fulva* type, *Centropyxis aculeata* type, and *Euglypha rotunda* type (Fig. 2). Taxa in less than four samples, including *Apodera vas*, *Microgromia* type, *Hyalospheonia minuta*, *Netzelia oviformis*, and likely *Nebela* species, were excluded from further quantitative analyses. Páramo testate amoebae included many that are commonly found in temperate and boreal regions of the Northern and Southern Hemispheres, although taxa restricted to the Southern

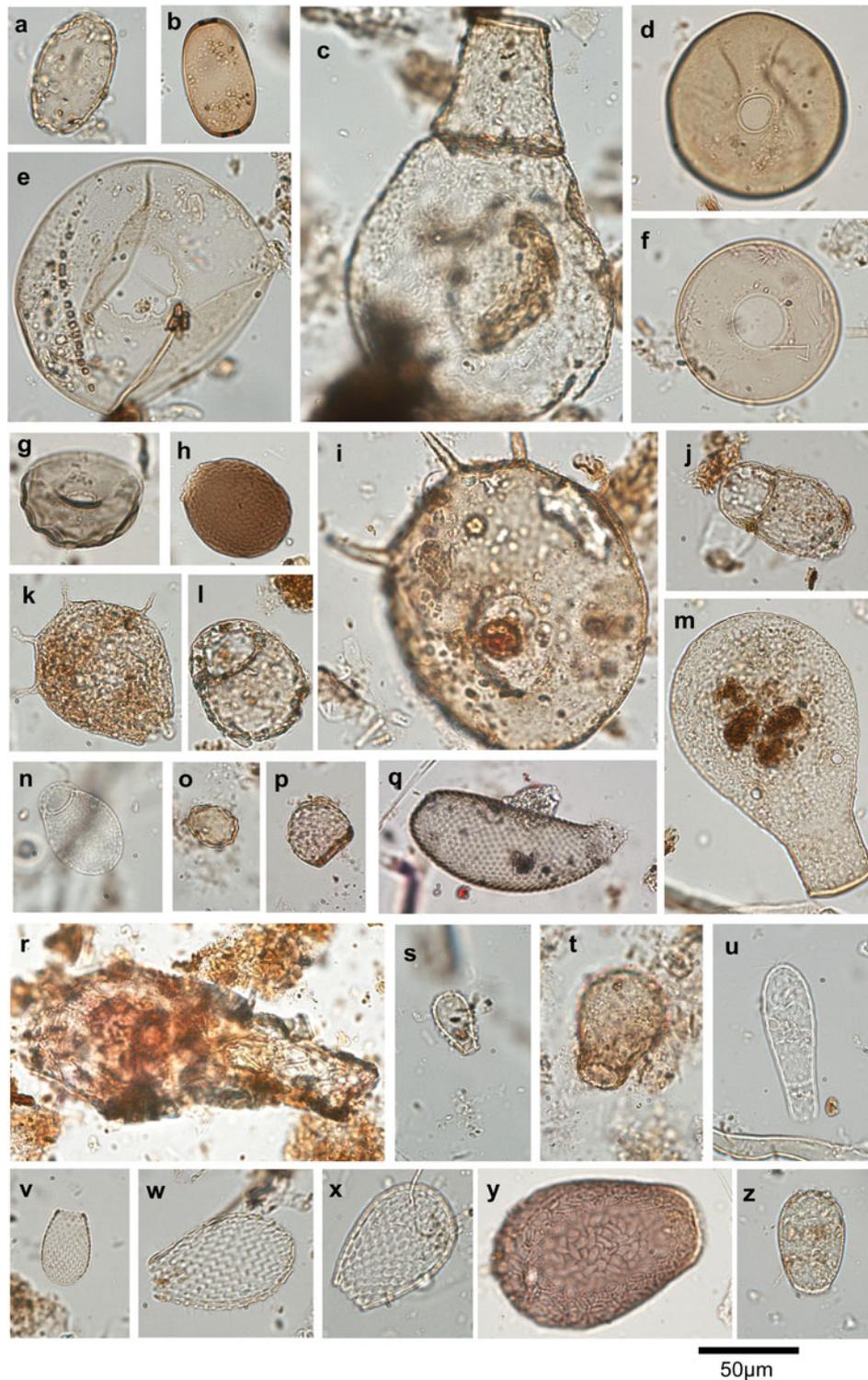


Figure 2. (color online) Photomicrographs of abundant and unusual testate amoebae in the modern samples from the Colombian páramo. (a) *Amphitrema wrightianum/stenostoma* type, (b) *Archellera flavum*, (c) *Apodera vas* type, (d) *Arcella catinus* type, (e) *Arcella crenulata* type, (f) *Arcella discoides* type, (g) *Arcella hemispherica* type, (h) *Assulina muscorum* type, (i) *Centropyxis aculeata* type, (j) *Centropyxis platystoma* type, (k) *Argynnia caudata* type, (l) *Centropyxis cassis* type, (m) *Certesella martiali*, (n) *Corythion* spp., (o) *Cryptodiffugia crenulata* type, (p) *Cyclopyxis arcelloides* type, (q) *Cyphodera ampulla*, (r) *Diffugia oblonga* type, (s) *Diffugia pulex* type, (t) *Diffugia rubescens* type, (u) *Euglypha cristata* type, (v) *Euglypha rotunda* type, (w) *Euglypha strigosa* type, (x) *Euglypha tuberculata* type, (y) *Heleopera rosea* type, (z) *Heleopera sylvatica*, (aa) *Heleopera sphagni-petricola* type, (bb) *Lesquereusia spiralis* type, (cc) *Nebela collaris-bohemica* type, (dd) *Nebela galeata* type, (ee) *Nebela militaris*, (ff) *Nebela parvula*, (gg) *Nebela tincta*, (hh) *Nebela penardiana* type, (ii) *Nebela wailesi* type, (jj) *Paraquadrula irregularis* type, (kk) *Pseudodiffugia fulva* type, (ll) *Plagiopyxis labiata* type, (mm) *Placocista spinosa*, (nn) *Quadrueella-Apodera* type, (oo) *Quadrueella symmetrica* type, (pp) *Tracheuglypha dentata* type, (qq) *Quadrueella wailesi* type.

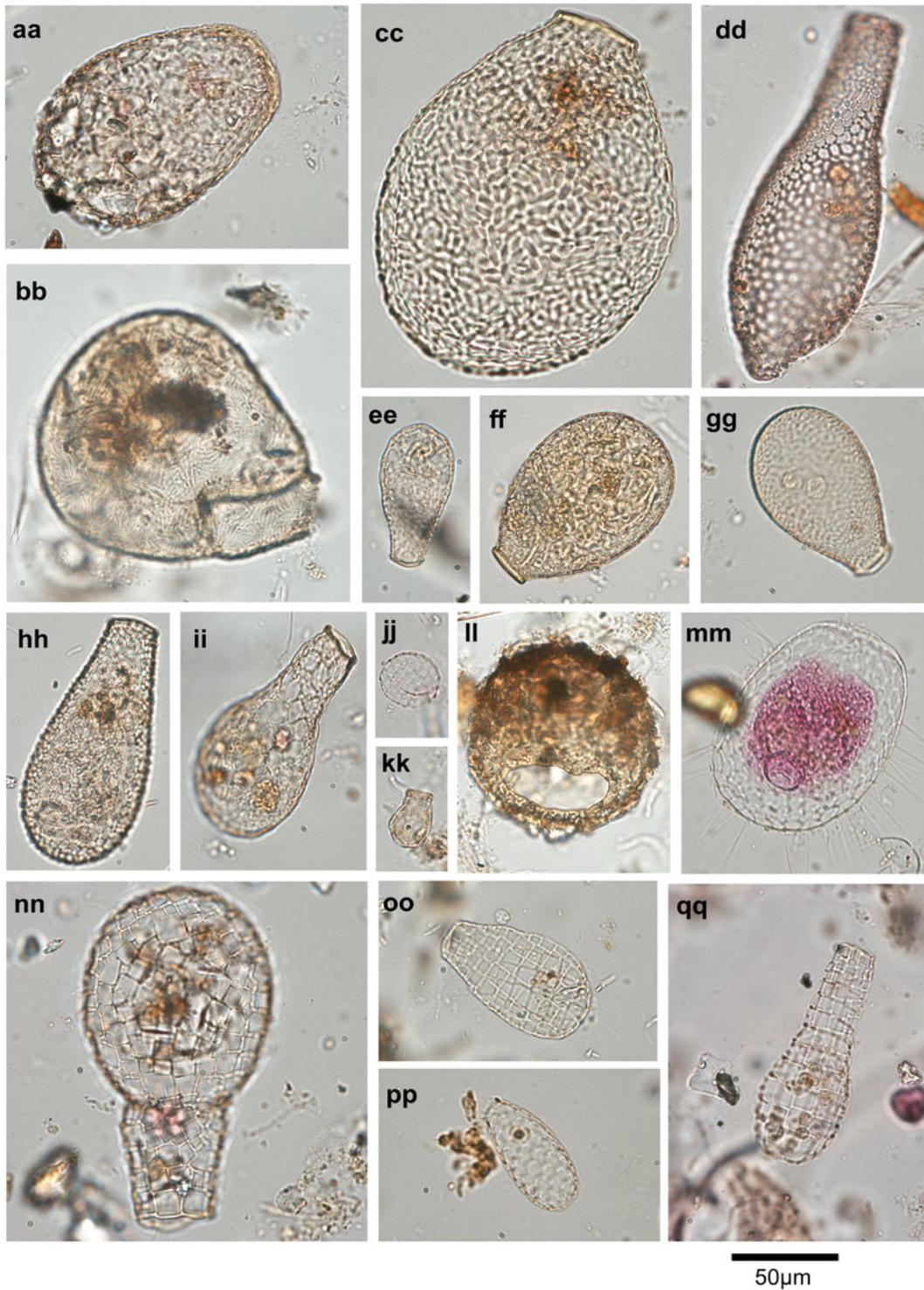


Figure 2. (color online) Continued.

Hemisphere were also encountered (e.g., *Certesella martiali*, *Apodera vas*), as well as a few unusual taxa (Fig. 2, Table 2). A two-dimensional NMDS ordination (final stress = 15.5) represented about 87% of the compositional variability in the data set and revealed considerable overlap in testate amoeba community composition among the five peatlands (Fig. 3). The primary and secondary compositional gradients,

as revealed by axis 1 and axis 2 of the NMDS ordination, represented 70% and 17% of the variability in the data set, respectively (Fig. 3).

Correlations among the NMDS axes and measured environmental variables indicated significant relationships between community composition and both pH and water-table depth (Fig. 3). For the samples for which pH was measured ($n = 43$)

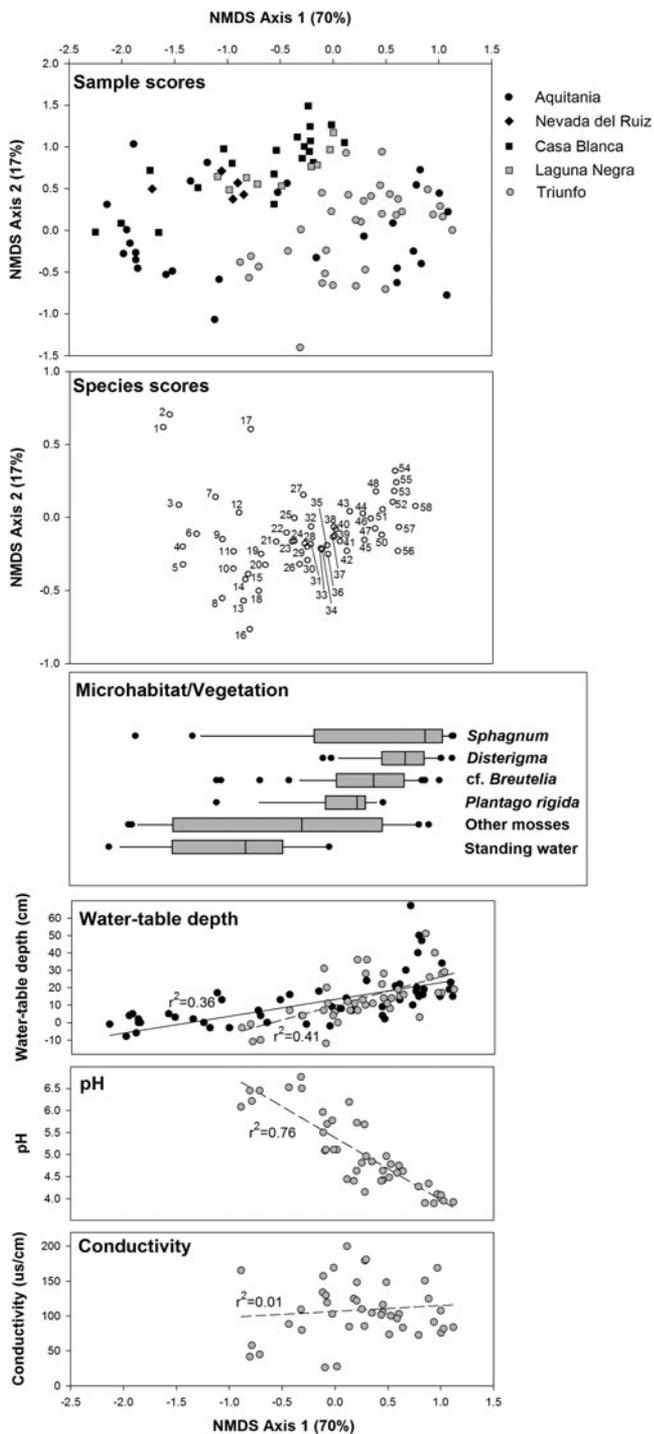


Figure 3. Nonmetric multidimensional ordination (NMDS) of modern testate amoeba communities of the Colombian páramo, showing the distributions of samples and species in the top two panels (species numbers are defined in Table 2). Axis 1 relationships with habitat and environmental variables are shown in the bottom three panels. Presence/absence data for vegetation and standing water are summarized with box plots in the center panel, showing the full range of axis 1 scores for samples (dots), 75% range (whiskers), 25% range (boxes), and median. The main gradient of testate amoeba compositional change is correlated with both pH and water-table depth.

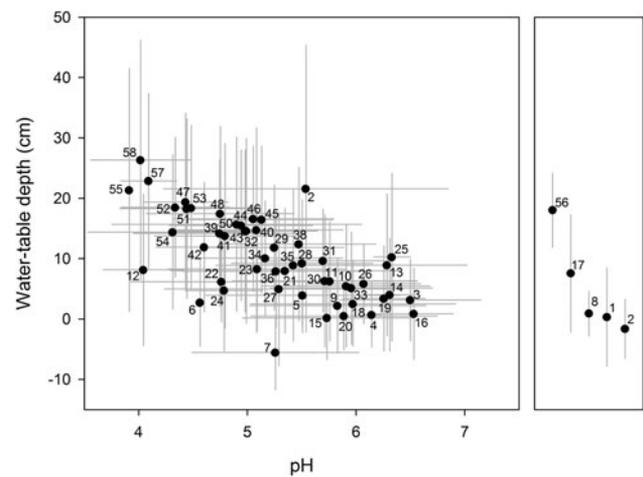


Figure 4. Water-table and pH optima (dots) and tolerances (gray lines) for testate amoeba taxa. Numbers for taxa are defined in Table 2 and correspond to the position of each taxon along the axis 1 community gradient shown in Figure 3. The five taxa on the right were not encountered at sites where pH was measured.

the correlation with NMDS axis 1 was particularly strong ($r^2 = 0.76$, $p < 0.0001$). When the same reduced data set was used, correlations between water-table depth and NMDS axis 1 were also significant but weaker ($r^2 = 0.34$, $p < 0.0001$), and a similar correlation between NMDS axis 1 and water-table depth was found using the entire data set ($r^2 = 0.35$, $p < 0.0001$). No significant correlation was found between conductivity and NMDS axis scores. Water-table depth and pH measurements were also correlated with each other, with drier habitats tending to be more acidic and wetter habitats generally more circumneutral ($r^2 = 0.41$, $p < 0.0001$). Microhabitats of the samples, as represented by presence/absence data for dominant plant groups and standing water, also tended to array themselves along axis 1 according to the pH and water-table gradient (Fig. 3). Optimal water-table depth and pH for individual testate amoeba taxa further highlight the correlation between water-table depth and pH, with taxa such as *Argynnia caudata* type, *Nebela militaris*, and *Plagiopyxis labiata* type more abundant on drier, acidic substrates, and taxa such as *Quadruella wailesi* type, *Diffflugia rubescens* type, and *Paraquadrula irregularis* more common in wetter, more circumneutral habitats (Fig. 4).

Transfer function development and cross-validation

Because pH and water-table depth showed significant relationships with testate amoeba community composition, transfer functions for these variables were developed and performance was assessed through bootstrapped cross-validation ($n = 1000$). Performance among the models was relatively similar (Table 3), and therefore we used the commonly applied WA-PLS (two-component) model for all subsequent validation and reconstructions (Fig. 5). Water-table depth and

Table 3. Cross-validation statistics showing performance of transfer functions for water-table depth and pH using a range of commonly applied transfer function models. The bootstrapped ($n = 1000$) root-mean-square error of prediction (RMSEP) and r^2 values indicate similar performance of the models. Values in parentheses provide statistics for water-table depth using only the samples with pH measurements.

Model	n	r^2_{boot}	RMSEP _{boot}
Water-table depth			
Weighted average (WA)	96 (43)	0.41 (0.39)	11.0 (10.9)
Weighted average–tolerance down weighted (WA-tol)	96 (43)	0.44 (0.43)	11.0 (11.0)
Weighted-average partial least squares (WA-PLS, two-component)	96 (43)	0.46 (0.37)	10.9 (11.6)
Modern analog technique (MAT)	96 (43)	0.57 (0.30)	10.3 (12.1)
pH			
Weighted average (WA)	43	0.82	0.39
Weighted average–tolerance down weighted (WA-tol)	43	0.70	0.59
Weighted-average partial least squares (WA-PLS, two-component)	43	0.83	0.38
Modern analog technique (MAT)	43	0.77	0.50

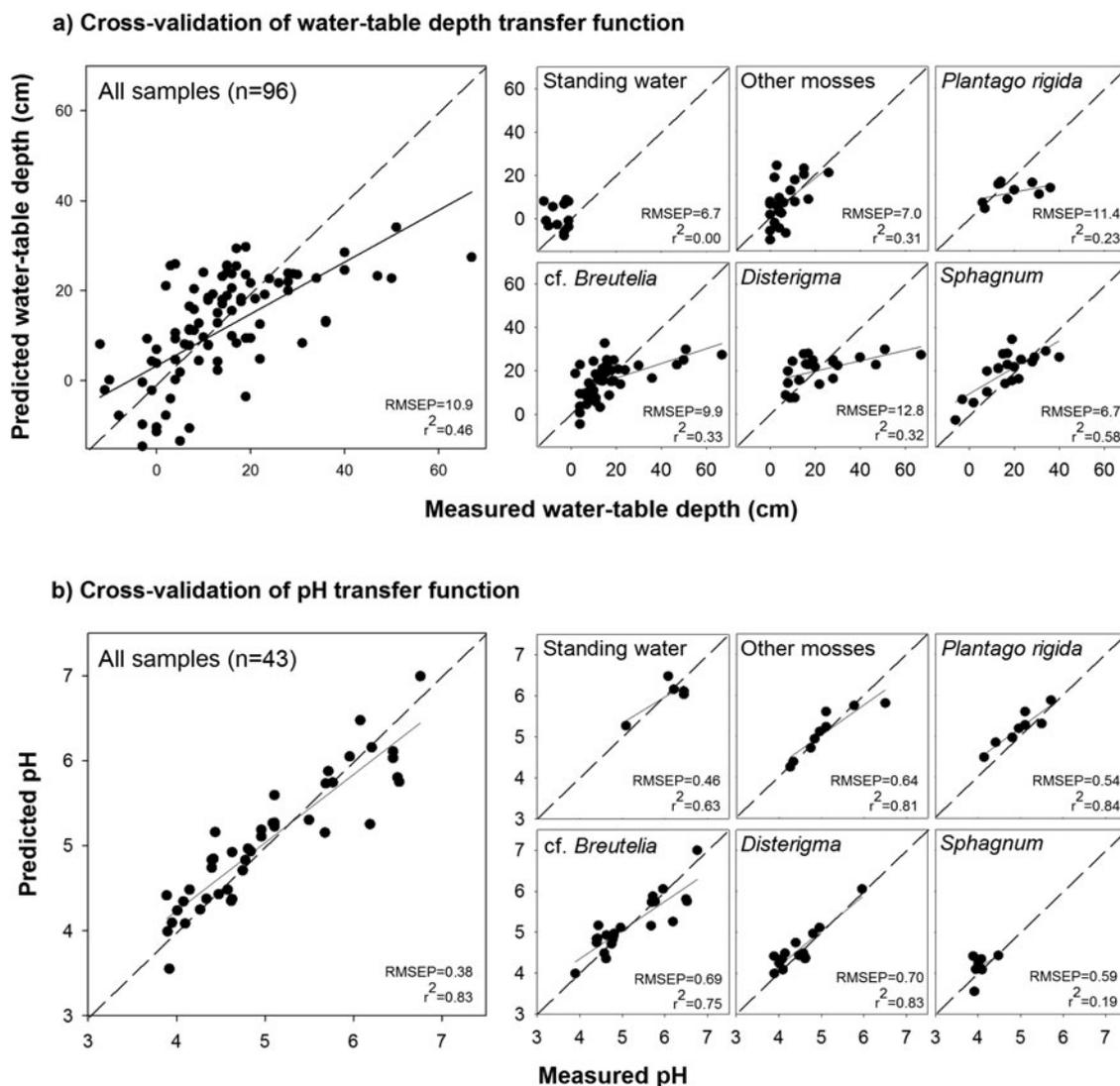


Figure 5. Bootstrapped ($n = 1000$) cross-validation for transfer functions for (a) water-table depth and (b) pH, comparing measured and predicted values to demonstrate the ability to estimate these variables from testate amoeba community composition. Cross-validation for transfer functions developed independently for each vegetation/habitat grouping are also shown to the right. The transfer function for pH outperforms water-table depth, except for within *Sphagnum* samples. RMSEP, root-mean-square error of prediction.

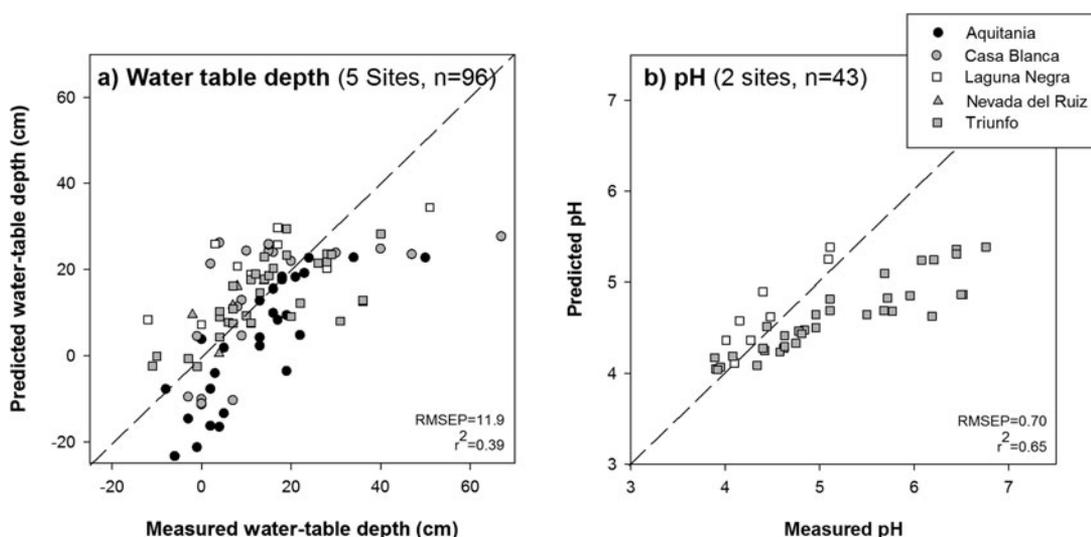


Figure 6. Leave one-site-out cross-validation of transfer functions for (a) water-table depth and (b) pH to assess the potential impact of spatial autocorrelation on performance. RMSEP, root-mean-square error of prediction.

pH were both successfully estimated from testate amoeba community composition, although cross-validation for pH indicated better performance for this variable (Fig. 5). To examine transfer function performance as a function of microhabitat, we also performed transfer function development and cross-validation within each vegetation/microhabitat grouping. Interestingly, within *Sphagnum* microhabitats, transfer function performance for water-table depth was better than performance for pH, while pH transfer functions outperformed water-table depth transfer functions in all other microhabitats (Fig. 5).

In addition, because spatial autocorrelation can lead to unrealistic estimates of uncertainty (e.g., Payne et al., 2012), we examined the results of sequentially leaving each site out of the transfer function model and using the resulting models to infer the environmental variables at each omitted site (Fig. 6). For water-table depth, uncertainty estimates were only slightly larger using this approach (Fig. 6a). The effects of spatial autocorrelation were difficult to fully assess for pH, because there were only two sites where pH measurements were obtained, and one of these sites (Laguna Negra) only captured about half of the pH gradient recorded at the other (Triunfo) (Fig. 6b).

Paleoecological record and transfer function application

The peat core was composed of fibric peat interbedded with ash and lapilli layers (Cardona and Monroy, 2015). An age-depth model developed from 11 ^{14}C dates (Table 4) and the surface of the peat core indicates average deposition times of ~ 7.5 yr/cm, and accumulation spanning the past 5000 years (Fig. 7). Testate amoebae were analyzed to a depth of 580 cm, or approximately the past 4300 yr, below which sufficient count totals were not obtainable. Low count totals

prevented analyses at some depths between 500 and 580 cm as well. A total of 53 testate amoeba taxa were identified from the core analyses, with a few major changes in community composition over the duration of the record, particularly in the lower 100 cm, as well as long intervals of relatively stable community composition (Fig. 8). Some dominant taxa included *Cyclopyxis arcelloides* type, *Centropyxis aculeata* type, *Centropyxis platystoma* type, *Assulina muscorum*, *Pseudodiffugia fulva* type, *Centropyxis cassis* type, and *Nebela penardiana* (Fig. 8).

Four biostratigraphic zones were identified using the guidance provided by stratigraphically constrained cluster analysis to facilitate discussion of the major changes in the record. Zone 1 (~ 4300 – 3200 cal yr BP) contained exceptionally variable testate amoeba communities, with abundant *Assulina muscorum* and *Cyclopyxis arcelloides* type in the lowermost samples, abundant *Arcella catinus* type in samples in the middle of the zone, and very abundant *Centropyxis aculeata* type in the upper two samples of the zone. Some *Arcella catinus* type individuals were large enough to be attributable to *Arcella artocrea*, although considerable numbers were of intermediate size between the typical size ranges of *Arcella catinus* and *Arcella artocrea*. Communities of Zone 2 (~ 3200 – 1100 cal yr BP) and Zone 3 (~ 1200 – 250 cal yr BP) had higher diversity than Zone 1 (~ 250 cal yr BP to present), with variable amounts of *Cyclopyxis arcelloides* type, *Centropyxis* spp., *Assulina muscorum*, and *Pseudodiffugia fulva* type characterizing the two zones. *Trinema enchelys* type increased in Zone 4, along with corresponding declines in other taxa, particularly *Pseudodiffugia fulva* type (Fig. 8).

Reconstructed pH and water-table depths based on changes in testate amoeba community composition were similar to each other, which was not surprising, given the correlation between pH and water-table depth in the modern samples (Fig. 9). Inferred environmental conditions from Zone 1 samples were the driest and most acidic of the reconstruction, with

Table 4. Radiocarbon dates and sample-specific information from the Triunfo peat core. Calibrated age is the date with highest probability followed by the 2-sigma range. Radiocarbon dates were calibrated using Calib 7.1 software and IntCal13 (Stuiver et al. 2018).

Depth (cm)	Material dated	Lab number	Radiocarbon age	$^{13}\text{C}/^{12}\text{C}$ ratio (‰)	Calibrated age (yr BP)
54	Bulk peat	Beta-368996	90 ± 30	-27.0	108 (22–265)
100	Bulk peat	Beta-330145	410 ± 30	-26.1	483 (330–519)
119	Bulk peat	Beta-368997	480 ± 30	-27.3	520 (499–542)
154	Bulk peat	Beta-368998	570 ± 30	-26.5	601 (527–646)
188	Bulk peat	Beta-369001	730 ± 30	-26.1	677 (653–726)
208	Bulk peat	Beta-368999	830 ± 30	-26.5	737 (688–789)
284	Bulk peat	Beta-369000	1070 ± 30	-26.7	974 (929–1054)
353	Bulk peat	Beta-369002	1580 ± 30	-27.0	1469 (1404–1540)
384	Bulk peat	Beta-369003	1810 ± 30	-26.2	1750 (1628–1822)
578	Bulk peat	Beta-369004	3970 ± 30	-27.1	4444 (4299–4523)
700	Bulk peat	Beta-330146	4430 ± 40	-26.1	5027 (4871–5279)

the exception of the upper two samples dominated by *Centro-pyxis aculeata* type, which had wet and circumneutral pH estimates. Zone 2 reconstructions were less variable than those of Zone 4, but contained both dry/acidic and wet/circumneutral estimates. Most water-table and pH estimates from Zone 3 samples were drier than average and slightly acidic. Wetter and more neutral pH conditions occurred within Zone 4. The pH and water-table depth reconstructions show a correspondence with C/N measurements from the same samples, at least for much of the record, but less association with carbon and nitrogen isotope measurements (Fig. 9). For example, the coherence between the C/N measurements and the reconstructions is relatively good between

about 1200 and 4300 cal yr BP ($n = 34$, $r = -0.70$); however, the correspondence is weak from about 1200 to 250 cal yr BP. Interestingly, during this time period of lower overall correspondence between testate amoeba-inferred estimates and C/N measurements, the two largest dry/acidic anomalies are associated with the two most positive $\delta^{13}\text{C}$ values (Fig. 9). Little obvious relationship between $\delta^{15}\text{N}$ values and testate amoeba-inferred conditions exists for most the record, although similar trends occur in the last 200 yr (Fig. 9).

Some correspondence exists between the reconstructions and the pollen-based interpretation of temperature and moisture changes for the past 1900 yr (Fig. 9), although the testate amoeba data suggest that not all pollen-inferred warmer time periods were necessarily drier (Giraldo-Giraldo et al., 2017). Both pollen and testate amoebae suggest that a shift to more persistently wet conditions occurred at about 1300 cal yr BP, and this was followed by dry conditions after about 1000 cal yr BP (Fig. 9). However, although pollen data suggest cooler and perhaps wetter conditions from about 700 to 400 cal yr BP, the testate amoeba record suggests moderately dry and slightly acidic conditions on the peatland surface (Fig. 9). Testate amoeba data also suggest that the warming during the past century has been associated with somewhat wetter and less acidic conditions.

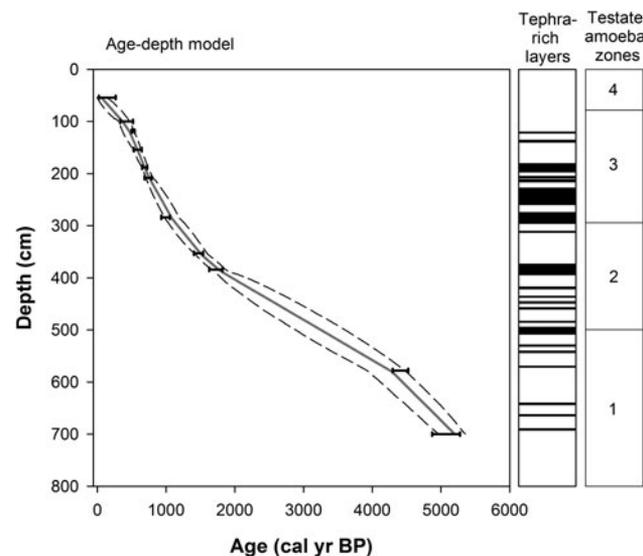


Figure 7. Age–depth model for the Triunfo peatland core, with the gray line indicating the model with highest probability and the dashed lines indicating the 95% confidence interval. Black vertical lines show the 2-sigma range of calibrated radiocarbon dates used in the model. The distribution of tephra-rich layers and the testate amoeba zonation from Figure 8 is shown to the right.

DISCUSSION

Testate amoeba ecology and potential for environmental inference

Although testate amoebae have been widely used as surface-moisture proxies in north-temperate and boreal peatlands, the ecology of testate amoebae in tropical peatlands is comparably less well understood. In tropical South America, recent work in the lowland Amazon basin demonstrated good preservation potential and the sensitivity of community composition to surface-moisture conditions (Swindles et al., 2014,

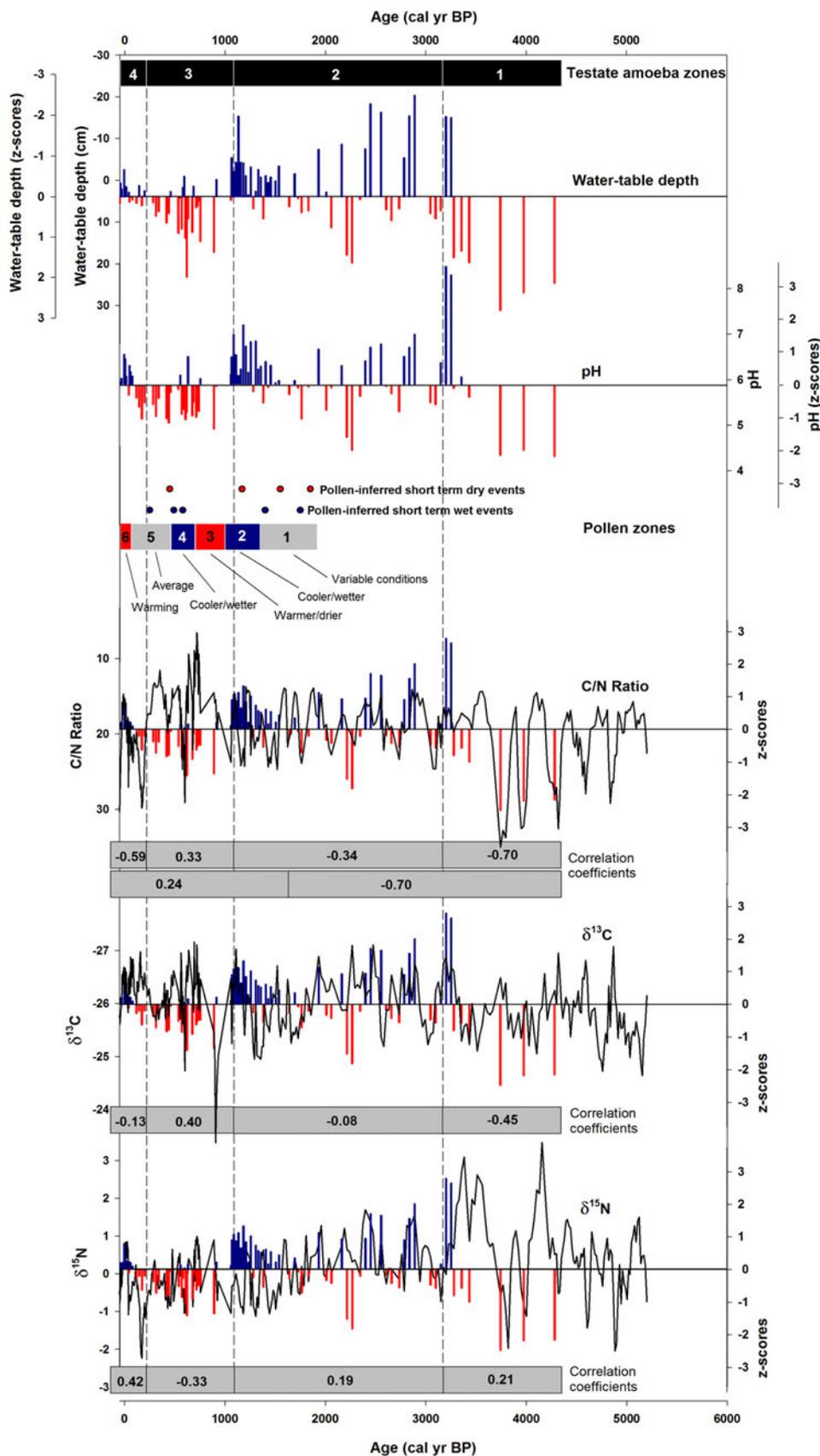


Figure 9. (color online) Comparison of water-table depth and pH reconstructions based on testate amoebae, pollen, and geochemical measurements from the peat core, including C/N ratios, $\delta^{13}\text{C}$, and $\delta^{15}\text{N}$. Correlation coefficients between testate amoeba-inferred paleoenvironmental conditions (average z-scores of pH and water-table depth reconstructions) and the geochemical measurements are shown for each zone.

gradient, with similar estimates of water-table depth for samples with measured water-table depths greater than about 30 cm from the peatland surface (Fig. 5a). Interestingly, pH estimates do not show the same degree of decreased performance on the circumneutral side of the gradient, although performance is still reduced (Fig. 5b). pH may better reflect the annual microenvironment of the peatland surface experienced by testate amoebae at these drier sites. However, the patterns indicate that water-table depth reconstructions deeper than a threshold of about 30 cm do not necessarily indicate increasingly dry conditions at the surface, and interpretation of fossil reconstructions should reflect this decreased performance on the dry side of the gradient.

Paleoenvironmental reconstruction at Triunfo peatland

Pollen analyses on the Triunfo core have been performed on the last 1900 yr of the record, and reveal that the site has been surrounded by páramo vegetation during this entire time and that the local wetland vegetation has been dominated by many of the same taxa that are present today (Giraldo-Giraldo et al., 2018). Although the relative abundance of wetland pollen taxa has been used to qualitatively discuss potential moisture changes, our reconstructions provide more direct estimates of pH and hydrology. Water-table depth and pH reconstructions based on the testate amoeba record indicate changes between standing water and subsurface water tables throughout much of the past 4300 yr. Before 3200 cal yr BP, reconstructions indicate dry and acidic conditions at the coring site. An abrupt increase in water levels and associated increase in pH occurred at 3200 cal yr BP, and then variable conditions occurred from 3200 cal yr BP until about 1000 cal yr BP. Drier and more acidic conditions became more common after 1000 cal yr BP, a pattern also evident in the pollen data (Fig. 9). Over the past century or two, when pollen data indicate a warming trend, environmental conditions at the surface of the peatland have generally become wetter and less acidic. These changes in both the mean state and variability of the reconstructions correspond with the broad changes in testate amoeba community composition reflected in the stratigraphic zones (Fig. 9).

The correlation between testate amoeba-based environmental reconstructions and the C/N ratio of bulk peat samples from 4300 to 1200 cal yr BP provides additional support for the testate amoeba-based paleohydrologic inferences during this time window (Fig. 9). Given that water-table depth reconstructions indicate shifts between standing water and subsurface water levels, the relative contribution of terrestrial and algal-derived organic matter to the sediments would be expected to change during these water-level changes. In lake ecosystems, C/N ratios greater than or less than 15 are often interpreted as reflecting higher inputs of terrestrial- or algal-derived organic matter, respectively (Meyers and Ishiwatari, 1993). Although carbon inputs from wetland plants

would have continued at Triunfo even during periods of standing water, the relative contribution of algal carbon would likely have increased during wetter time periods with standing water. Algae and biofilms are very abundant in the pools on the surface of the peatland today. However, little correlation exists between C/N and the testate amoeba reconstruction during the interval between 1200 and 250 cal yr BP. If the testate amoeba reconstruction is correct, this time period was associated with mostly dry and acidic conditions, and the lack of any standing water for long periods of time would have limited algal carbon contributions. Surprisingly, however, the C/N ratios are the lowest of the record and are also quite variable. The time period from about 1200 to 500 cal yr BP contained abundant tephra, including a series of closely spaced, thick tephra deposits (Fig. 6). How ecological and biogeochemical dynamics would have been impacted by repeated deposition of large amounts of tephra is unclear, but abundant tephra-derived minerals like silica may have stimulated algal productivity, decreased terrestrial-derived carbon from wetland plants via disturbance, or impacted testate amoebae. Despite a few studies in northern peatlands (Payne and Blackford, 2008; Hughes et al., 2013), impacts of tephra deposition are still not well understood.

More research is needed to assess the relationship between climatic variability and páramo peatland hydrology, as well as the potential role of nonclimatic effects on hydrology. However, regional-scale climate changes likely drove the largest-magnitude fluctuations in the Triunfo reconstruction. For example, the nearest high-resolution hydrologic reconstruction is from Lake Ubaque (2070 m asl) which is located in the Eastern Cordillera about 160 km east of the Triunfo peatland. C/N ratios and percent sand in this record have been interpreted as reflecting frontal slope hydroclimatic variability along the Eastern Cordillera (Bird et al., 2018). The record contains an abrupt shift from drier to wetter conditions at about 3200 cal BP, very similar to the major transition in testate amoeba communities and inferred environmental conditions at Triunfo (Fig. 10). Furthermore, although the regions might be expected to record different climates at some time scales (Marchant et al., 2001; Bird et al., 2018), both records suggest some of the wettest time periods of the late Holocene between about 3200 and 2200 cal BP and drier conditions after this time (Fig. 10). In the last 200 yr, both sites also have experienced a wetting trend.

In summary, our research results attest to the potential of using testate amoebae in peatlands of the páramo to reconstruct surface-moisture and pH changes. Testate amoebae are of particular value to high-resolution studies because their short generation time and rapid response to environmental change (e.g., Koenig et al., 2017) make them highly sensitive to decadal-scale changes in peatland hydrology (e.g., Booth, 2010). Although testate amoebae are widely used as proxies in north-temperate and boreal ecosystems, our work demonstrates that testate amoebae are useful paleohydrologic indicators in the páramo, where they are well preserved in

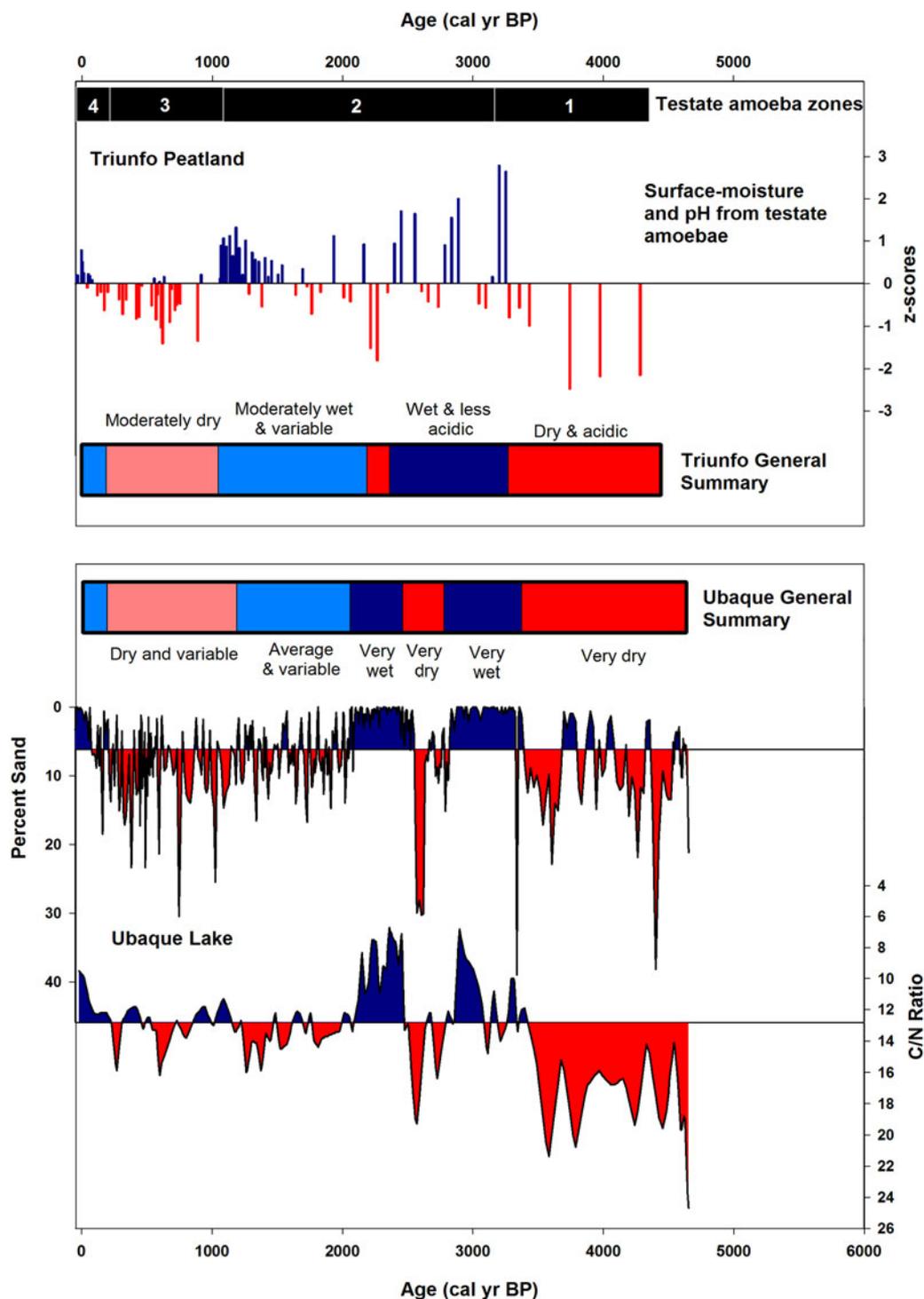


Figure 10. (color online) Comparison of the Triunfo peatland testate amoeba reconstruction with a lake-level record from Lake Ubaque (Bird et al., 2018). All proxies are arranged so that downward changes are indicative of drier conditions. Conditions wetter and drier than the long-term average are shown in blue and red, respectively. Qualitative summaries of the record are also shown for comparison.

peatland deposits. In combination with other recent ecological and paleoecological studies of testate amoebae in lowland tropical peatlands (Swindles et al., 2014, 2016, 2018a, 2018b), our results suggest that testate amoebae could

potentially be applied across a broad elevation gradient to address hydrologic and climatological questions in topographically heterogeneous tropical regions, particularly when used as part of multiproxy studies.

ACKNOWLEDGMENTS

Fieldwork and sample collection costs were supported by a Lehigh University Faculty Grant for International Connections to RKB. BL and ZW were supported during their work on this project by a South China Normal University Grant for Excellent Students Visiting and Studying Abroad, which allowed them to reside at Lehigh University for 6 months. Fieldwork, sample collection, and geochemistry analysis were partially covered by a grant from the Inter-American Institute for Global Change Research (CRN3038) awarded to JE, which was supported by the U.S. National Science Foundation (GEO-1128040). JE also was partially funded by the Canadian Queen Elizabeth II Diamond Jubilee Advanced Scholar Program (QES-AS), a partnership among universities in Canada, the Rideau Hall Foundation, and the Community Foundations of Canada. The QES-AS was made possible with financial support from the International Development Research Centre and the Social Science and Humanities Research Council. All authors thank Mark Brenner, Felipe Velasco, Diego Felipe Vallejo, and Raul Andres Trejo for their help in the field. The article was improved by constructive feedback from Henry Hooghiemstra and Graeme Swindles.

REFERENCES

- Amesbury, M.J., Swindles, G.T., Bobrov, A., Charman, D.J., Holden, J., Lamentowicz, M., Mallon, G., et al., 2016. Development of a new pan-European testate amoeba transfer function for reconstructing peatland palaeohydrology. *Quaternary Science Reviews* 152, 132–151.
- Baker, P.A., Fritz, S.C., 2015. Nature and causes of Quaternary climate variation of tropical South America. *Quaternary Science Reviews* 124, 31–47.
- Bakker, J., Olivera, M.M., Hooghiemstra, H., 2008. Holocene environmental change at the upper forest line in northern Ecuador. *The Holocene* 18, 877–893.
- Baruch, Z., 1984. Ordination and classification of vegetation along an altitudinal gradient in the Venezuelan páramos. *Vegetatio* 55, 115–126.
- Bird, B.W., Rudloff, O., Escobar, J., Gilhooly, W.P., III, Correa-Metrio, A., Vélez, M., Polissar, P.J., 2018. Paleoclimate support for a persistent dry island effect in the Colombian Andes during the last 4700 years. *The Holocene* 28, 217–228.
- Blaauw, M., Christen, J. 2011. Flexible paleoclimate age-depth models using an autoregressive gamma process. *Bayesian Analysis* 6, 457–474.
- Bogota-A, R.G., Groot, M.H.M., Hooghiemstra, H., Lourens, L.J., Van der Linden, M., Berrio, J.C., 2011. Rapid climate change from north Andean Lake Fuquene pollen records driven by obliquity: implications for a basin-wide biostratigraphic zonation for the last 284 ka. *Quaternary Science Reviews* 30, 3321–3337.
- Booth, R.K., 2001. Ecology of testate amoebae (Protozoa) in two Lake Superior coastal wetlands: implications for paleoecology and environmental monitoring. *Wetlands* 21, 564–576.
- Booth, R.K., 2008. Testate amoebae as proxies of mean annual water-table depth in *Sphagnum*-dominated peatlands of North America. *Journal of Quaternary Science* 23, 43–57.
- Booth, R.K., 2010. Testing the climate sensitivity of peat-based paleoclimate reconstructions in mid-continental North America. *Quaternary Science Reviews* 29, 720–731.
- Booth, R.K., Jackson, S.T., Sousa, V.A., Sullivan, M.E., Minckley, T.A., Clifford, M.J., 2012. Multidecadal drought and amplified moisture variability drove rapid forest community change in a humid region. *Ecology* 93, 219–226.
- Booth, R.K., Lamentowicz, M., Charman, D.J., 2010. Preparation and analysis of testate amoebae in peatland paleoenvironmental studies. *Mires and Peat* 7, 1–7.
- Bosman, A.F., Hooghiemstra, H., Cleef, A.M., 1994. Holocene mire development and climatic change from a high Andean *Plantago rigida* cushion mire. *The Holocene* 4, 233–243.
- Buytaert, W., Celleri, R., De Bievre, B., Cisneros, F., Wyseure, G., Deckers, J., Hofstede, R., 2006. Human impact on the hydrology of the Andean Páramos. *Earth-Science Reviews* 79, 53–72.
- Buytaert, W., Cuesta-Camacho, F., Tobón, C., 2011. Potential impacts of climate change on the environmental services of humid tropical alpine regions. *Global Ecology and Biogeography* 20, 19–33.
- Calder, W.J., Shuman, B., 2017. Extensive wildfires, climate change, and an abrupt state change in subalpine ribbon forests, Colorado. *Ecology* 98, 2585–2600.
- Cardona, V., Monroy, D., 2015. Datación, petrografía y morfología de las tefras acumuladas en el registro Holocénico (0–4973 años) del humedal El Triunfo (Parque Nacional Los Nevados). Undergraduate honors thesis, Universidad de Caldas, Manizales, Colombia. 76 p.
- Chardez, D., 1969. Le genre *Phryganella* Penard (Protozoa, Rhizopoda, Testacea). *Bulletin de la Station de Recherche Agronomique de Gembloux* 4, 315–322.
- Charman, D.J., 1997. Modelling hydrological relationships of testate amoebae (Protozoa: Rhizopoda) on New Zealand peatlands. *Journal of the Royal Society of New Zealand* 27, 465–483.
- Charman, D.J., 2001. Biostratigraphic and paleoenvironmental applications of testate amoebae. *Quaternary Science Reviews* 20, 1753–1764.
- Charman, D.J., Hendon, D., Woodland, W.A., 2000. *The Identification of Testate Amoebae (Protozoa: Rhizopoda) in Peats*. Quaternary Research Association, London.
- Chen, F., Yu, Z., Yang, M., Ito, E., Wang, S., Madsen, D.B., Huang, X., et al., 2008. Holocene moisture evolution in arid central Asia and its out-of-phase relationship with Asian monsoon history. *Quaternary Science Reviews* 27, 351–364.
- Cincotta, R.P., Wisniewski, J., Engelman, R., 2000. Human population in the biodiversity hotspots. *Nature* 404, 990–992.
- Cleef, A.M., 1981. *The Vegetation of the Páramos of the Colombian Cordillera Oriental*. Dissertationes Botanicae 61. J. Cramer, Vaduz, Liechtenstein.
- Clifford, M.J., Booth, R.K., 2015. Late-Holocene drought and fire drove a widespread change in forest community composition in eastern North America. *The Holocene* 25, 1102–1110.
- Deflandre, G., 1928. Le genre *Arcella* Ehrenberg. Morphologie-Biologie. Essai phylogénétique et systématique. *Archiv für Protistenkunde* 64, 152–287.
- Deflandre, G., 1932. *Paraquadrula* nov. gen. irregularis (Archer). *Conjugaison et enkystement: Comptes rendus hebdomadaires des Séances de la Société de Biologie et de ses filiales et associées (Paris)*, 109, 1346–1347.
- Ehrenberg, G.C., 1840. Über die Bildung der Kreidefelsen und des Kreidemergels durch unsichtbare Organismen. *Abhandlungen der Königliche Akademie der Wissenschaften zu Berlin (1838)*, pp. 59–147.
- Flantua, S., Hooghiemstra, H., Vuille, M., Behling, H., Carson, J.F., Gosling, W. D., Hoyos, I., et al., 2016. Climate variability and human impact in South America during the last 2000 years: synthesis and perspectives from pollen records. *Climate of the Past* 12, 483–523.

- Giraldo-Giraldo, M.J., Velásquez-Ruiz, C.A., Pardo-Trujillo, A., 2017. Late-Holocene pollen-based paleoenvironmental reconstruction of the El Triunfo wetland, Los Nevados National Park (Central Cordillera of Colombia). *The Holocene* 28, 183–194.
- Gonzalez, H., 2001. *Geología de las Planchas 206 Manizales y 225 Nevado del Ruiz*. Ministerio de Minas y Energía, INGEOMINAS, Bogotá D.C., Colombia. 93 p.
- González-Carranza, Z., Hooghiemstra, H., Vélez, M.I., 2012. Major altitudinal shifts in Andean vegetation on the Amazonian flank show temporary loss of biota in the Holocene. *The Holocene* 22, 1227–1241.
- Grabandt, R., 1990. Presence and ecological range of some algae, Testacea and fungal remains in moss samples of Colombian páramo vegetation. In: Rabassa, J., Salemme, M. (Eds.), *Quaternary of South America and Antarctic Peninsula*, Vol. 8. Balkema, Rotterdam, pp. 85–105.
- Grimm, E.C., 1987. CONISS: a Fortran 77 program for stratigraphically constrained cluster analysis by the method of incremental sum of squares. *Computers and Geosciences* 13, 13–35.
- Herd, D., 1982. *Glacial and Volcanic Geology of the Ruiz—Tolima Volcanic Complex, Cordillera Central, Colombia*. Instituto Nacional de Investigaciones Geológico-Mineras Pres, Bogotá, Colombia. 48 p.
- Hughes, P.D.M., Mallon, G., Brown, A., Essex, H.J., Stanford, J.D., Hotes, S., 2013. The impact of high tephra loading on late-Holocene carbon accumulation and vegetation succession in peatland communities. *Quaternary Science Reviews* 67, 160–175.
- Instituto Alexander Von Humboldt, 2012. Identificación cartográfica de los páramos de Colombia a escala 1:100.000 (versión a junio de 2012). Proyecto: Actualización del Atlas de Páramos de Colombia. Convenio Interadministrativo de Asociación 11-103, Instituto de investigación de recursos biológicos Alexander von Humboldt y Ministerio de Ambiente y Desarrollo Sostenible, Bogotá D.C., Colombia.
- Juggins, S., 2003. *C2 Software for Ecological and Palaeoecological Data Analysis and Visualization*. University of Newcastle, Newcastle-upon-Tyne, UK.
- Juggins, S., 2013. Quantitative reconstructions in palaeolimnology: new paradigm or sick science? *Quaternary Science Reviews* 64, 20–32.
- Kerr, A.C., Tarney, J., 2005. Tectonic evolution of the Caribbean and northwestern South America: the case for accretion of two Late Cretaceous oceanic plateaus. *Geology* 33, 269–272.
- Koenig, I., Schwendener, F., Mulot, M., Mitchell, E.A.D., 2017. Response of *Sphagnum* testate amoebae to drainage, subsequent re-wetting and associated changes in the moss carpet: results from a three year mesocosm experiment. *Acta Protozoologica* 56, 191–210.
- Kummu, M., Varis, O., 2010. The world by latitudes: a global analysis of human population, development level and environment across the north–south axis over the past half century. *Applied Geography* 31, 495–507.
- Kruskal, J.B., 1964. Nonmetric multidimensional scaling: a numerical method. *Psychometrika* 29, 115–129.
- Lamarre, A., Magnan, G., Garneau, M., Boucher, É., 2013. A testate amoeba-based transfer function for paleohydrological reconstruction from boreal and subarctic peatlands in northeastern Canada. *Quaternary International* 306, 88–96.
- Lamentowicz, Ł., Lamentowicz, M., Gąbka, M., 2008. Testate amoebae ecology and a local transfer function from a peatland in western Poland. *Wetlands* 28, 164–175.
- Lamentowicz, M., Mitchell, E.A.D., 2005. The ecology of testate amoebae (Protists) in *Sphagnum* in north-western Poland in relation to peatland ecology. *Microbial Ecology* 50, 48–63.
- Leidy, J., 1879. *Fresh-water Rhizopods of North America*. Report of the United States Geological Survey of the Territories, Vol. 12. U.S. Government Printing Office, Washington, DC. 324 p.
- Li, H., Wang, S., Zhao, H., Wang, M., 2015. A testate amoebae transfer function from *Sphagnum*-dominated peatlands in the Lesser Khingan Mountains, NE China. *Journal of Paleolimnology* 54, 189–203.
- Loeblich, A.R., Jr., Tappan, H., 1961. Remarks on the systematics of the Sarkodina (Protozoa) renamed homonyms and new and validated genera. *Proceedings of the Biological Society of Washington* 74, 213–234.
- Luteyn, J.L., 1999. Páramos: A checklist of plant diversity, geographical distribution, and botanical literature. *Memoirs of the New York Botanical Garden*, 84. 278 p.
- Marchant, R., Behling, H., Berrio, J.C., Cleef, A., Duivenvoorden, J., Hooghiemstra, H., Kuhry, P., et al., 2001. Mid- to Late-Holocene pollen-based biome reconstructions for Colombia. *Quaternary Science Reviews* 20, 1289–1308.
- Markel, E., Booth, R.K., Qin, Y., 2010. Testate amoebae and $\delta^{13}\text{C}$ of *Sphagnum* as surface-moisture proxies in Alaskan peatlands. *The Holocene* 20, 463–475.
- Marlon, J.R., Bartlein, P.J., Daniau, A.-L., Harrison, S.P., Maezumi, S.Y., Power, M.J., Tinner, W., Vannié, B., 2013. Global biomass burning: a synthesis and review of Holocene paleofire records and their controls. *Quaternary Science Reviews* 65, 5–25.
- Marlon, J.R., Pederson, N., Nolan, C., Goring, S., Shuman, B., Booth, R., Bartlein, P., et al., 2017. Temperature and hydroclimatic histories of the northeastern United States during the past 3000 years and their ecological implications. *Climate of the Past* 13, 1355–1379.
- Martinez, J.I., Obrochta, S., Yokoyama, Y., Battarbee, R.W., 2015. Atlantic Multidecadal Oscillation (AMO) forcing on the late Holocene Cauca paleolake dynamics, northern Andes of Colombia. *Climate of the Past* 11, 2649–2664.
- McCune, B., Grace, J.B., Urban, D.L., 2002. *Analysis of Ecological Communities*. MjM Software Design, Gleneden Beach, OR.
- McCune, B., Mefford, M.J., 2011. *PC-ORD. Multivariate Analysis of Ecological Data*. Version 6 [computer software]. MjM Software, Gleneden Beach, OR.
- Mesa, O., Poveda, G., Carvajal, L., 1997. *Introducción al clima de Colombia*. Universidad Nacional de Colombia, Bogotá, Colombia. 390 p.
- Meyers, P.A., Ishiwatari, R., 1993. Lacustrine organic geochemistry—an overview of indicators of organic matter sources and diagenesis in lake sediments. *Organic Geochemistry* 20, 867–900.
- Mitchell, E.A.D., Charman, D.J., Warner, B.G., 2008. Testate amoebae analysis in ecological and paleoecological studies of wetlands: past, present and future. *Biodiversity and Conservation* 17, 2115–2137.
- Montoya, E., Rull, V., van Geel, B., 2010. Non-pollen palynomorphs from surface sediments along an altitudinal transect of the Venezuelan Andes. *Palaeogeography Palaeoclimatology Palaeoecology* 297, 169–183.
- Munoz, P., Gorin, G., Parra, N., Velásquez, C., Lemus, D., Monsalve-M, C., Jojoa, M., 2017. Holocene climatic variations in the Western Cordillera of Colombia: a multiproxy high-resolution record unravels the dual influence of ENSO and ITCZ. *Quaternary Science Reviews* 155, 159–178.
- Myers, N., Mittermeier, R.A., Mittermeier, C.G., da Fonseca, G.A.B., Kent, J., 2000. Biodiversity hotspots for conservation priorities. *Nature* 403, 853–858.

- Ogden, C.G., 1979. Siliceous structures secreted by members of the subclass Lobosia (Rhizopodea: Protozoa). *Bulletin of the British Museum of Natural History (Zoology)* 36, 203–207.
- Ogden, C.G., 1980. Aspects of shell structure in genus *Diffflugia* (Rhizopodea). *Journal of Protozoology* 27, 57A–58A.
- Opravilova, V., Hajek, M., 2006. The variation of testacean assemblages (Rhizopoda) along the complete base-richness gradient in fens: a case study from the western Carpathians. *Acta Protozoologica* 45, 191–204.
- Payne, R., Blackford, J., 2008. Distal volcanic impacts on peatlands: palaeoecological evidence from Alaska. *Quaternary Science Reviews* 27, 21–22.
- Payne, R.J., Charman, D.J., Matthews, S., Eastwood, W.J., 2008. Testate amoebae as palaeohydrological proxies in sürmene ağaçbaşı yaylası peatland (northeast Turkey). *Wetlands* 28, 311–323.
- Payne, R.J., Telford, R.J., Blackford, J.J., Blundell, A., Booth, R.K., Charman, D.J., Lamentowicz, Ł., et al., 2012. Testing peatland testate amoeba transfer functions: appropriate methods for clustered training-sets. *The Holocene* 22, 819–825.
- Penard, E., 1902. *Les Rhizopodes du bassin du Léman* Kundig, Genève, Kundig ed., 714 p.
- Penard, E., 1910. Rhizopodes nouveaux. *Revue suisse de zoologie* 17, fasc. 4, pp. 929–940.
- Playfair, G.I., 1917. Rhizopods of Sydney and Lismore. *Proceedings of the Linnean Society of New South Wales* 42, 633–675.
- Poveda, G., Alvarez, D.M., Rueda, O.A., 2011. Hydro-climatic variability over the Andes of Colombia associated with ENSO: a review of climatic processes and their impact on one of the Earth's most important biodiversity hotspots. *Climate Dynamics* 36, 2233–2249.
- Poveda, G., Jaramillo, A., Gil, M., Quiceno, N., Mantilla, R., 2001. Seasonality in ENSO-related precipitation, river discharges, soil moisture, and vegetation index in Colombia. *Water Resources Research* 37, 2169–2178.
- Poveda, G., Mesa, O., 2004. On the existence of Lloro (the rainiest locality on Earth): enhanced ocean–atmosphere–land interaction by a low-level jet. *Geophysical Research Letters* 27, 1675–1678.
- Poveda, G., Vélez, J.I., Mesa, O., Hoyos, C.D., Mejía, J.F., Barco, O., Correa, P.L., 2002. Influencia de fenómenos macroclimáticos sobre el ciclo anual de la hidrología colombiana: cuantificación lineal, no lineal, y percentiles probabilísticos. *Meteorología Colombiana* 6, 121–130.
- Qin, Y., Mitchell, E.A.D., Lamentowicz, M., Payne, R.J., Lara, E., Gu, Y., Huang, X., Wang, H., 2013. Ecology of testate amoebae in peatlands of central China and development of a transfer function for paleohydrological reconstruction. *Journal of Paleolimnology* 50, 319–330.
- Schönborn, W., Peschke, T., 1990. Evolutionary studies on the *Assulina-Valkanovia* complex (Rhizopoda, Testaceafilosia) in *Sphagnum* and soil. *Biology and Fertility of Soils* 9, 95–100.
- Sklenář, P., Luteyn, J.L., Ulloa Ulloa, C., Jørgensen, P.M., Dillon, M.O., 2005. Flora generica de los paramos; Guía ilustrada de las plantas vasculares. *Memoirs of the New York Botanical Garden* 92, 499 p.
- Song, L., Li, H., Wang, K., Wu, D., Wu, H., 2014. Ecology of testate amoebae and their potential use as palaeohydrological indicators from peatland in Sanjiang Plain, Northeast China. *Frontiers of Earth Science* 8, 564–572.
- Spikings, R.A., Cochran, R., Villagomez, D., van der Lelij, R., Vallejo, C., Winker, W., Beate, B., 2015. The geological history of northwestern South America: from Pangaea to the early collision of the Caribbean Large Igneous Province (290–75 Ma). *Gondwana Research* 27, 96–139.
- Swindles, G.T., Amesbury, M.J., Turner, T.E., Carrivick, J.L., Woulds, C., Raby, C., Mullan, D., et al., 2015. Evaluating the use of testate amoebae for palaeohydrological reconstruction in permafrost peatlands. *Palaeogeography Palaeoclimatology Palaeoecology* 424: 111–122.
- Swindles, G.T., Charman, D.J., Roe, R.M., Sansum, P.A., 2009. Environmental controls on peatland testate amoebae (Protozoa: Rhizopoda) in the North of Ireland: implications for Holocene palaeoclimate studies. *Journal of Paleolimnology* 42, 123–140.
- Swindles, G.T., Lamentowicz, M., Reczuga, M., Galloway, J.M., 2016. Palaeoecology of testate amoebae in a tropical peatland. *European Journal of Protistology* 55, 181–189.
- Swindles, G.T., Kelly, T.J., Roucoux, K.H., Lawson, I.T., 2018a. Response of testate amoebae to a late Holocene ecosystem shift in an Amazonian peatland. *European Journal of Protistology* 64, 13–19.
- Swindles, G.T., Morris, P.J., Whitney, B., Galloway, J.M., Gatka, M., Gallego-Sala, A., Macumber, A.L., et al., 2018b. Ecosystem state shifts during long-term development of an Amazonian peatland. *Global Change Biology* 24, 738–757.
- Swindles, G.T., Reczuga, M., Lamentowicz, M., Raby, C.L., Turner, T.E., Charman, D.J., Gallego-Sala, A., et al., 2014. Ecology of testate amoebae in an Amazonian peatland and development of a transfer function for palaeohydrological reconstruction. *Microbial Ecology* 68, 284–298.
- Stuiver, M., Reimer, P.J., Reimer, R.W., 2018. CALIB 7.1 [www program] (accessed March 15, 2018). <http://calib.org>.
- Van Bellen, S., Mauquoy, D., Payne, R.J., Roland, T.P., Daley, T.J., Hughes, P.D.M., Loader, N.J., Street-Perrott, F.J., Rice, E.M., Pancotto, V.A., 2014. Testate amoebae as a proxy for reconstructing Holocene water table dynamics in southern Patagonian peat bogs. *Journal of Quaternary Science* 29, 463–474.
- Velásquez, C., Hooghiemstra, H., 2013. Pollen-based 17-kyr forest dynamics and climate change from the Western Cordillera of Colombia; no-analogue associations and temporarily lost biomes. *Review of Paleobotany and Palynology* 194, 38–49.
- Vuilleumier, F., Monasterio, M., 1986. *High Altitude Tropical Biogeography*. Oxford University Press, Oxford.
- Wanner, H., Beer, J., Bütikofer, J., Crowley, T.J., Cubasch, U., Flückiger, J., Goosse, H., et al., 2008. Mid- to Late Holocene climate change: an overview. *Quaternary Science Reviews* 27, 1791–1828.
- Wilmschurst, J.M., Wiser, S.K., Charman, D.J., 2003. Reconstructing Holocene water tables in New Zealand using testate amoebae: differential preservation of tests and implications for the use of transfer functions. *The Holocene* 13, 61–72.
- Woodland, W.A., Charman, D.J., Sims, P.C., 1998. Quantitative estimates of water tables and soil moisture in Holocene peatlands from testate amoebae. *The Holocene* 8, 261–27.