Annals of Environmental Science and Ecology

Research Article

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Reconstructing the Evolutionary History of an Ephemeral Groundwater-Fed Turlough Wetland Using *n*-Alkane Geochemistry

Vane CH[1*], Farr G[2], Bevan J[3], Moss-Hayes V[1], Bosanquet S[4], Jones PS[5], Rutt G[3] and Naughton O[6]

[1]British Geological Survey, Keyworth, Nottingham, UK

[2]Mining and Remediation Authority, Mansfield, Nottinghamshire, UK

[3]Natural Resources Wales, Maes Newydd, Llandarcy, Neath Port Talbot

[4]Natural Resources Wales, Hadnock Road, Monmouthshire

[5]Natural Resources Wales, Maes y Ffynnon, Penrhosgarnedd

[6]Trinity College Dublin, College Green, Ireland

Summary

This study provides the first description of the evolution of an ephemeral groundwater fed wetland (turlough) during the last 12K years BP. The study site; 'Pant-y-llyn' is the only recorded turlough in mainland Britain. Associated with karstic Carboniferous Limestone terrain, turloughs form in depressions and their ephemeral hydrology is controlled by fluctuations in the local groundwater regime. Turloughs are common in the Republic of Ireland however similar ephemeral water bodies occur in many karstic areas around the world. Coring identified 5.5 m of basin infill comprising 3.5m of peat underlain by 2m of basal silt-clay deposits. Geochemical *n*-alkane and initial macrofossil analysis suggests the site has evolved from a fully aquatic water body infilling with fine sediments, towards a more emergent system. ^{14}C radiocarbon dating suggests progression from a dominantly aquatic to more emergent conditions and the onset of peat formation between 12589–12105 cal BP and 8932-8545 cal BP. This study illustrates the use of *n*-alkane analysis to reconstruct the evolutionary history of ephemeral wetlands.

Keywords: *n*-alkane; Geochemistry groundwater; Turlough; Karst; limestone.

INTRODUCTION

A turlough is defined as *'a topographic depression in karst which is intermittently inundated on an annual basis, mainly from groundwater, and which has a substrate and/or ecological communities characteristic of wetlands'* [1]. Annual changes in water levels are the most striking hydrological feature of turloughs, and they are primarily considered to be an Irish habitat, with over 300 sites documented in the Republic of Ireland [2,3], three sites ; in Northern Ireland [4], and one example, Panty-llyn in south Wales [5]. Similar ephemeral water bodies are found in Spain, Slovenia and Canada [6,7]. Owing to their rarity and vulnerability, turloughs have been classified as an Annex I priority habitat under the EU Habitats Directive 92/43/EC [8].

Pant-y-llyn (Figure 1) (Easting 260620 Northing 216650) is a small turlough measuring just 160 m in length and 60 m in width. Situated in an area of karstic limestone at an intermediate altitude of ~150 m above sea level, it floods to depths of 3 m (Farr et al. 2012) in response to changes in the elevation of the local groundwater table. Pant-y-llyn is designated as a Special Area of Conservation (SAC) and Site of Special Scientific Interest (SSSI) for both its ecological and hydrological features. Previous studies at Pant-y-llyn describe both ecological and hydrological aspects including its classification as a turlough [5], hydrological functioning and groundwater catchment delineation using dye tracing [10], hydrological modelling and water chemistry [9] and identification of ancient volcanic tephra associated with ash clouds [12]. Ecological research includes characterization and monitoring of the vegetation zonation [13,14] which is currently dominated by amphibious bryophytes and *Callitriche* in the lowest-lying zone and by *Carex* sedges in the upper zones, with no *Sphagnum* at all. The invertebrate fauna has been characterized [15,13, 16] and compared to other temporary water bodies [17]. The dry phase of turloughs deters fish and other predators, making them ideal breading sites for amphibians such as the common toad *Bufo bufo* [18].

The purpose of this study was to use *n*-alkane chemistry in combination with macrofossil analysis to better understand the evolutionary history of this rare form of wetland..

Submitted: 19 Sept 2026 | **Accepted:** 12 Nov 2026 | **Published:** 13 Nov 2026

***Corresponding author:** Christopher H Vane, chv@bgs.ac.uk, British Geological Survey, Keyworth, Nottingham, UK

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Citation: Vane CH, Farr G, Bevan J, Moss-Hayes V, Bosanquet S, et al. (2026) Reconstructing the Evolutionary History of an Ephemeral Ground-water-Fed Turlough Wetland Using *n*-Alkane Geochemistry. Ann Environ Sci Ecol 6(1): 1024.

METHODS

Core retrieval

An initial survey of the turlough basin was undertaken

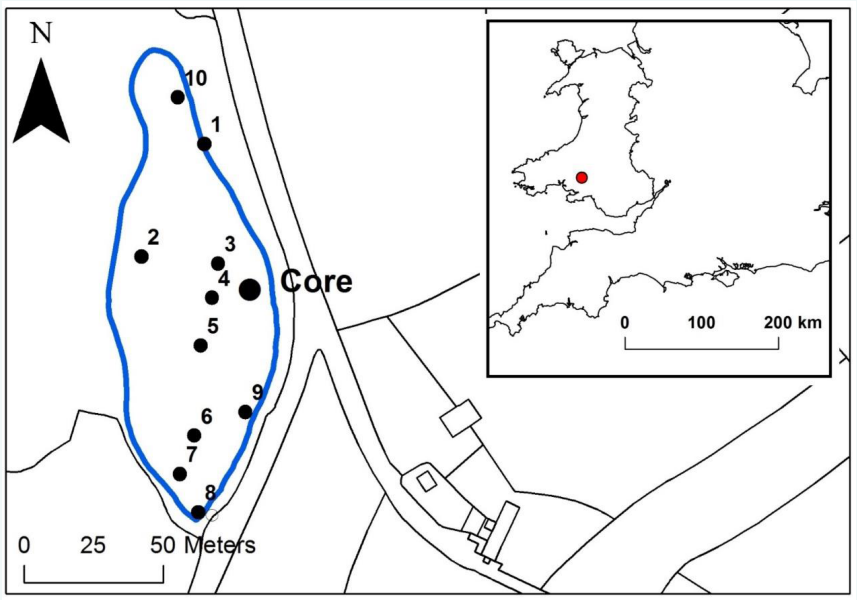


Figure 1: Location of Pant-y-llyn with location of trial holes (1-10) and core location (Core). Contains Ordnance Survey Master Map data © Crown Copyright and database rights 2019.

using a peat probe and hand auger to investigate the depth of the unconsolidated fill in the basin. In total ten locations were investigated within the turlough basin. Following this initial survey the part of the basin with the deepest unconsolidated infill was selected as a location from which to retrieve a core (Easting 260634 Northing 216648). A single 5.5 m deep core was obtained on the 28th August 2013 using a peat or 'Russian' corer with a 0.5 L sample volume, stored in PVC casing and wrapped in cling film before being stored in a fridge at BGS, Keyworth (British Geological Survey borehole reference SN61NW12). Sub-samples for organic geochemistry were freeze dried and ground to powder using a ball mill (Retsch PM100) to < 250 µm.

This same core was also used in a simultaneous project to identify tephra – a volcanic glass associated with ash clouds [12].

Marcofossils

Macrofossil were sampled and identified to species level where possible from horizons where there were visual changes in the lithology: at -423 cm, -417 cm.-270 cm,-245 cm. at -100 cm.

Gas chromatography of saturated hydrocarbons

Geochemical analysis of the 5.5 m core was undertaken in order to construct an *n*-alkane profile. Normal alkanes (*n*-alkanes) are a sub-class of wax lipid widely used to provenance organic matter in modern and palaeo lakes sediments [19-21]. Twenty-one dried sediments (0.5 g) were each spiked with tetracosane-D50, squalane, hexatriacontane-D74 at 5µg/g and allowed to equilibrate for 12 hours. Sediments were mixed with clean sand and copper powder and extracted with dichloromethane/methanol (9:1 v/v) at 75°C and 750 psi using ASE (accelerated solvent extraction). Extracts were reduced to dryness using a TurboVap and reconstituted in a minimum volume of dichloromethane/propan-2-ol (2:1 v/v) for column chromatography. Silica gel (Si60 200-500µm, 5% deactivated) was used in a 1.5 × 20 cm column and eluted with three column volumes of *n*-hexane to isolate the saturate fraction. The effluent was reduced in volume (1mL) and reconstituted in an appropriate volume of *n*-hexane for geochemical analysis. The saturates were analysed on a Hewlett Packard 6890 series GC-FID system fitted with an Agilent DB-1ms UI column (60 m length, 0.25 mm id., 0.1 µ film thickness). Oven temperature was: 60°C (isothermal for 1 minute) to 320°C (isothermal for 15 minutes) at 10°C / minute.

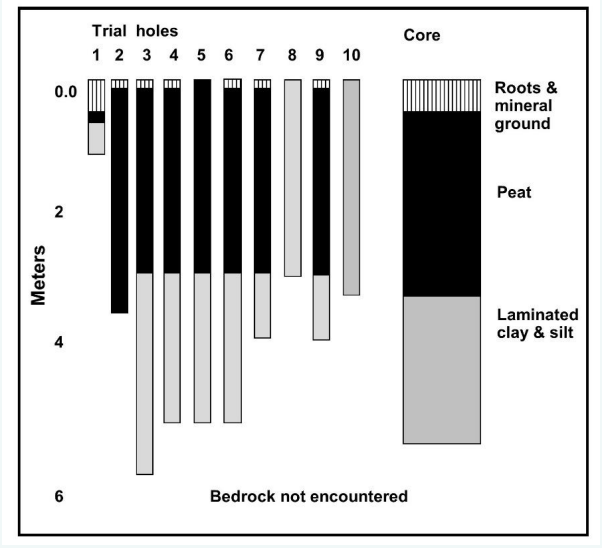


Figure 2: Generalised summary of the stratigraphy of trial holes (1-10) and main core

1µL was injected at 280°C in splitless mode for 0.7 min, split 1:10 thereafter and the Helium carrier gas was 1mL/min. Peak areas were integrated using Clarity software.

Radiocarbon dating

Four bulk 100 g samples were removed from the 5.5 m core and stored in glass vials. The samples were taken from 200, 245, 395 and 510 cm depths, corresponding with key *n*-alkane sample horizons. ^{14}C AMS (Accelerator Mass Spectrometry) radiocarbon dating of the four samples was undertaken at the ^{14}CHRONO Centre, Queens University, Belfast. The samples were measured on an AMS for fraction correction for the radiocarbon age. Calibration was undertaken using the Intca113 and Maringe13 radiocarbon age calibration curves [22], ages are reported as ranges cal BP. The same radiocarbon dates are also used in a simultaneous project undertaken to identify tephra – a volcanic glass associated with ash clouds [12].

RESULTS

The following results are presented and interpreted with evidence for aquatic or terrestrial conditions and are used to create an initial evolutionary history of Pant-y-llyn during the last 13,000 years (CalBP).

Core

The investigation demonstrated a basin depth of 5.5 m of unconsolidated material (Figure 2). Core stratigraphy can be summarised as: mineral ground (0 to -22 cm), peat (-22 to -364 cm), laminated organic-rich silt-clay material (-364 to -500 cm), underlain by unlaminated clay-rich sediment (-500 to -550 cm); the auger hole collapsed after -550 cm with no bedrock encountered and thus the true depth of the basin is not known. The depth of infill material was unexpected as the core was obtained fairly close to the eastern margin of the site where it was assumed bedrock or glacial till would be in close proximity to the surface. There is no evidence within the core of significant erosional periods and thus it is possible that the material deposited within this basin represents a continual depositional record throughout the last ~ 13,000 years (CalBP).

Marcofossils

Macrofossil analysis from key horizons provides a tantalizing glimpse into the evolutionary history of Pant-y-llyn. Deep within the sequence at -423 cm, seeds of *Potamogeton crispus* and *P. natans* suggest periods of permanent flooding, with the shallow water alga *Gleotrichia* noted at -417 cm. The more permanently flooded pond then moves into a swamp phase evidenced by the presence of *Equisetum* fragments and *Sparganium* or *Typha* at -270 cm, followed by the development of a *Sphagnum*-rich fen with abundant *Sphagnum palustre* remains at -245 cm. *Equisetum* and wood (*Salix cinerea*) fragments also occur later in the sequence at -100 cm. A more detailed study of macrofossils would provide a higher resolution history of habitat evolution at Pant-y-llyn.

Gas Chromatography of saturated hydrocarbons

Normal alkanes (*n*-alkanes) are a sub-class of wax lipid used to provenance organic matter in river-estuaries [23], tropical and riparian wetlands [24,25], as well as modern and palaeo lake sediments [19-21]. Aquatic algae and plankton synthesise mainly odd numbered short-chain homologues (C_{13}-C_{25}) maximising at about C_{15} or C_{17}; in contrast to vascular plants and particularly the waxy protective coatings of leaf cuticles which biosynthesise mainly odd numbered long-chain homologues (C_{23}-C_{35}) maximising at C_{29}, C_{31} or C_{33} [26,27]. Similarly, certain *Sphagnum* species show either C_{31} dominated distribution or, in the case of *S.*

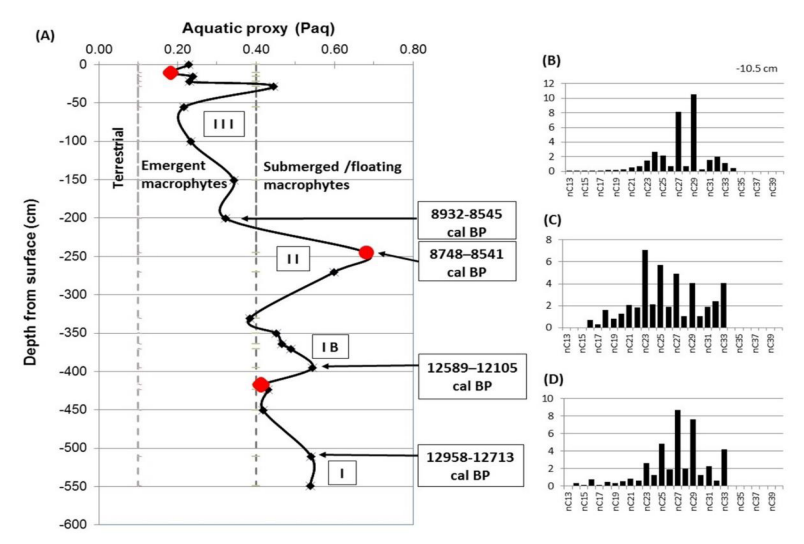


Figure 3: A) Down core aquatic proxy (Paq) for submerged/floating (0.4-1), emergent (0.1-0.4) and terrestrial (<0.1) macrophytes, The up-profile trend suggests increasing terrestrialisation, radiocarbon dates for key horizons are also shown; B,C,D) Contrasting exemplar distributions of *n*-alkanes interpreted to suggest submerged/floating (-423 cm), *Sphagnum palustre* (-245 cm), emergent terrestrial (-10.5 cm).

palustre and *S. papillosum*, a bimodal distribution with maxima at C_{23} and C_{31} [28]. Although identification of a specific floral source may be confounded by the ubiquity of C_{29}, C_{31} and C_{33}, in terrestrial systems, Ficken et al. [29], showed that submerged and floating leaved aquatic plants produce elevated abundances of C_{23} and C_{25} as compared to C_{29}, C_{31} and C_{33} homologues. These compositional relationships were summarised by the aquatic proxy (P*aq* = $(C_{23}+C_{25})/(C_{23}+C_{25}+C_{29}+C_{31})$ to distinguish *n*-alkanes from plants that grow in fresh water/ submerged (0.40-1.00) from those that are emergent (0.1-0.40) and those growing in a fully terrestrial environment (≤ 0.1) [29].

At Pant-y-llyn (Figure 3A) sediments from the base of the core (-550 cm to -330 cm) give Paq values that ranged between 0.4 and 0.54 corresponding to *n*-alkane input from submerged/floating macrophytes, which in turn suggests that initially Pant-y-llyn was a fully aquatic environment (Zone I) that gradually received an increased amount of alkanes sourced from emergent flora, such as, but not limited to *Typha* and / or *Phragmites*. This interpretation is entirely consistent with the overall distribution of the *n*-alkanes and is also supported by the presence of *Potamogeton crispus* and *P. natans* seeds at -423 cm and the shallow water alga Gleotrichia at -417 cm. The subsequent increase to Paq values of 0.54 (-395 cm) and then a gradual decrease to 0.40 (-330 cm) suggests a second phase of deeper aquatic conditions (Zone IB) followed by a return to shallower conditions with corresponding greater input from emergent flora.

Inspection of the *n*-alkane profiles between -270.5 and -245.5 cm suggest a significant change in organic matter source (Zone II). Although the P*aq* proxy indicates *n*-alkanes sourced from submerged floating macrophytes, both distribution profiles mitigate against this interpretation. The sample at -245.5 cm centred at C_{23} with diminishing amounts of C_{25} and C_{27} (Figure 3 C) is similar to that previously reported for specific *Sphagnum* moss species, namely, *S. palustre* which is most commonly found at the margins of bogs [28] and as a more general component of poor-fens. This interpretation was confirmed by the presence of abundant *S. palustre* remains at this horizon in the core. On balance the organic geochemical data in Zone II suggests a transitional phase to generally less aquatic conditions. The *n*-alkane profiles from sediments between -200 cm and surface (Figure 3 C-D) are all unimodal and mainly dominated by C_{27} and C_{29} with much lower amounts of other homologues, a distribution characteristic of grass and tree leaf waxes. This, combined with the general up-profile decrease in P*aq* from 0.32 at -200 cm to 0.23 at surface suggests a systematic decrease in inputs from fully submerged/ floating flora and a concomitant increase in *n*-alkanes from emergent and terrestrial (tree leaves) which probably indicates a shift to shallower water and/or only episodic aquatic conditions.

Radiocarbon dating

^{14}C radiocarbon dating of the four bulk samples from the 5.5 m core (Table 1) has dated the basal (-510 cm) silt-clay sample to 12958-12713 cal BP indicating a considerable age to the deeper sediments. The silt-clay sample at -395 cm depth was dated at 12589–12105 cal BP. These samples correspond to Zone I and Zone 1B in the gas chromatography analysis, representing a fully aquatic environment. The start of peat formation, at approximately -364 cm in the core, would have commenced sometime between 12589–12105 cal BP (-395 cm sample) and 8748–8541 cal BP (-245 cm peat sample). The -200 cm peat sample, corresponding to Zone III in the gas chromatography analysis, characterized by less submerged conditions, was dated at 8932-8545 cal BP.

Evolution of Pant-y-llyn over the last 13 Ka (BPCal)

A series of generic graphical illustrations, Figure 4, propose an initial time line for the evolution of Pant-y-llyn over the last 13 Ka (BPCal). Following the melting of the ice sheets and erosion of the landscape, the first known hydrological setting is that of open water body. During the open water body period clays and silts are deposited at the lake bottom. Following this there is a transitional phase with succession to swamp and a terrestrial *Sphagnum*-rich fen. Following this the *n*-alkane evidence and marco fossil evidence suggests that Pant-y-llyn moves towards a more emergent system from about 8932-8545 cal BP. It generally remains in this terrestrial setting until the present day however it is not possible to define the onset of fluctuating turlough conditions, as a higher resolution analysis would be required of the upper part of the core.

DISCUSSION

The epehrmal nautre of Pant-y-llyn is dependant entierly upon fluctuations in the local groundwater regime. It is unkown for how long exactly it has been operating as an ephermal wetland, with annual empty and fill cycles. With a maximum flood depth of just 3 m this poses an interesting question as to how Pant-y-

Table 1: Peat core radiocarbon dates (modified from Jones et al.2017)

Depth	14C age years BP	Calibrated age range (CalBP) (95.4%)	Description of sample
200 cm	7857±41 ^{14}C	8932-8545 cal BP	Zone III: start of emerging macrophytes
245 cm	7833±37 ^{14}C	8748–8541 cal BP	Zone II: transitional phase
395 cm	10479±65 ^{14}C	12589–12105 cal BP	Zone IB: second phase of submergence
510 cm	10953±47 ^{14}C	12958-12713 cal BP	Zone I: fully aquatic and start of peat formation

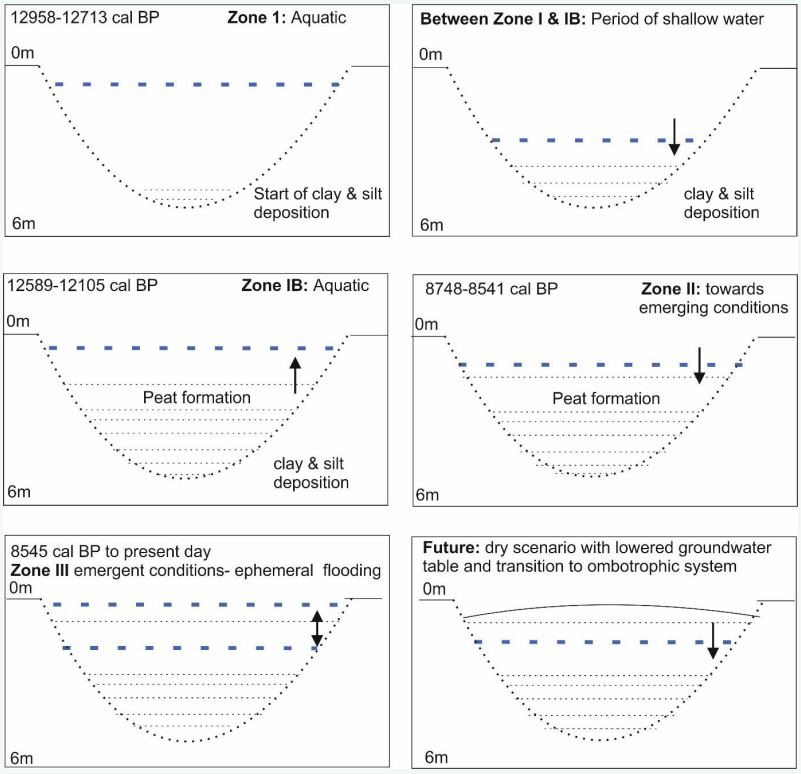


Figure 4: Schematic conceptual models to represent key phases in Pant-y-llyn since ~13,000 years cal BP and possible future scenario in drier climate. *Diagram not to scale; depth to rock head and geomorphology of basin not known; water levels are estimated.*

llyn may respond to a changing climate. Changing weather and climate patters could manifest as higher or lower groundwater levels, thus there is every possiliblity that even with a small overall change in the annual mean groundwater regime Pant-y-llyn could become either permiantly flooded or permemantly terrestrial, thus loosing its epehermal nature. Wetter climatic conditions could result in permanent flooding with Pant-y-llyn returning to an open water body. Conversly a drier climate could result in lower groundwater levels, thus driving Pant-y-llyn towards a wholly emergant system, even with the possbility of transittion towards a dominatly ombrogenous system. To better understand how long Pant-y-llyn has been ephermal a much higher resolution *n*-alkane, macrofossil and ^{14}C analysis of the core, would be required. This information would be useful when reviewing site management targets, many of which are 'set in stone' and may not take into account the potential dynamic nature of the site.

CONCLUSIONS

Analysis of the core using *n*-alkanes, ^{14}C and macrofossil analysis has given an initial insight into the evolution of Pant-y-llyn turlough.. Analysis of *n*-alkanes combined with C14 analysis and macrofossil identification suggests that Pant-y-llyn was initially a fully aquatic water body (129580 -12105 cal BP). This early aquatic stage was followed by succession to swamp vegetation and then development of a terrestrial *Sphagnum*-rich fen. ^{14}C dating suggests the transition from dominantly aquatic to a more emergent system and the start of peat formation occurred sometime between 12589–12105 cal BP and 8932-8545 cal BP. The presence of abundant *Sphagnum palustre* remains, dated to 8748–8541 cal BP, suggesting a fully terrestrial wetland, before subsequent development at a currently unknown time, to the present day fluctuating turlough conditions. The method of investigation could be applied to other ephemeral wetlands where retrieval of continually deposited infill can be obtained.

ACKNOWLEDGMENTS

The authors would like to thank Tristan Hatton Ellis, Tracey Lovering, Rhys Heath, Mike Jenkins, Simon Neale and David A Jones from Natural Resources Wales. Christopher H Vane and Vicky Moss-Hayes publish with the permission of the Executive Director of the British Geological Survey (UKRI).

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