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ORIGINAL PAPER

Response of testate amoeba assemblages to peatland drain blocking

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Abstract Peatlands represent globally important habitats and carbon stores. However, human impacts and climate change leave peatlands with a substantial management challenge. Degradation of peatland habitats and their hydrological integrity is increasingly counteracted through the rehabilitation of peatlands including re-wetting and drain blocking. Research into how such management interventions affect peatland microbial assemblages is limited. Here, we investigate the response of testate amoebae (established unicellular amoeboid protist indicators of hydrological conditions in peatlands) to drain blocking on three small lowland raised bogs in Northern Ireland, UK. We sampled *Sphagnum* adjacent to areas of focused flow near sites of damming in addition to control sites away from dam blocking. These restoration measures show complex but meaningful results after restoration. We observe several key developments following dam blocking: (i) species diversity increases; (ii) unambiguous wet indicator taxa appear in increasing abundance at dammed sites; (iii) and transfer-function reconstructed water-table depths show wetter conditions in the dammed sites. These findings imply wetter conditions after restoration, where routine monitoring presented no clear trend in water-table depths. We found no statistically significant assemblage-level response to experimental or environmental variables, which may be related to antecedent conditions and significant periods of drought during the study period. Thus, caution is advised when utilising testate amoebae for bioindication until their assemblage-level response to restoration is better understood. Nevertheless, this study emphasises the potential of an indicator-taxa based approach to applying testate amoebae as contemporary bioindicators of peatland restoration—particularly on short-term timescales immediately following restoration.

Keywords Peatland · Restoration · Testate amoebae · *Sphagnum* · Biomonitoring

Introduction

Peatlands represent critically important habitats that anthropogenic pressures are putting at great risk (Auterives et al. 2011; Turner et al. 2014;

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Basińska et al. 2020). Predominantly these habitats are threatened by peat extraction, drainage, fire, afforestation, and land-use change (Schaible and Dickson 1990; Cooper et al. 2003; Tomlinson 2010; Douglas et al. 2015). In extreme circumstances, peatlands have been converted wholesale into profitable spaces (e.g., Oil Sands Mining—Rooney et al. 2012). Where peatlands are protected from direct exploitation, climate change and pollution can have major negative impacts on these habitats (e.g., atmospheric ammonia deposition on protected peatlands—Kelleghan et al. 2021). Peatlands provide several important ecosystem services. In the UK, peatlands provide flood relief (Edokpa et al. 2022), cultural heritage/public leisure spaces (Flint and Jennings 2022), and as sites for game bird management (Douglas et al. 2015). Internationally, peatlands prove to be unsustainable yet undeniably valuable agricultural areas (e.g., rice farming on peatland soils—Verhoeven and Setter 2010; Surahman et al. 2018). Most notably, peatlands have global carbon stores exceeding 600 GtC (Yu et al. 2010) and act as carbon sinks when healthy (Schwieger et al. 2021).

Threats from anthropogenic activity leave peatlands with a substantial management challenge. Direct exploitation needs to be opposed and legislated against, while omnipresent forms of large-scale damage such as agricultural pollution and climate change need to be reduced or counteracted. If this is not accomplished, the sizeable global store of carbon in peatlands could potentially be released to the atmosphere at an alarming rate (Ribeiro et al. 2021). Emissions from peatlands are around 23.1 Mt CO_2 equivalent in the UK annually (Parliamentary Office of Science and Technology 2022), as high as 1.91 Gt CO_2 equivalent per year globally (Leifeld and Menichetti 2018), and > 3.26 Mt CO_2 equivalent specifically for the island of Ireland (Wilson et al. 2013; Centre for Ecology & Hydrology 2017). The Intergovernmental Panel on Climate Change (IPCC 2018) reported that a decrease of only 25–30 Gt CO_2 equivalent per year of current global emissions could represent the difference between 1.5 and 2 °C warming by 2030. The effects of 2 °C warming could be extreme; for example, it could cause an increase in permafrost thaw by 2.5 million km^2 (IPCC 2018). International cooperation to see current emissions fall to acceptable levels is perhaps the largest challenge in generations. Adding 1.91 Gt CO_2 of avoidable carbon equivalents per year from peatlands would thwart these efforts.

Through pervasive anthropogenic pressures, it is thought that > 60% of UK and Irish peatlands are in a degraded state (Wilson et al. 2013; IUCN 2018; Tanneberger et al. 2021). A marked trend towards drying has been observed across European peatlands (Charman et al. 2006; Marcisz et al. 2015), which is linked to direct and indirect anthropogenic pressures on peatlands (Swindles et al. 2019). However, the last two decades have seen ambitious projects to re-wet peatlands across UK and Ireland (e.g., Parry et al. 2014; Holden et al. 2017). Re-wetting through drainage ditch damming can promote carbon sequestration (Beyer et al. 2021; Wilson et al. 2022). Re-wetting inhibits the decomposition of peat by reintroducing anoxic conditions by raising water-tables potentially causing peatlands that have shifted into becoming carbon sources to become carbon sinks again (Nugent et al. 2018). This process leads to peatlands becoming moderate sinks of CO_2 while generally emitting more CH_4, resulting in overall reductions in total greenhouse gas emissions (Nyberg et al. 2022; Wilson et al. 2022). Vegetation appears to recover in peatlands that undergo re-wetting restoration (Hancock et al. 2018), with important peatland species expanding, such as peat forming *Sphagnum* mosses (Malmer et al. 2003)—though this colonisation can take many years (Anderson and Peace 2017). *Sphagnum* colonisation following re-wetting can occur spontaneously (Graf et al. 2008; Mahmood and Strack 2011). However, practices are being developed to improve peatland re-vegetation, and to reduce greenhouse gas emissions, after re-wetting restoration (e.g., moss layer-transfer technique Lazcano et al. 2018; Purre et al. 2020). Evaluation of restoration success is a complex issue. The succession of vegetation, particularly *Sphagnum* in the case of peatlands, can be used to indicate restoration success (Soini et al. 2010). Different indicator species may have merit in predicting restoration outcomes in early stages post-restoration (González et al. 2013). Despite this, even over multi-year time series it can be difficult to determine the effect of restoration in the hydrology of a site (Green et al. 2017). Furthermore, shifts in greenhouse gas emissions following restoration can be multi-faceted, with reductions in carbon emissions but increases in methane emissions (Strack et al. 2014; Nugent et al. 2018; Nyberg et al.

2022; Wilson et al. 2022). Improved understanding of how to quantify the effectiveness of peatland restoration, which can be used to adapt and improve restoration methods, would be an invaluable tool in future peatland restoration projects.

Testate amoebae are a cosmopolitan group of protists that fulfil the role of dominant microbial consumers in peatlands (Mitchell et al. 2008; Jassey et al. 2013; Kuuri-Riutta et al. 2022). Testate amoebae can be found living on the stems of *Sphagnum* and in the still oxygenated layers of peat found close to the surface (Roe et al. 2017; Kuuri-Riutta et al. 2022). Testate amoebae have been shown to respond to a number of changing peatland conditions (Payne et al. 2012; Marcisz et al. 2020) and provide an established means of inferring past water levels from Holocene peat cores (Hendon and Charman 1997; Booth et al. 2010). The shell or 'test' (made from protective autogenous or xenogenous material) of these organisms generally resists decay, allowing them to preserve well in fossil peat (Charman et al. 2000). Assemblage response has been demonstrated to be quite rapid (Koenig et al. 2018a, b), allowing them to be used as contemporary indicators of peatland conditions. Testate amoeba taxa respond to varying peatland conditions differently (e.g., changes in moisture availability; nutrients; light; pH), meaning their response may be useful in inferring subtle changes in these habitats (Marcisz et al. 2020), where measured changes could take much longer to be fully understood. Testate amoebae could prove valuable for organisations trying to secure competitive funding for restoration work, where measured metrics are ambiguous or respond to management intervention too slowly.

Swindles et al. (2016) presented a contemporary time-series approach to testate amoebae assemblage response to changing peatland conditions before and after management inventions on a Welsh blanket bog—prior to this, similar research analysed testate amoebae assemblage changes in peat profiles after restoration (e.g., Davis and Wilkinson 2004; Valentine et al. 2013). Swindles et al. (2016) described a complex assemblage response of testate amoebae following restoration, where diversity increased, and the appearance of key unambiguous wet indicator taxa reflected the observed shift towards wetter peatland conditions. Additionally, Swindles et al. (2016) observed potential interactions between drainage ditches/treatment sites but could not easily discern this result due to the close proximity of control and treatment sites. For this study, control site locations were carefully considered to avoid obscuring potential interactions between drainage ditches or broader site-wide change. Creevy et al. (2018, 2023) demonstrated promising signs of peatland and testate amoebae assemblage recovery in a forest-to-bog restoration effort. Recent research highlights that further work is needed to provide a clear understanding of testate amoeba response to peatland restoration (Swindles et al. 2016; Creevy et al. 2018, 2023; Kuuri-Riutta et al. 2022). In this study, we investigate the response of testate amoebae to drainage ditch blocking from 2019 to 2021 on a lowland raised bog in Northern Ireland. A site from the Collaborative Action for the Natura Network (CANN) project was selected, which underwent drain blocking restoration in early 2020. This project provided regular water-table monitoring (Fig. 2) which continued for several years to align with restoration. This allowed us to collect *Sphagnum* samples months before the restoration began and have access to routine-monitoring data for the entire duration of the study.

Hypotheses

We tested the following hypotheses:

H1 Drain blocking leads to a change in testate amoebae assemblage dynamics.

H2 Unambiguous wet-indicator taxa abundance increases in response to restoration.

H3 Increased testate amoebae taxa diversity is observed following restoration.

Methods

Field site

This study was conducted at Cranny Bogs, a Special Area of Conservation (SAC) close to the town of Fintona in County Tyrone, Northern Ireland (54° 31′ 24.0″ N, 7° 20′ 37.0″ W). The site consists of three small, lowland raised bogs: Fallaghearn and

Killymoonan bogs to the east; and Cavan bog to the south-west (Fig. 1). The bogs are a combination of M18 *Erica tetralix-Sphagnum papillosum* raised and blanket bog, and M2 *Sphagnum cuspidatum/recurvum* bog pool communities under the UK National Vegetation Classification (NVC) (Rodwell 2006; DAERA 2015). The bog-myrtle *Myrica gale* can be found across all three bogs with its highest presence felt on Fallaghearn bog and least on Killymoonan bog. Cranny bogs have been predominantly damaged by peat cutting. Although areas of old hand cutting have since begun to regenerate, the historic activity left exposed cut faces as high as 2.5 m. A review of the site in 2015 (DAERA 2015) indicated that it is not known if an extant consent for peat cutting exists and that mechanised peat extraction has occurred in recent years. All drainage at the site is believed to be associated with this legacy of peat cutting. Sporadic burning is reported on all three bogs with the eastern bogs (Fallaghearn and Killymoonan bogs) having the largest extent of this past damage (DAERA 2015). The site also experiences nitrogen deposition (24.44 kg N/ha/year) above the calculated critical load for raised and blanket bogs (5–10 kg N/ha/year) (DAERA 2015; APIS 2020). The average annual rainfall at the site is between 1050 and 1650 mm year, with average temperatures of 4.2 °C in January and 15.4 °C in July (averages calculated from Edenfel Park and Castlederg MIDAS land surface weather stations for 2019, 2020, and 2021—Met Office 2022a, b) with an elevation of around 110–120 m.

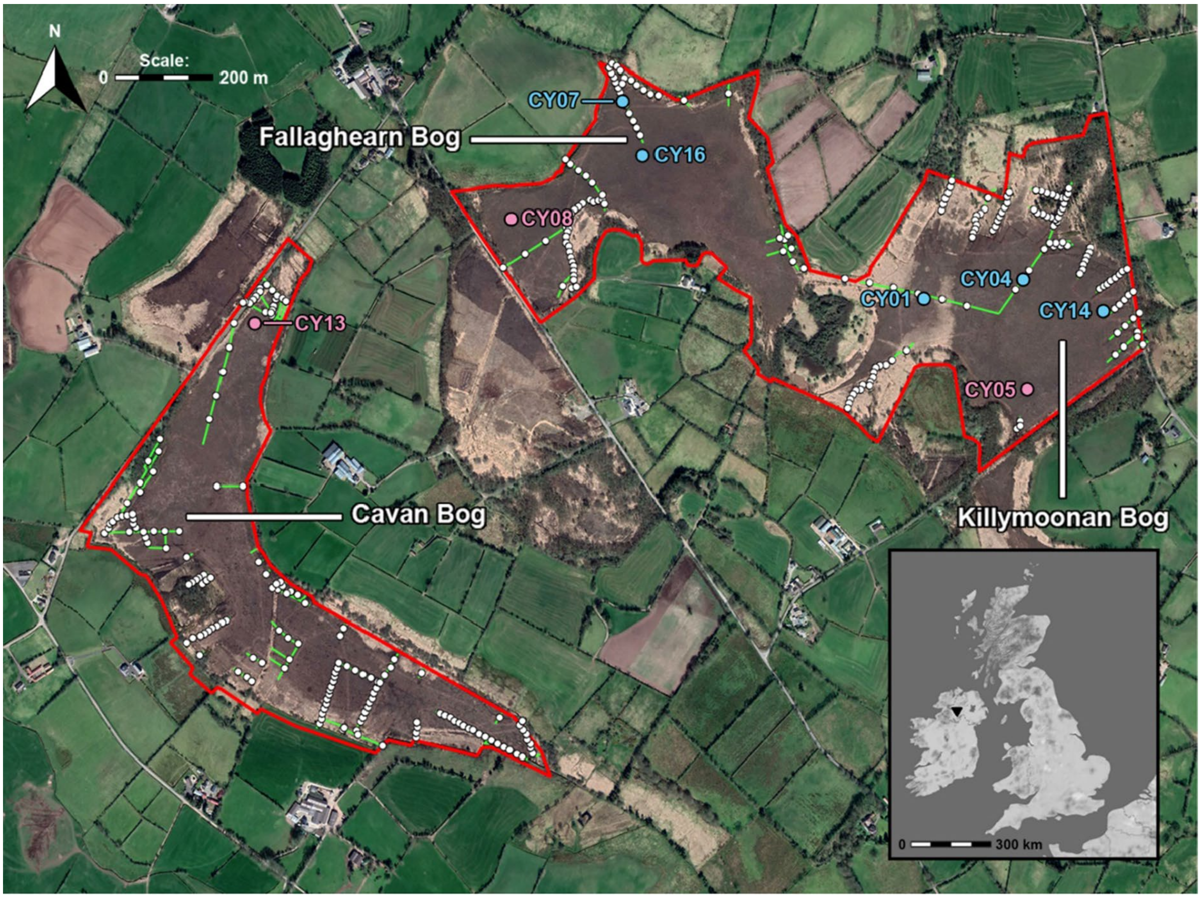

Fig. 1 Location map of the study site, Cranny Bogs (Cavan, Killymoonan, and Fallaghearn bogs) in County Tyrone, Northern Ireland. *Sphagnum* samples taken approximately 1 m from piezometer locations. Pink = control piezometer and sample (e.g., CY05); Blue = dammed piezometer and sample (e.g., CY01); White circles = dam locations; Green line = drainage ditches; Red line = SAC boundary (Sources: DAERA 2015; Google Earth 2022)

Restoration measures

Sphagnum samples were collected from eight locations across the three bogs and were selected based on their proximity to the drainage ditches present on the site. The replicates were classified into one of two restoration measures, either ‘dammed’ or ‘control’. The five ‘dammed’ replicates were located in areas of focused hydrological flow near drains whilst the three ‘control’ replicates were collected from areas away from drains and with no contributing hydrological catchment (Fig. 1). All five ‘dammed’ replicates were sampled from the two eastern bogs (Fallaghearn and Killymoonan bogs), whereas the ‘control’ replicates were located in each of the three bogs (Fallaghearn, Killymoonan, and Cavan bogs). The moss species chosen at each sample location were most commonly *Sphagnum capillifolium*. One dammed sample site (CY01) had a mix of *S. capillifolium* and *Sphagnum papillosum*, with a second dammed sample site (CY16) having a mix of *S. capillifolium* and *Sphagnum magellanicum*.

Routine monitoring

Baseline-monitoring began in the summer of 2018, for the CANN project. Regular water-table monitoring started on 8th July 2019, concluding several months after the final *Sphagnum* samples were collected for this study, on 25th October 2021. Damming of the drainage ditches on the site margins (Fig. 1) began on 30th January 2020 and was complete on all three bogs by 28th February 2020. Dams were constructed using peat, a common technique with smaller width drainage ditches (Armstrong et al. 2009; Parry et al. 2014).

Measurement of meteorological conditions

The Met Office Integrated Data Archive System (MIDAS) land surface station data was the source of meteorological data used in this study (Fig. 2) (Met Office 2022a, b), including air temperature (°C) and rainfall (mm) which the system reports on daily and hourly intervals. The closest station available for temperature data was situated approximately 8.8 km to the north of the study site in Edenfel Park (54° 35′ 38″ N, 7° 16′ 59″ W—Met Office 2022a), with the most suitable station for rainfall data being 24.8 km to the north-west of the study site in Castlederg (54° 42′ 25″ N, 7° 34′ 37″ W—Met Office 2022b). Other potential weather stations that were considered were further from the site or had incomplete data for the time period required.

Measurement of water-table depths

Sixteen piezometers were installed across Cranny Bogs in 2019 as part of the CANN project (The CANN Project 2022) to record water-table depth in areas of varying peatland condition (Fig. 2). Twelve piezometers were installed in areas of established raised bog. Of these, eight were located close to drains that were subsequently dammed, and three were placed far from the catchment areas and drains of the peatland. A single piezometer was positioned near catchment areas but away from proposed dams. All eight *Sphagnum* sampling sites (five ‘dammed’ and three ‘control’ restoration measures) were next to these piezometers on the raised bog. The remaining four piezometers were installed in areas of cutover bog are connected to the catchment areas of the peatland.

Sampling of testate amoebae

Live *Sphagnum* samples were collected 113 (07/11/2019—t_{-2}) and 81 (09/12/2019—t_{-1}) days before, and 3 (02/03/2020—t_0), 75 (13/05/2020—t_1), 175 (21/08/2020—t_2), 242 (26/10/2020—t_3), 293 (16/12/2020—t_4), 391 (24/03/2021—t_5), and 485 (28/06/2021—t_6) days after ditch dam construction was concluded (28/02/2020), corresponding to dates in which routine monitoring occurred at Cranny Bogs. The upper-most part of the *Sphagnum* moss (including a minimum of approximately 2 cm^3 of capitulum, stem, and branches) was selected in the field because the aerobic portions of *Sphagnum* stems have been shown to have the most live testate amoebae (Booth 2002). Each date corresponds with a date of routine water-table monitoring on Cranny Bogs with piezometers near all eight sample sites. *Sphagnum* samples measured greater than 2 cm^3 and were collected from a plot approximately 1 m from a piezometer and kept in individually labelled Ziplock bags. Samples were stored in a freezer until further preparation was carried out. *Sphagnum* samples were prepared using a mostly unmodified version of the

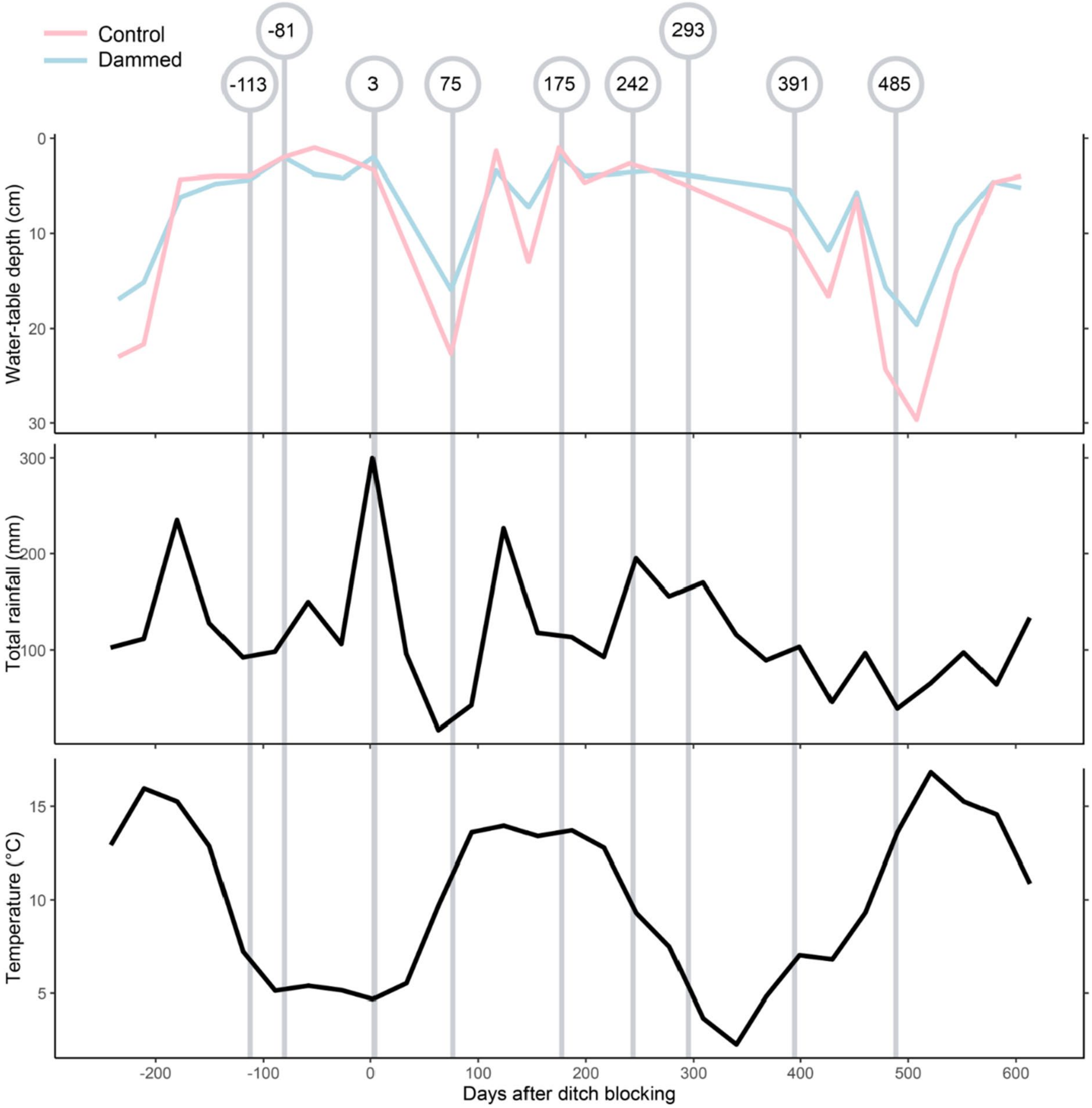

Fig. 2 Environmental variables over the course of the experiment. Water-table depths were routinely monitored on-site at dammed and control sample locations. Total monthly rainfall and mean monthly temperature data are from the Met Office Integrated Data Archive System weather stations (Met Office 2022a, b)

standard method (Booth et al. 2010) with the resulting testate amoebae samples being stored at 4 °C in the laboratory for analysis and identification over the following weeks. *Sphagnum* samples in boiling water were passed through a coarse-sieve of 300 µm, then back-sieved at 15 µm. Testate amoebae were then counted and noted as dead (only the test or partial test present) or 'live' (with a clearly visible cytoplasm) under transmitted light microscopy at × 200. Identification was performed at this time using illustrated taxonomic guides (Charman et al. 2000; Siemensma 2022). A minimum of 100 testate amoebae were identified and counted per sample (mean = 197). This minimum threshold is recommended by Payne and Mitchell (2009) for peatland water-table depth studies. Two *Lycopodium* spore tablets were added to samples and were later counted as recommended by the standard method (Booth et al. 2010), and test concentrations were calculated using an established formula (Stockmarr 1971).

Statistical analysis

Statistical analysis and data presentation was carried out using R version 4.2.1 (R Core Team 2022). The R package ‘vegan’ (version 2.6-2) was used for Non-metric Multidimensional Scaling (NMDS), permutational MANOVA (PERMANOVA), and Analysis of Similarity (ANOSIM), for use in investigating the response of testate amoebae assemblages to several experimental and environmental variables (time, restoration measure, rainfall, temperature, and measured and reconstructed water-table depth). Nonmetric multidimensional scaling (NMDS), PERMANOVA, and ANOSIM analysis were carried out using the Bray–Curtis dissimilarity index. The Shannon Diversity Index (Shannon and Weaver 1949) was calculated for each sample to explore species diversity. Water-table depth reconstructions were generated using the pan-European testate amoeba-based transfer function of Amesbury et al. (2016) using weighted average tolerance downweighing with inverse de-shrinking (WA-Tol). To achieve precision in the testate amoeba-based transfer function reconstructed water-table depths, a minimum count of 100 testate amoebae was strictly adhered to (Payne and Mitchell 2009).

Results

The most commonly occurring testate amoebae taxa identified at Cranny Bogs were *Nebela tincta*, *Assulina muscorum*, *Archerella flavum*, *Hyalosphenia elegans, Euglypha ciliata*, and *Heleopera sylvatica* (Fig. 6). The six most common taxa represented the vast majority (75%) of the 38 testate amoebae taxa identified. In total, 12,850 individuals were counted (mean 27.65% live testate amoebae—3463 individuals).

The Shannon Diversity Index (SDI) scores of the assemblages ranged from 0.35 to 2.46 and generally increased both in the control and dammed restoration measures after damming occurred (t_0 onwards) (Fig. 3). At the dammed sites the SDI scores/values were variable before management occurred, showing a high range and including the largest recorded value (CY07 t_{-2}—2.46) of the study prior to t_0 (Fig. 3). Additionally, the diversity in the control sites was substantially lower than the dammed sites prior to t_0.

Measured water-table depths at Cranny Bogs (Fig. 4a) appeared to be dominated by seasonality, with the data collected showing a trend towards deeper and drier water-tables in summer 2021 (t_6)—coinciding with lower observed rainfall during this period. Water-table depths were also reconstructed using a pan-European transfer function (Amesbury et al. 2016) (Fig. 4b) which showed substantial changes to the water-table depths of the dammed restoration measure sites. Reconstructed water-table depths were shallower and wetter in the dammed sites before intervention compared with the control sites.

A number of environmental variables were measured to test against this assemblage dataset. Observed and reconstructed water-table depths, air temperature, rainfall, time, and restoration measures could all be used in association with the testate amoebae assemblages to infer significant effects. Nonmetric multidimensional scaling with ANOSIM and PERMANOVA were used to evaluate the effect these environmental variables might have had on the assemblage composition of testate amoebae (NMDS – ‘stress’ ~ 0.25). All combinations of the dataset were considered, including how the results differed through time (before and after management). None of these variables were found to have a statistically significant effect on the testate amoebae assemblage composition (95% level).

The occurrence of non-ambiguous wet indicator taxa was notable in that prior to installation of drainage ditch dams (t_{-2} and t_{-1}) the only occurrence of these key taxa was a single occurrence (0.43% abundance, $n = 1$ specimen) of *Amphitrema wrightianum* in a control site (CY13, t_{-1}) and a peak of 24.89% abundance of *Archerella flavum* in a dammed site (CY14, t_{-1}) (Fig. 5). After management occurred (t_0 onwards) these key taxa did not appear in greater abundance in control sites (Fig. 5), where *A. wrightianum* was never observed again and *A. flavum* appeared in a single occurrence (CY13, t_1—0.45% abundance). *Amphitrema flavum* was observed in increasing abundance in the dammed sites, peaking at 66.19% relative abundance (CY14, t_6) (Fig. 5). Additionally, the key indicator taxa *A.*

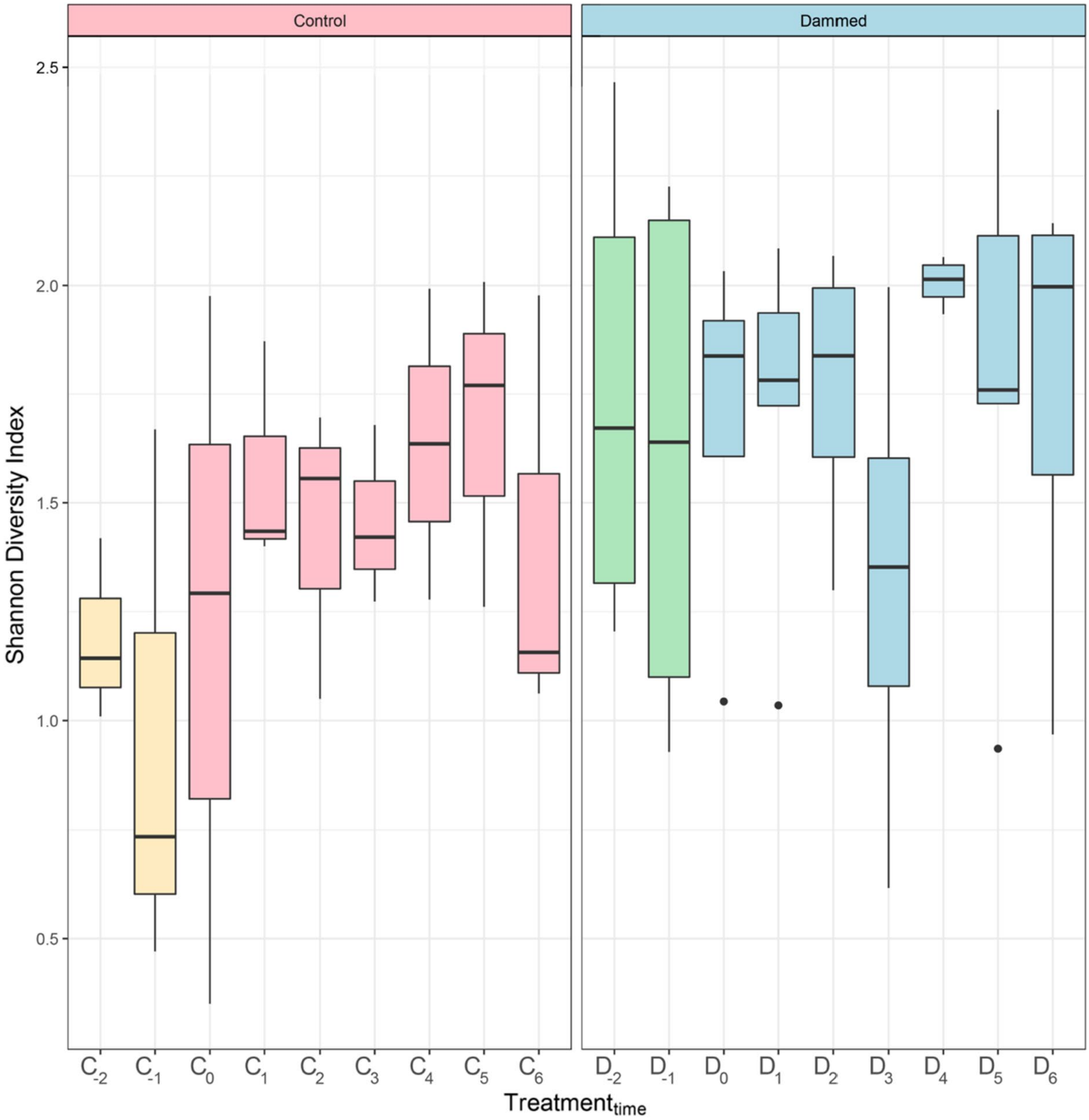

Fig. 3 Boxplot of testate amoebae assemblage Shannon Diversity Index from control and dammed sample sites. Results prior to restoration (t-2 and t-1) are highlighted

wrightianum, Amphitrema stenostoma, and *Centropyxis aculeata* type were recorded in a dammed site (CY07) after management occurred (Fig. 5). The single occurrences of *A. wrightianum* and *A. flavum* in the control sites is not likely to be through interaction between the five dammed sites of this study as these taxa occurred at CY13, the sole sampling location on the southernmost bog. Cavan Bog, where CY13 is located, is separated from the two northern bogs by a road and farmland (Fig. 1).

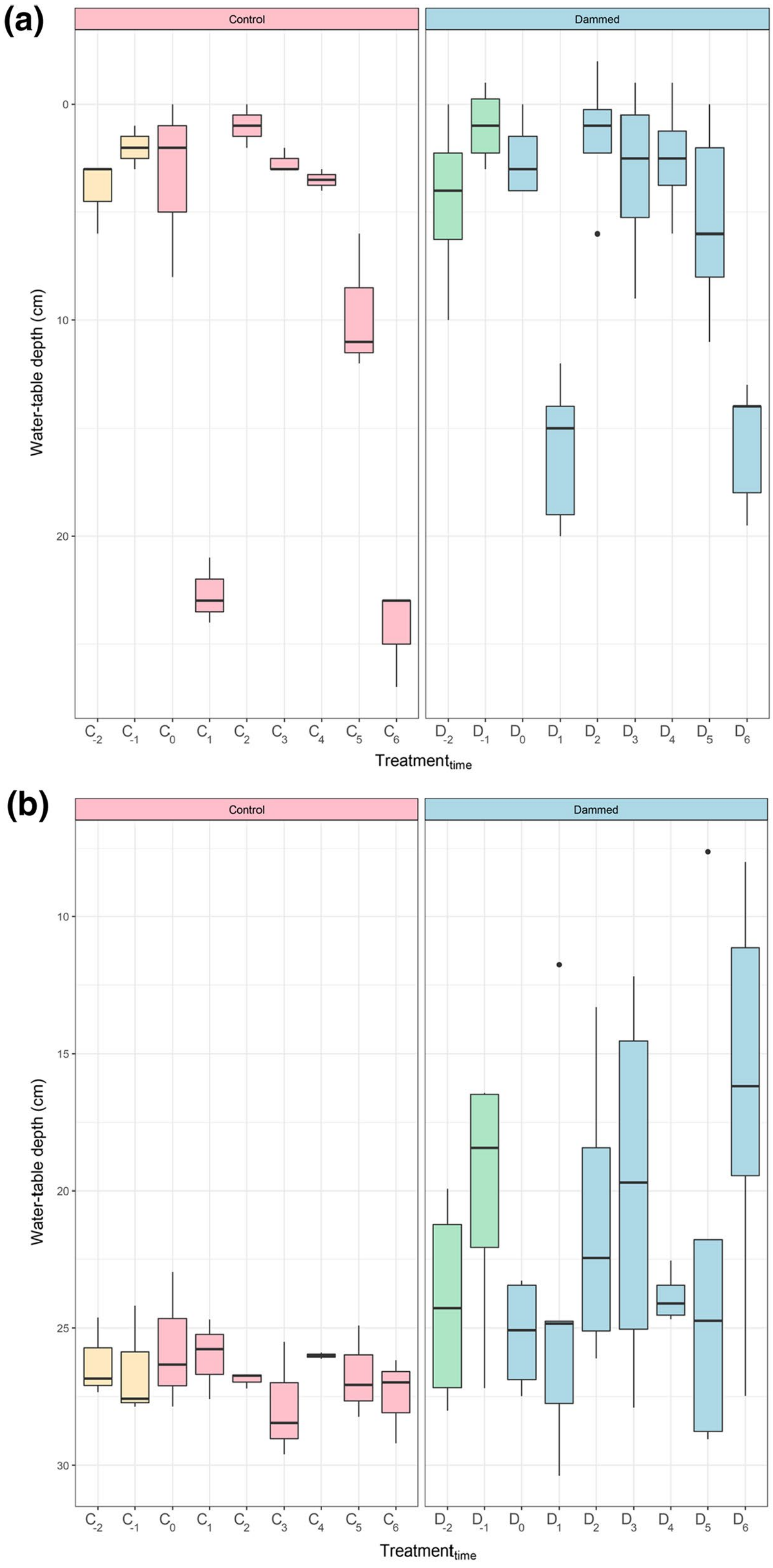

Fig. 4 **a** Boxplot of measured water-table depths. Results prior to restoration (t-2 and t-1) are highlighted. **b** Boxplot of testate-amoebae based transfer function reconstructed water-table depths. Results prior to restoration (t-2 and t-1) are highlighted

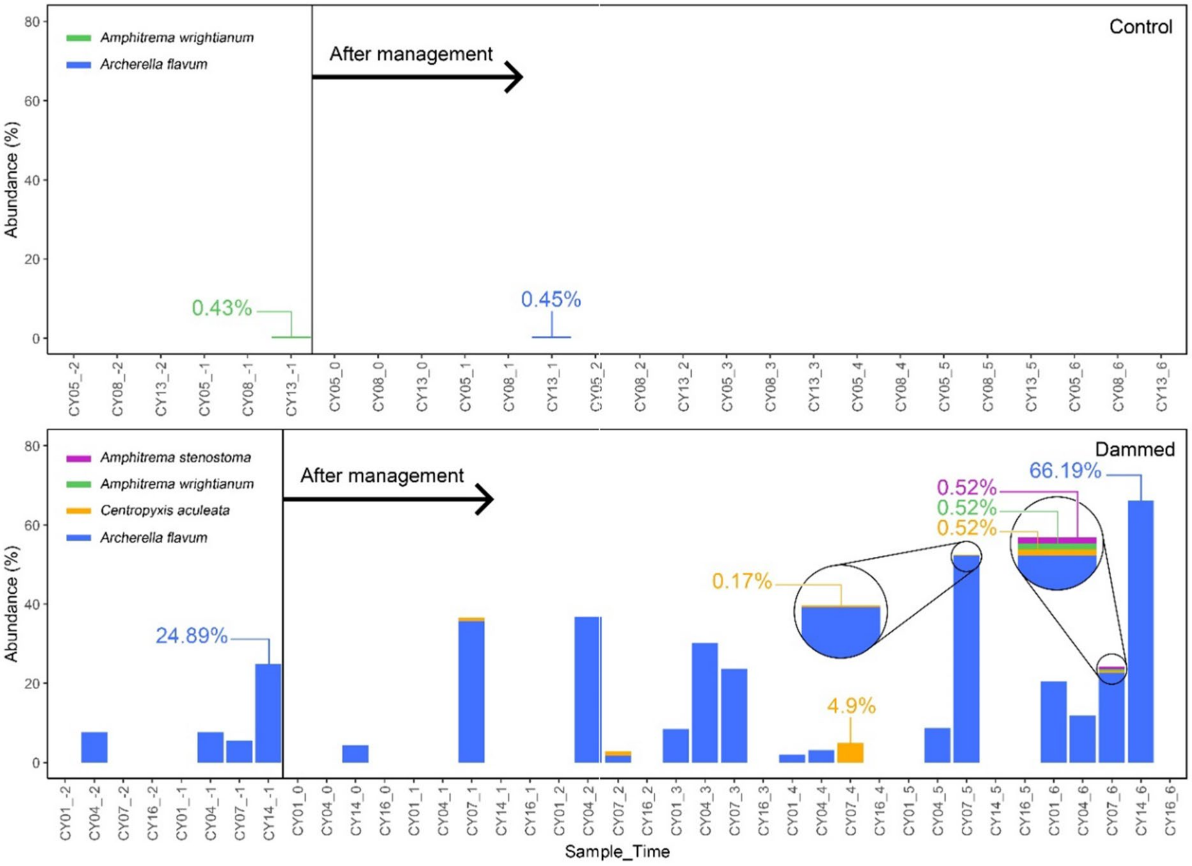

Fig. 5 Abundance (%) of unambiguous wet indicator taxa before and after restoration (t0 onwards). Sample site codes (e.g., CY16) are denoted with time of collection (e.g., − 2, − 1, 0 etc.) as seen in Fig. 1

Discussion

Peatland restoration at Cranny Bogs was characterised by a complex response of testate amoebae. Measured water-table depth indicated no clear response following management intervention (t_0 onwards) (Fig. 4a). Unseasonably dry conditions towards the end of the experiment ($t_4 - t_6$) appear to manifest as a slight trend towards deeper water-table depths (Figs. 2, 4a). Contrary to the measured results, testate-amoeba based transfer function reconstructed water-table depths show a notable trend toward shallower water-table depths in dammed restoration measure sites (Fig. 4). The appearance and proliferation of key unambiguous wet indicator taxa in the dammed sites (e.g., *A. flavum*) (Fig. 5) implies that restoration is having its intended effects. A general increase in SDI across the sites may indicate that management is having a wider ecohydrological impact across Cranny Bogs (Fig. 3). The large increase in wet indicator taxa specifically in the dammed sites (Fig. 5) suggests that this diversity increase is not caused by interactions between control and dammed sites as seen with similar studies (Swindles et al. 2016). Environmental variables and restoration measures had no statistically significant effect (95% level) on the testate amoebae assemblages. This lack of significance could be in part due to the antecedent conditions of the sites prior to the start of data collection. We believe *N. tincta* may have caused the lack of significance in our multivariate analysis. *Nebela tincta,* a ubiquitous taxon which is regarded as a poor hydrological indicator (Mitchell and Buttler 1999; Charman et al. 2000; Beaulne et al. 2018), often appears in very wet conditions (Warner 1987) but is commonly interpreted as a dry indicator (Booth 2008; Koenig et al. 2018a, b).

We observed very high abundances of *N. tincta* in our samples (Fig. 6), with this taxon even appearing in post-restoration samples at as much as 90% of the testate amoebae identified. The dominance of *N. tincta* in this site is problematic given the taxon's indifferent or ambiguous response to hydrological conditions. Nevertheless, the abundance of unambiguous wet indicator taxa increased substantially in the dammed sites (Fig. 5), despite decreased rainfall that could have led to drier conditions in 2021 (t_5 and t_6), suggesting improved hydrology at Cranny Bogs following management intervention.

Although the response of testate amoebae to peatland drain blocking in this study has been complex, we can accept two of our three proposed hypotheses. The appearance and proliferation of unambiguous wet-indicator taxa in dammed restoration measure sites and the general increase of SDI on Cranny Bogs allows us to accept our second and third hypotheses (H_2—Unambiguous wet-indicator taxa abundance increases in response to restoration; H_3—Increased testate amoebae taxa diversity is observed following restoration). Despite promising signs of testate amoebae assemblage change (e.g., increasingly wet reconstructed water-table depth), the remaining hypothesis (H_1—Drain blocking leads to a change in testate amoebae assemblage dynamics) must be rejected, as NMDS, PERMANOVA, and ANOSIM multivariate analysis illustrated a lack of statistically significant change at the testate amoebae assemblage-level.

Sample collection at Cranny Bogs began opportunistically, including antecedent conditions that may have caused a lack of significance in our multivariate analysis of testate amoebae assemblages. However, several previous experiments have demonstrated the significant influence environmental factors have at the testate amoebae assemblage-level. Variables associated with anthropogenic pressures have been shown to effect testate amoebae assemblages significantly. Daza Secco et al. (2018) reported that as much as 75% of the difference between testate amoeba assemblages in samples from three raised bogs in Finland were driven by land use. Further understanding of how these assemblages respond to environmental variables may be critical for restoration success. Creevy et al. (2018, 2023) examined forest-to-bog restoration sites where even after 17 years testate amoebae communities had not recovered due to limited expansion of *Sphagnum* after restoration. There is evidence to suggest that peatlands may be more resistant to hydrological change than previously thought (Lamentowicz et al. 2019), meaning a detailed paleo record at Cranny Bogs could aid in interpreting the sites' ability to recover from present and future disturbance. Further knowledge of the assemblage response of testate amoebae to restoration and critical environmental variables could inform effective restoration efforts. Our results show that these testate amoebae can respond rapidly to restoration, which could avoid decades of stagnant or ineffectual management practice.

Specific methods for testate amoebae sample preparation and analysis can be contentious (e.g., peat/*Sphagnum* sample boiling—Avel and Pensa 2013). Of note to this study, and our employed *Sphagnum* preparation methods (Booth et al. 2010), is the issue of micro-sieving (e.g., 15 µm back-sieve) which has been shown to exclude small testate amoebae taxa (Avel and Pensa 2013; McKeown et al. 2019). Many unambiguous wet indicator taxa, such as those found on Cranny Bogs (*A. flavum*, *C. aculeata* type, *A. stenostoma*, and *A. wrightianum*), are too large (> 45 µm) to be affected by these issues with micro-sieving. However, the multivariate analysis carried out for this study may have been affected by the omission of these small testate amoeba taxa. For instance, the common taxon *Cryptodifflugia oviformis*, was not found to be widespread on Cranny Bogs before or after restoration occurred. Other widespread but notably larger taxa (e.g., *Assulina muscorum*) were observed, which could highlight a loss of small taxa in this study.

At present the use of testate amoebae as bioindicators of peatland restoration success is better understood through an indicator-based approach, where known responses to changing environmental variables by specific taxa are used to infer the condition of the peatland. In this study we demonstrated that this method can be applied relatively simply and can be effective in scenarios where antecedent conditions and climatic events may cause issue with other forms of analysis. Here we used this approach to understand the hydrological response of a raised bog before and after re-wetting occurred. Using key unambiguous wet indicator taxa (*A. flavum*, *C. aculeata* type, *A. stenostoma*, and *A. wrightianum*) we have been able to infer wetter conditions over the course of nearly two years. In this same time frame, direct observation of water-table depths did not clearly indicate wetter

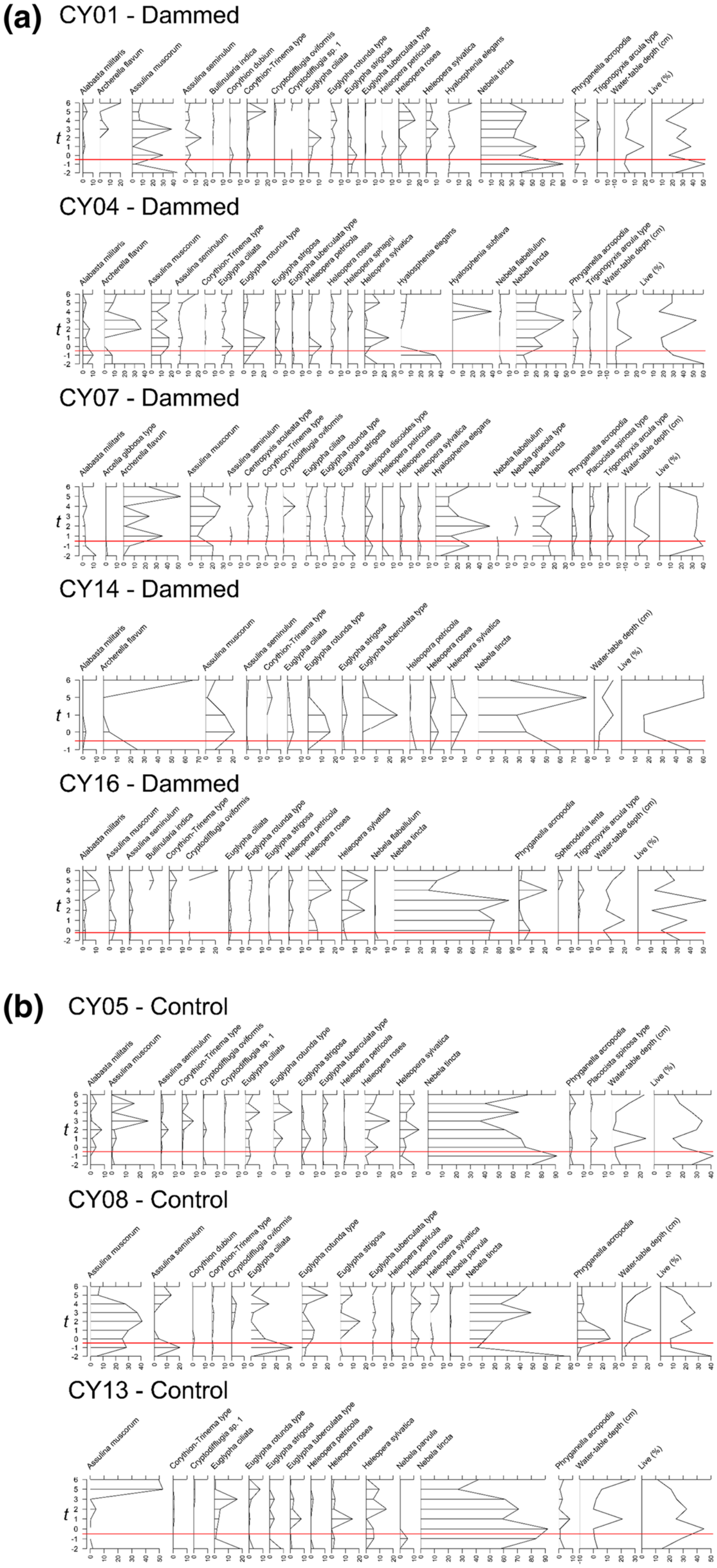

Fig. 6 **a** Percentage testate amoebae data for dammed sample sites (CY01, CY04, CY07, CY14, and CY16—Fig. 1). Measured water-table depth and the percentage of live testate amoebae are included. Red line denotes date of restoration. Samples were collected on the following dates: 07/11/2019 (− 2); 09/12/2019 (− 1); 02/03/2020 (0); 13/05/2020 (1); 21/08/2020 (2); 26/10/2020 (3); 16/12/2020 (4); 24/03/2021 (5); and 28/06/2021 (6). **b** Percentage testate amoebae data for control sample sites (CY05, CY08, and CY13—Fig. 1). Measured water-table depth and the percentage of live testate amoebae are included. Red line denotes date of restoration. Samples were collected on the following dates: 07/11/2019 (− 2); 09/12/2019 (− 1); 02/03/2020 (0); 13/05/2020 (1); 21/08/2020 (2); 26/10/2020 (3); 16/12/2020 (4); 24/03/2021 (5); and 28/06/2021 (6)

conditions due to periods of drought. Koenig et al. (2015) highlighted how straightforward this approach can be, by demonstrating that the use of as few as ten easily identifiable testate amoebae taxa, can be used to outperform bryophytes and vascular plants in 'bioindication value'. Furthermore, the use of testate amoeba indicator taxa may have applications in monitoring other forms of anthropogenic pressures on peatlands (e.g., ammonia pollution indicators—Payne et al. 2013).

If more is not done to understand the effectiveness of peatland restoration efforts, greenhouse gas emissions from degraded peatland sites could worsen. Emissions estimates from degraded peatlands are contentious (Leifeld and Menichetti 2018; Xu et al. 2018) with estimates of global total emissions and carbon stores of peatlands consistently uncertain (e.g., Congo Basin peatland extent; Dargie et al. 2017). Degraded UK peatland emissions were only formally included in the national greenhouse gas inventory as late as 2019 (Parliamentary Office of Science and Technology 2022). Official estimates for the Republic of Ireland have historically had considerable error ranges (Wilson et al. 2013; Donlan et al. 2016). Tropical peatlands account for the majority of peatland greenhouse gas emissions at this time (Leifeld and Menichetti 2018; Ribeiro et al. 2021), though notably they account for a major part of the global store of peatland carbon (Xu et al. 2018; Ribeiro et al. 2021). The response of tropical peatlands to restoration is poorly understood, particularly in terms of greenhouse gas emissions changes following management intervention (Kumar et al. 2020). Uncertain estimates for global peatland greenhouse gas emissions already suggest that as much as 1.91 Gt of CO_2 equivalents are released per year (Leifeld and Menichetti 2018). If global emissions are actually much higher, the case for understanding and implementing improved peatland restoration is urgent. Testate amoebae could be a vital part of these future restoration efforts, informing management practice and aiding in accurate and effective monitoring. However, the use of testate amebae in this manner is not well understood in tropical peatlands (Swindles et al. 2014; Liu et al. 2019; Krashevska et al. 2020) so we extend recommendations of further research on testate amoebae and restoration effectiveness in these regions, and investigations into the use of testate amoebae as bioindicators in agriculture and forestry on drained peatlands. In Germany greenhouse gas emissions from drained peatlands make up the largest part of emissions from agriculture (Tiemeyer et al. 2020), nevertheless a growing body of research suggests that agriculture and forestry on drained peatlands could potentially remain productive while greatly reducing emissions (Tanneberger et al. 2020; Evans et al. 2021). However, study of testate amoebae assemblage response to agricultural systems and forestry on drained peatlands is limited: Daza Secco et al. (2018) demonstrated the use of testate amoebae assemblages as bioindicators on drained peatlands for forestry; and Qin et al. (2020) presented their use on drained peatlands used for agricultural. In this study we have noted that restoration at Cranny Bogs has likely influenced the hydrology of the site—creating wetter conditions that favour these wet-indicator taxon (e.g., *Archerella flavum* and *Centropyxis aculeata* type). However, testate amoebae assemblages also respond to a number of ecological controls (Arrieira et al. 2015; Roe et al. 2017) such as: food availability; temperature; light; oxygen; and access to minerals for 'test' construction. The ways in which re-wetting influences these controls should be studied in greater detail.

Swindles et al. (2016) advised caution when using testate amoebae for biomonitoring of peatland restoration, especially when trying to analyse their assemblage-level response, due to this method needing further and more robust experimentation. Here we extend that recommendation of caution and highlight that more research needs to be targeted at understanding the assemblage-level response of testate amoebae following restoration. However, we note that the use of unambiguous wet indicator taxa has merit in rapid bioindication for early stages of peatland restoration.

Conclusions

This study investigated the use of testate amoebae as contemporary bioindicators of peatland hydrology following restoration on three small lowland raised bogs in Northern Ireland. Unambiguous wet indicator taxa (*A. flavum, C. aculeata* type*, A. stenostoma,* and *A. wrightianum*) were observed in increasing abundance on dammed restoration measure sites after management intervention (t_0 onward)—with some taxa appearing for the first time. These unambiguous wet indicator taxa were not observed in increasing abundance on any of our control sites. Diversity increased in both the control and dammed sites. This increase of unambiguous wet indicator taxa on dammed sites, lack of proliferation of these taxa in control sites (despite single occurrences of *A. flavum* and *A. wrightianum*), and a general site-wide diversity increase, suggest management has led to wetter conditions in and around former drainage ditches. Multivariate analysis was conducted to evaluate the assemblage-level response of testate amoebae in relation to experimental and environmental variables. Drought and the antecedent conditions of the study site appear to have affected the findings of this analysis, and as such we observed a lack of statistically significant assemblage-level change in response to these variables. Though complex, the findings of this investigation contribute to the growing body of research illustrating the value of testate amoebae as contemporary bioindicators of peatland restoration. An indicator-taxa based approach remains the clearest way of utilising testate amoebae for bioindication, with their assemblage-level response remaining complex and in need of further study at this time.

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Data availability The datasets generated during and/or analysed during the current study are available from the corresponding author on reasonable request.

Declarations

Competing interests The authors declare that they have no conflict of interest.

References

Amesbury MJ, Swindles GT, Bobrov A, Charman DJ, Holden J, Lamentowicz M, Mallon G, Mazei Y, Mitchell EAD, Payne RJ, Roland TP, Turner TE, Warner BG (2016) Development of a new pan-European testate amoeba transfer function for reconstructing peatland palaeohydrology. Quatern Sci Rev 152:132–151. https://doi.org/10.1016/j.quascirev.2016.09.024

Anderson R, Peace A (2017) Ten-year results of a comparison of methods for restoring afforested blanket bog. Mires Peat. https://doi.org/10.19189/MaP.2015.OMB.214

APIS (2020) UK air pollution information system. https://www.apis.ac.uk/. Accessed 3 April 2023

Armstrong A, Holden J, Kay P, Foulger M, Gledhill S, McDonald AT, Walker A (2009) Drain-blocking techniques on blanket peat: a framework for best practice. J Environ Manag 90:3512–3519. https://doi.org/10.1016/j.jenvman.2009.06.003

Arrieira RL, Schwind LTF, Bonecker CC, Lansac-Tôha FA (2015) Use of functional diversity to assess determinant assembly processes of testate amoebae community. Aquat Ecol 49:561–571. https://doi.org/10.1007/s10452-015-9546-z

Auterives C, Aquilina L, Bour O, Davranche M, Paquereau V (2011) Contribution of climatic and anthropogenic effects to the hydric deficit of peatlands. Hydrol Process 25:2890–2906. https://doi.org/10.1002/hyp.8052

Avel E, Pensa M (2013) Preparation of testate amoebae samples affects water table depth reconstructions in peatland palaeoecological studies. Est J Earth Sci 62:113. https://doi.org/10.3176/earth.2013.09

Basińska AM, Reczuga MK, Gąbka M, Stróżecki M, Łuców D, Samson M, Urbaniak M, Leśny J, Chojnicki BH, Gilbert D, Sobczyński T, Olejnik J, Silvennoinen H, Juszczak R, Lamentowicz M (2020) Experimental warming and precipitation reduction affect the biomass of microbial communities in a Sphagnum peatland. Ecol Indic 112:106059. https://doi.org/10.1016/j.ecolind.2019.106059

Beaulne J, Magnan G, Garneau M (2018) Evaluating the potential of testate amoebae as indicators of hydrological conditions in boreal forested peatlands. Ecol Ind 91:386–394. https://doi.org/10.1016/j.ecolind.2018.01.034

Beyer F, Jansen F, Jurasinski G, Koch M, Schröder B, Koebsch F (2021) Drought years in peatland rewetting: rapid vegetation succession can maintain the net CO_2 sink function. Biogeosciences 18:917–935. https://doi.org/10.5194/bg-18-917-2021

Booth RK (2002) Testate amoebae as paleoindicators of surface-moisture changes on Michigan peatlands: modern ecology and hydrological calibration. J Paleolimnol 28:329–348. https://doi.org/10.1023/A:1021675225099

Booth RK (2008) Testate amoebae as proxies for mean annual water-table depth in Sphagnum-dominated peatlands of North America. J Quat Sci 23:43–57. https://doi.org/10.1002/jqs.1114

Booth RK, Lamentowicz M, Charman DJ (2010) Preparation and analysis of testate amoebae in peatland palaeoenvironmental studies. Mires Peat 7:1–7

Centre for Ecology & Hydrology (2017) Implementation of an Emissions Inventory for UK Peatlands. https://uk-air.defra.gov.uk/assets/documents/reports/cat07/1904111135_UK_peatland_GHG_emissions.pdf. Accessed 6 Feb 2023

Charman DJ, Hendon D, Woodland WA (2000) The identification of testate amoebae (Protozoa: Rhizopoda) in peats. Quaternary Research Association, London

Charman DJ, Blundell A, Chiverrell RC, Hendon D, Langdon PG (2006) Compilation of non-annually resolved Holocene proxy climate records: stacked Holocene peatland palaeo-water table reconstructions from northern Britain. Quatern Sci Rev 25:336–350. https://doi.org/10.1016/j.quascirev.2005.05.005

Cooper A, McCann T, Meharg MJ (2003) Sampling Broad Habitat change to assess biodiversity conservation action in Northern Ireland. J Environ Manag 67:283–290. https://doi.org/10.1016/S0301-4797(02)00180-9

Creevy AL, Andersen R, Rowson JG, Payne RJ (2018) Testate amoebae as functionally significant bioindicators in forest-to-bog restoration. Ecol Ind 84:274–282. https://doi.org/10.1016/j.ecolind.2017.08.062

Creevy AL, Wilkinson DM, Andersen R, Rowson JG, Payne RJ (2023) Testate amoebae response and vegetation composition after plantationremoval on a former raised bog. Eur J Protistol 89:125977. https://doi.org/10.1016/j.ejop.2023.125977

Davis SR, Wilkinson DM (2004) The conservation management value of testate amoebae as 'restoration' indicators: speculations based on two damaged raised mires in northwest England. The Holocene 14:135–143. https://doi.org/10.1191/0959683604hl696rp

Daza Secco E, Haimi J, Högmander H, Taskinen S, Niku J, Meissner K (2018) Testate amoebae community analysis as a tool to assess biological impacts of peatland use. Wetl Ecol Manag 26:597–611. https://doi.org/10.1007/s11273-018-9594-y

Department of Agriculture, Environment and Rural Affairs (2015) Cranny Bogs SAC, UK0030321, CONSERVATION OBJECTIVES. https://www.daera-ni.gov.uk/sites/default/files/publications/doe/land-information-cladagh-cranny-bogs-conservation-objectives-2015.pdf. Accessed 10 Nov 2022

Donlan J, O'Dwyer J, Byrne KA (2016) Area estimations of cultivated organic soils in Ireland: reducing GHG reporting uncertainties. Mires Peat. https://doi.org/10.19189/MaP.2016.OMB.230

Douglas DJT, Buchanan GM, Thompson P, Amar A, Fielding DA, Redpath SM, Wilson JD (2015) Vegetation burning for game management in the UK uplands is increasing and overlaps spatially with soil carbon and protected areas. Biol Conserv 191:243–250. https://doi.org/10.1016/j.biocon.2015.06.014

Edokpa D, Milledge D, Allott T, Holden J, Shuttleworth E, Kay M, Johnston A, Millin-Chalabi G, Scott-Campbell M, Chandler D, Freestone J, Evans M (2022) Rainfall intensity and catchment size control storm runoff in a gullied blanket peatland. J Hydrol 609:127688. https://doi.org/10.1016/j.jhydrol.2022.127688

Evans CD, Peacock M, Baird AJ, Artz RRE, Burden A, Callaghan N, Chapman PJ, Cooper HM, Coyle M, Craig E, Cumming A, Dixon S, Gauci V, Grayson RP, Helfter C, Heppell CM, Holden J, Jones DL, Kaduk J, Levy P, Matthews R, McNamara NP, Misselbrook T, Oakley S, Page SE, Rayment M, Ridley LM, Stanley KM, Williamson JL, Worrall F, Morrison R (2021) Overriding water table control on managed peatland greenhouse gas emissions. Nature 593:548–552. https://doi.org/10.1038/s41586-021-03523-1

Flint A, Jennings B (2022) The role of cultural heritage in visitor narratives of peatlands: analysis of online user-generated reviews from three peatland sites in England. Int J Herit Stud 28:163–179. https://doi.org/10.1080/13527258.2021.1941198

González E, Rochefort L, Boudreau S, Hugron S, Poulin M (2013) Can indicator species predict restoration outcomes early in the monitoring process? A case study with peatlands. Ecol Ind 32:232–238. https://doi.org/10.1016/j.ecolind.2013.03.019

Google Earth (2022) County Tyrone, Northern Ireland. 54°31'33 " N, 7°21'13" W, Eye alt 7 km. Maxar Technologies 2023. http://www.earth.google.com. Accessed 27 Mar 2023

Graf MD, Rochefort L, Poulin M (2008) Spontaneous revegetation of cutwaway peatlands of North America. Wetlands 28:28–39. https://doi.org/10.1672/06-136.1

Green SM, Baird AJ, Holden J, Reed D, Birch K, Jones P (2017) An experimental study on the response of blanket bog vegetation and water tables to ditch blocking. Wetl Ecol Manag 25:703–716. https://doi.org/10.1007/s11273-017-9545-z

Hancock MH, Klein D, Andersen R, Cowie NR (2018) Vegetation response to restoration management of a blanket bog damaged by drainage and afforestation. Appl Veg Sci 21:167–178. https://doi.org/10.1111/avsc.12367

Hendon D, Charman DJ (1997) The preparation of testate amoebae (Protozoa: Rhizopoda) samples from peat. The Holocene 7:199–205. https://doi.org/10.1177/095968369700700207

Holden J, Green SM, Baird AJ, Grayson RP, Dooling GP, Chapman PJ, Evans CD, Peacock M, Swindles GT (2017) The impact of ditch blocking on the hydrological functioning of blanket peatlands: blanket peat ditch blocking impacts. Hydrol Process 31:525–539. https://doi.org/10.1002/hyp.11031

IPCC (2018) Global Warming of 1.5 °C. An IPCC special report on the impacts of global warming of 1.5 °C above pre-industrial levels and related global greenhouse gas emission pathways, in the context of strengthening the global response to the threat of climate change, sustainable development, and efforts to eradicate poverty. Cambridge University Press, Cambridge

IUCN (2018) UK Peatland Strategy. 2018–2040. https://www.iucn-uk-peatlandprogramme.org/sites/default/files/2022-04/UK%20Peatland%20Strategy%202018_2040.pdf. Accessed 10 Jan 2023

Jassey VEJ, Meyer C, Dupuy C, Bernard N, Mitchell EAD, Toussaint ML, Metian M, Chatelain AP, Gilbert D (2013) To what extent do food preferences explain the trophic position of heterotrophic and mixotrophic microbial consumers in a Sphagnum peatland? Microb Ecol 66:571–580. https://doi.org/10.1007/s00248-013-0262-8

Kelleghan DB, Hayes ET, Everard M, Keating P, Lesniak-Podsiadlo A, Curran TP (2021) Atmospheric ammonia and nitrogen deposition on Irish Natura 2000 sites: implications for Irish agriculture. Atmos Environ 261:118611. https://doi.org/10.1016/j.atmosenv.2021.118611

Koenig I, Feldmeyer-Christe E, Mitchell EAD (2015) Comparative ecology of vascular plant, bryophyte and testate amoeba communities in four Sphagnum peatlands along an altitudinal gradient in Switzerland. Ecol Ind 54:48–59. https://doi.org/10.1016/j.ecolind.2015.01.043

Koenig I, Christinat K, d'Inverno M, Mitchell EAD (2018a) Impact of two hot and dry summers on the community structure and functional diversity of testate amoebae in an artificial bog, illustrating their use as bioindicators of peatland health. Mires Peat 21:1–24. https://doi.org/10.19189/MaP.2018.OMB.327

Koenig I, Mulot M, Mitchell EAD (2018b) Taxonomic and functional traits responses of Sphagnum peatland testate amoebae to experimentally manipulated water table. Ecol Ind 85:342–351. https://doi.org/10.1016/j.ecolind.2017.10.017

Krashevska V, Tsyganov AN, Esaulov AS, Mazei YA, Hapsari KA, Saad A, Sabiham S, Behling H, Biagioni S (2020) Testate amoeba species- and trait-based transfer functions for reconstruction of hydrological regime in tropical peatland of central Sumatra, Indonesia. Front Ecol Evol 8:225. https://doi.org/10.3389/fevo.2020.00225

Kumar P, Adelodun AA, Khan MF, Krisnawati H, Garcia-Menendez F (2020) Towards an improved understanding of greenhouse gas emissions and fluxes in tropical peatlands of Southeast Asia. Sustain Cities Soc 53:101881. https://doi.org/10.1016/j.scs.2019.101881

Kuuri-Riutta O, Väliranta M, Tuittila E-S (2022) Literature review on testate amoebae as environmental indicators and as a functional part of the microbial community in northern peatlands. Mires Peat 28:1–16. https://doi.org/10.19189/MaP.2022.OMB.StA.2412

Lamentowicz M, Gałka M, Marcisz K, Słowiński M, Kajukało-Drygalska K, Dayras MD, Jassey VEJ (2019) Unveiling tipping points in long-term ecological records from Sphagnum-dominated peatlands. Biol Lett 15:20190043. https://doi.org/10.1098/rsbl.2019.0043

Lazcano C, Robinson C, Hassanpour G, Strack M (2018) Short-term effects of fen peatland restoration through the moss layer transfer technique on the soil CO_2 and CH_4 efflux. Ecol Eng 125:149–158. https://doi.org/10.1016/j.ecoleng.2018.10.018

Leifeld J, Menichetti L (2018) The underappreciated potential of peatlands in global climate change mitigation strategies. Nat Commun 9:1071. https://doi.org/10.1038/s41467-018-03406-6

Liu B, Booth RK, Escobar J, Wei Z, Bird BW, Pardo A, Curtis JH, Ouyang J (2019) Ecology and paleoenvironmental application of testate amoebae in peatlands of the high-elevation Colombian páramo. Quat Res 92:14–32. https://doi.org/10.1017/qua.2018.143

Mahmood MdS, Strack M (2011) Methane dynamics of recolonized cutover minerotrophic peatland: Implications for restoration. Ecol Eng 37:1859–1868. https://doi.org/10.1016/j.ecoleng.2011.06.007

Malmer N, Albinsson C, Svensson BM, Wallén B (2003) Interferences between Sphagnum and vascular plants: effects on plant community structure and peat formation. Oikos 100:469–482. https://doi.org/10.1034/j.1600-0706.2003.12170.x

Marcisz K, Tinner W, Colombaroli D, Kołaczek P, Słowiński M, Fiałkiewicz-Kozieł B, Łokas E, Lamentowicz M (2015) Long-term hydrological dynamics and fire history over the last 2000 years in CE Europe reconstructed from a high-resolution peat archive. Quatern Sci Rev 112:138–152. https://doi.org/10.1016/j.quascirev.2015.01.019

Marcisz K, Jassey VEJ, Kosakyan A, Krashevska V, Lahr DJG, Lara E, Lamentowicz Ł, Lamentowicz M, Macumber A, Mazei Y, Mitchell EAD, Nasser NA, Patterson RT, Roe HM, Singer D, Tsyganov AN, Fournier B (2020) Testate amoeba functional traits and their use in paleoecology. Front Ecol Evol 8:575966. https://doi.org/10.3389/fevo.2020.575966

Met Office (2022a) MIDAS Open: UK daily temperature data, v202207. NERC EDS Centre for Environmental Data Analysis, 22 September 2022. https://doi.org/10.5285/8bcf6925cddc4681b96f94d424537b9e. Accessed 14 Dec 2022

Met Office (2022b) MIDAS Open: UK hourly rainfall data, v2022b07. NERC EDS Centre for Environmental Data Analysis, 22 September 2022b. https://doi.org/10.5285/64f5d7be890a4ac08cb2b4e78eb5fcc1. Accessed 14 Dec 2022

McKeown MM, Wilmshurst JM, Duckert C, Wood JR, Mitchell EAD (2019) Assessing the ecological value of small testate amoebae (<45 µm) in New Zealand peatlands. Eur J Protistol 68:1–16. https://doi.org/10.1016/j.ejop.2018.12.002

Mitchell EAD, Buttler AJ, Warner BG, Gobat J-M (1999) Ecology of testate amoebae (Protozoa: Rhizopoda) in

Sphagnum peatlands in the Jura mountains, Switzerland and France. Écoscience 6:565–576. https://doi.org/10.1080/11956860.1999.11682555

Mitchell EAD, Charman DJ, Warner BG (2008) Testate amoebae analysis in ecological and paleoecological studies of wetlands: past, present and future. Biodivers Conserv 17:2115–2137. https://doi.org/10.1007/s10531-007-9221-3

Nugent KA, Strachan IB, Strack M, Roulet NT, Rochefort L (2018) Multi-year net ecosystem carbon balance of a restored peatland reveals a return to carbon sink. Glob Change Biol 24:5751–5768. https://doi.org/10.1111/gcb.14449

Nyberg M, Black TA, Ketler R, Lee SC, Johnson M, Merkens M, Nugent KA, Knox SH (2022) Impacts of active versus passive re-wetting on the carbon balance of a previously drained bog. J Geophys Res 127:e2022006881. https://doi.org/10.1029/2022JG006881

Parliamentary Office of Science and Technology (2022) Reducing peatland emissions. https://post.parliament.uk/research-briefings/post-pn-0668/. Accessed 9 Jan 2023

Parry LE, Holden J, Chapman PJ (2014) Restoration of blanket peatlands. J Environ Manag 133:193–205. https://doi.org/10.1016/j.jenvman.2013.11.033

Payne RJ, Mitchell EAD (2009) How many is enough? Determining optimal count totals for ecological and palaeoecological studies of testate amoebae. J Paleolimnol 42:483–495. https://doi.org/10.1007/s10933-008-9299-y

Payne RJ, Lamentowicz M, van der Knaap WO, van Leeuwen JFN, Mitchell EAD, Mazei Y (2012) Testate amoebae in pollen slides. Rev Palaeobot Palynol 173:68–79. https://doi.org/10.1016/j.revpalbo.2011.09.006

Payne RJ, Jassey VEJ, Leith ID, Sheppard LJ, Dise NB, Gilbert D (2013) Ammonia exposure promotes algal biomass in an ombrotrophic peatland. Soil Biol Biochem 57:936–938. https://doi.org/10.1016/j.soilbio.2012.09.012

Purre A, Ilomets M, Truus L, Pajula R, Sepp K (2020) The effect of different treatments of moss layer transfer technique on plant functional types' biomass in revegetated milled peatlands. Restor Ecol 28:1584–1595. https://doi.org/10.1111/rec.13246

Qin Y, Puppe D, Payne R, Li L, Li J, Zhang Z, Xie S (2020) Land-use change effects on protozoic silicon pools in the Dajiuhu National Wetland Park. China. Geoderma 368:114305. https://doi.org/10.1016/j.geoderma.2020.114305

R Core Team (2022) R: a language and environment for statistical computing, Vienna, Austria. https://www.R-project.org/. Accessed 27 Mar 2023

Ribeiro K, Pacheco FS, Ferreira JW, Sousa-Neto ER, Hastie A, Krieger Filho GC, Alvalá PC, Forti MC, Ometto JP (2021) Tropical peatlands and their contribution to the global carbon cycle and climate change. Glob Change Biol 27:489–505. https://doi.org/10.1111/gcb.15408

Rodwell JS (2006) National vegetation classification: user's handbook. JNCC, Peterborough

Roe HM, Elliott SM, Patterson RT (2017) Re-assessing the vertical distribution of testate amoeba communities in surface peats: Implications for palaeohydrological studies. Eur J Protistol 60:13–27. https://doi.org/10.1016/j.ejop.2017.03.006

Rooney RC, Bayley SE, Schindler DW (2012) Oil sands mining and reclamation cause massive loss of peatland and stored carbon. Proc Natl Acad Sci USA 109:4933–4937. https://doi.org/10.1073/pnas.1117693108

Schaible R, Dickson DA (1990) Effects of drainage intensity and planting position on the growth and nutrition of second rotation Sitka spruce on shallow peat. Forestry 47:19–27

Schwieger S, Kreyling J, Couwenberg J, Smiljanić M, Weigel R, Wilmking M, Blume-Werry G (2021) Wetter is better: rewetting of minerotrophic peatlands increases plant production and moves them towards carbon sinks in a dry year. Ecosystems 24:1093–1109. https://doi.org/10.1007/s10021-020-00570-z

Shannon CE, Weaver W (1949) The mathematical theory of communication. University of Illinois Press, Illinois, Champaign

Siemensma FJ (2022) Microworld, world of amoeboid organisms. https://arcella.nl/. Accessed 24 Oct 2022

Soini P, Riutta T, Yli-Petäys M, Vasander H (2010) Comparison of vegetation and CO_2 dynamics between a restored cut-away peatland and a pristine fen: evaluation of the restoration success. Restor Ecol 18:894–903. https://doi.org/10.1111/j.1526-100X.2009.00520.x

Stockmarr J (1971) Tablets with spores used in absolute pollen analysis. Pollen Spores 13:615–621

Strack M, Keith AM, Xu B (2014) Growing season carbon dioxide and methane exchange at a restored peatland on the Western Boreal Plain. Ecol Eng 64:231–239. https://doi.org/10.1016/j.ecoleng.2013.12.013

Surahman A, Soni P, Shivakoti GP (2018) Are peatland farming systems sustainable? Case study on assessing existing farming systems in the peatland of Central Kalimantan, Indonesia. J Integr Environ Sci 15:1–19. https://doi.org/10.1080/1943815X.2017.1412326

Swindles GT, Reczuga M, Lamentowicz M, Raby CL, Turner TE, Charman DJ, Gallego-Sala A, Valderrama E, Williams C, Draper F, Honorio Coronado EN, Roucoux KH, Baker T, Mullan DJ (2014) Ecology of testate amoebae in an Amazonian peatland and development of a transfer function for Palaeohydrological reconstruction. Microb Ecol 68:284–298. https://doi.org/10.1007/s00248-014-0378-5

Swindles GT, Green SM, Brown L, Holden J, Raby CL, Turner TE, Smart R, Peacock M, Baird AJ (2016) Evaluating the use of dominant microbial consumers (testate amoebae) as indicators of blanket peatland restoration. Ecol Ind 69:318–330. https://doi.org/10.1016/j.ecolind.2016.04.038

Swindles GT, Morris PJ, Mullan DJ, Payne RJ, Roland TP, Amesbury MJ, Lamentowicz M, Turner TE, Gallego-Sala A, Sim T, Barr ID, Blaauw M, Blundell A, Chambers FM, Charman DJ, Feurdean A, Galloway JM, Gałka M, Green SM, Kajukało K, Karofeld E, Korhola A, Lamentowicz Ł, Langdon P, Marcisz K, Mauquoy D, Mazei YA, McKeown MM, Mitchell EAD, Novenko E, Plunkett G, Roe HM, Schoning K, Sillasoo Ü, Tsyganov AN, van der Linden M, Väliranta M, Warner B (2019) Widespread drying of European peatlands in recent centuries. Nat Geosci 12:922–928. https://doi.org/10.1038/s41561-019-0462-z

Tanneberger F, Schröder C, Hohlbein M, Lenschow U, Permien T, Wichmann S, Wichtmann W (2020) Climate change mitigation through land use on rewetted peatlands—cross-sectoral spatial planning for Paludiculture in Northeast Germany. Wetlands 40:2309–2320. https://doi.org/10.1007/s13157-020-01310-8

Tanneberger F, Moen A, Barthelmes A, Lewis E, Miles L, Sirin A, Tegetmeyer C, Joosten H (2021) Mires in Europe—regional diversity, condition and protection. Diversity 13:381. https://doi.org/10.3390/d13080381

The CANN Project (2022) Collaborative action for the natura network. https://thecannproject.org/. Accessed 2 Nov 2022

Tiemeyer B, Freibauer A, Borraz EA, Augustin J, Bechtold M, Beetz S, Beyer C, Ebli M, Eickenscheidt T, Fiedlere S, Förster C, Gensior A, Giebels M, Glatzel S, Heinichen J, Hoffmann M, Höper H, Jurasinski G, Laggner A, Leiber-Sauheitl K, Peichl-Brakg M, Drösler M (2020) A new methodology for organic soils in national greenhouse gas inventories: Data synthesis, derivation and application. Ecol Indic 109:105838. https://doi.org/10.1016/j.ecolind.2019.105838

Tomlinson RW (2010) Changes in the extent of peat extraction in Northern Ireland 1990–2008 and associated changes in carbon loss. Appl Geogr 30:294–301. https://doi.org/10.1016/j.apgeog.2009.08.004

Turner TE, Swindles GT, Roucoux KH (2014) Late Holocene ecohydrological and carbon dynamics of a UK raised bog: impact of human activity and climate change. Quatern Sci Rev 84:65–85. https://doi.org/10.1016/j.quascirev.2013.10.030

Valentine J, Davis SR, Kirby JR, Wilkinson DM (2013) The use of testate amoebae in monitoring peatland restoration management: case studies from North West England and Ireland. Acta Protozoologica 52:129–145. https://doi.org/10.4467/16890027AP.13.0013.1110

Verhoeven JTA, Setter TL (2010) Agricultural use of wetlands: opportunities and limitations. Ann Bot 105:155–163. https://doi.org/10.1093/aob/mcp172

Warner BG (1987) Abundance and diversity of testate amoebae (Rhizopoda, Testacea) in Sphagnum peatlands in southwestern Ontario, Canada. Arch Protistenk 133:173–189. https://doi.org/10.1016/S0003-9365(87)80051-9

Wilson D, Müller C, Renou-Wilson F (2013) Carbon emissions and removals from Irish peatlands: present trends and future mitigation measures. Ir Geogr 46:1–23. https://doi.org/10.1080/00750778.2013.848542

Wilson D, Mackin F, Tuovinen J, Moser G, Farrell C, Renou-Wilson F (2022) Carbon and climate implications of rewetting a raised bog in Ireland. Glob Change Biol 28:6349–6365. https://doi.org/10.1111/gcb.16359

Xu J, Morris PJ, Liu J, Holden J (2018) PEATMAP: refining estimates of global peatland distribution based on a meta-analysis. CATENA 160:134–140. https://doi.org/10.1016/j.catena.2017.09.010

Yu Z, Loisel J, Brosseau DP, Beilman DW, Hunt SJ (2010) Global peatland dynamics since the Last Glacial Maximum. Geophys Res Lett 37:L13402. https://doi.org/10.1029/2010GL043584