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## Assessing reference conditions and ecological status for lakes using subfossil diatoms

Science Report: SC030103/SR3

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This report is the result of research commissioned and funded by the Environment Agency's Science Programme.

**Published by:**

Environment Agency, Rio House, Waterside Drive, Aztec West,  
Almondsbury, Bristol, BS32 4UD  
Tel: 01454 624400 Fax: 01454 624409  
[www.environment-agency.gov.uk](http://www.environment-agency.gov.uk)

ISBN: 978-1-84432-902-1

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February 2009

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**Dissemination Status:**

Released to all regions  
Publicly available

**Keywords:**

diatoms, lakes, palaeolimnology, Water Framework Directive, ecological status, reference conditions

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**Science Project Number:**

SC030103

**Product Code:**

SCH00508BOBP-E-P

# Science at the Environment Agency

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Steve Killeen

**Head of Science**

# Executive Summary

This report is one of a series of reports generated from the project, *Development of a phytobenthos classification tool for lakes and lochs of UK (DALES – Diatom assessment of lake and loch ecological status)*, funded by the Environment Agency. This project forms part of the strategy for the implementation of the EU Water Framework Directive (WFD), which requires reference conditions to be set for all water body types including lakes.

This palaeoecological study focuses on the analysis of diatom remains in sediment core samples from a set of lakes across England, Wales and Scotland. Diatoms are sensitive to water quality changes and shifts in the diatom community often correspond to changes in water quality and other biological elements. In the summer of 2004, short sediment cores were collected by the Environmental Change Research Centre (ECRC) on behalf of the Environment Agency from 100 lakes in England and Wales as part of the WFD Lake Monitoring Phase 2 programme, and by the Scottish Environmental Protection Agency (SEPA) from 27 Scottish lochs. Surface sediment samples from many of these cores have been analysed as part of DALES; however, this study provides further analysis of core bottom material from around 50 lakes.

In selecting a subset of sites, preference was given to natural lakes and sites not previously subject to down-core diatom analysis. The selection of lakes spanned the range of GB lake types (Phillips, 2004). For each site, the core bottom diatom assemblages were compared with those of the surface sediment diatom assemblages, to provide an estimate of floristic change from the reference condition. Diatoms were not preserved in all core bottom lake samples, so mid-core samples were analysed from sites where screening revealed some degree of change in diatom floristic composition between core top and bottom. Following screening for diatom preservation and the subsequent elimination of sites displaying diatom dissolution, core bottoms were analysed from 30 lakes (ten England, seven in Wales and thirteen in Scotland). Mid-core samples were also analysed from a subset of 20 of these sites (seven England, five Wales and eight Scotland), bringing the total number of mid and bottom samples analysed to 50.

This project builds on existing palaeoecological work in the UK, in particular the *Identification of reference lakes and evaluation of palaeoecological approaches to define reference conditions for UK (England, Wales, Scotland and Northern Ireland) ecotypes, WFD08* (Bennion, 2004).

Furthermore, this study complements additional palaeoecological work recently undertaken on thirteen English lakes of conservation interest (Bennion *et al.*, 2005) funded by English Nature, and ongoing site condition assessments of nineteen lakes in Welsh Special Areas of Conservation funded by Countryside Council for Wales (Goldsmith *et al.*, in preparation). The data provides information on reference conditions that can subsequently be added to the large database of diatom reference samples analysed as part of the recently completed SNIFFER-funded WFD08 Project (Bennion, 2004). The output of this project provides valuable information on reference and impact sites and enables broad assessment of ecological change at impacted sites.

## Results

The results of this study indicate that 17 of the 30 sites appear to have experienced significant floristic change in diatom species assemblages and 13 sites show minimal floristic change. The majority of changes appear to relate to increases in trophic status, although at some sites, floristic changes suggest increasing acidity (such as Llyn Bodlyn, Llyn Egnant and Llyn Ogwen) or decreasing acidity (Lochs Tormasad and Shnathaid).

Ten of the 30 lakes are thought to be good examples of reference lakes, given the minimal change in their diatom species assemblages and low squared chord distance dissimilarity distances between core bottom and top samples. Tal-y-llyn Lake (deep, low alkalinity) is the only non-Scottish example of a reference lake in this study. All other potential reference sites are Scottish lochs, covering all lake types except marl lakes for which there are no Scottish examples in this study. Loch Lagain and Loch Ascaig are examples of peat lake reference sites, Lochs Skerrols, Ailsh and nan Gabhar of low alkalinity, shallow reference lakes, Lochs Craggie and Hope of medium alkalinity, deep lakes and Lochan Lùnn Dà-Bhrà of a medium alkalinity, shallow lake; Loch Kinnabus is the only high alkalinity, deep lake reference site. Some lakes such as Lochs Kinnabus, Hope and Craggie and Lochan Lùnn Dà-Bhrà show early warning signs of slight increases in trophic status. We recommend that the water chemistry and ecology of these sites are monitored closely over the coming years, to ascertain whether a shift from reference conditions is occurring.

A further eight lakes are potential reference lakes based on the relatively low degree of floristic change observed in the cores: Loch na Moracha and Loch Shnathaid (peat, deep); Thirlmere Reservoir (low alkalinity, deep); Loch Borralan (low alkalinity, shallow); Loch Tormasad and The Mere, Ellesmere (high alkalinity, deep); Shear Water and Llyn Coron (high alkalinity, shallow). However, further investigation is necessary to confirm their status.

One limitation of this study is that none of the cores are dated and for some lakes where sediment cores are short, core bottom samples may not represent true 'reference' samples. This is of particular concern at the high alkalinity sites which lie in productive catchments (such as The Mere, Ellesmere, Shear Water and Llyn Coron) where sediment accumulation rates are expected to be high. The lack of a chronology is of less concern where the diatom assemblages remain stable throughout the core, but it becomes a greater limitation when interpreting data from sites that exhibit floristic change, as we have no estimate of the time at which the changes occurred. Sediment accumulation rates are site specific and it is unlikely that all cores cover comparable time periods. Longer cores would need to be collected and radiometric or spheroidal carbonaceous particle (SCP) dating carried out to provide a more detailed assessment of the nature and timing of such ecological changes. In addition, sites such as Loch Borralan, where no change between top and bottom was seen but where the assemblages comprised many nutrient-tolerant diatom taxa, may represent naturally meso-eutrophic reference sites and further analysis is advised. Analysis of remains of other biological elements preserved in lake sediment cores, such as plant macrofossils, cladocera and chironomids, would enable more holistic ecological reference conditions to be defined, and would provide valuable information on changes in ecological structure and function (see Sayer *et al.*, 1999; Bennion, 2001).

## Conclusion

In conclusion, this project shows that relatively low-resolution analysis of lake sediment cores and the use of simple techniques such as dissimilarity scores and ordination analyses to palaeoecological data can provide valuable information for defining ecological reference conditions and assessing deviation from the reference state at impacted sites. This information aids implementation of the WFD at the national level.

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# 1 Introduction

## 1.1 Study rationale and objectives

There is a pressing need for progress in our understanding of lakes in the UK. Annex II of the EU Water Framework Directive (WFD) requires the identification of candidate reference lakes, while Annex V calls for the development of tools to determine reference conditions and classify status (European Union, 2000). Furthermore, the EU Habitats and Species Directive require the setting of conservation objectives, which may in turn lead to the need for restoration targets for lakes. The use of palaeolimnological techniques could help meet these requirements. A number of recently completed and ongoing projects employ palaeolimnological methods to identify reference lakes, describe reference conditions and assess ecological status of UK lakes. For example, the project, *Identification of reference lakes and evaluation of palaeoecological approaches to define reference conditions for UK (England, Wales, Scotland and Northern Ireland) ecotypes, WFD08* (Bennion, 2004) involved the analysis of 219 core top and bottoms to identify reference lakes.

Sediment cores were taken by the Environmental Change Research Centre (ECRC) on behalf of the Environment Agency from 100 lakes in England and Wales in 2004 as part of the *Lake monitoring to support Environment Agency Water Framework Directive intercalibration exercise and classification tool development, and CCW Site Condition Assessment - Phase 2* programme. Around half of these lakes had not previously been the subject of down-core diatom analysis. Short cores were also collected by the Scottish Environmental Protection Agency (SEPA) from 27 Scottish lochs in the summer of 2004, with 20 never having been subject to palaeoecological analysis. The ongoing project *Development of a phytobenthos classification tool for lakes and lochs of UK, DALES* includes analysis of surface sediment samples from a number of these cores. The project reported here provides further analysis of core bottoms from a subset of 50 sites across England, Wales and Scotland.

This project used the 'top and bottom' approach, a low-resolution technique for broadly assessing the degree of change at a large numbers of sites. Where floristic change was exhibited between the core top and bottom samples, an additional mid-core sample was analysed. In the absence of core chronologies, the timing and rate of change could not be established, although the mid-core samples provided further insight into the point of change.

The objectives of the project were to:

1. Analyse subfossil diatoms in core bottoms from 50 sites across England, Wales and Scotland.
2. Compare the bottom and mid-core diatom assemblages to those of the surface sediment diatom assemblages already being analysed as part of DALES, to provide an estimate of floristic change from reference conditions.
3. Perform detrended correspondence analysis (DCA) and a dissimilarity measure to assess the nature and degree of change in the diatom assemblages.
4. Provide information on reference conditions, verify the choice of reference lakes and assess shifts in ecological status at impacted sites.



## 1.2 Background

The EU Water Framework Directive (WFD) 2000/60/EC came into force in 2000 (European Union, 2000) and aims to achieve good ecological quality in all relevant water bodies within 15 years. It requires biological, hydromorphological and chemical elements of water quality to be assessed by the degree to which present day conditions deviate from those expected in the absence of human influence, termed reference conditions. The WFD requires the determination of reference conditions for different water body types in order to identify sites of high status, where the various elements correspond totally or almost totally to undisturbed conditions. The four categories of good, moderate, poor and bad status are defined according to the degree of deviation from the reference state. In the absence of long-term data, the WFD states that reference conditions based on modelling may be derived using hindcasting methods, and palaeolimnology is given as one such technique (Pollard and Huxham, 1998; European Union, 2000).

The study of sediment accumulated in a water body can provide a record of its past biology and chemistry, a science known as palaeoecology. Diatoms (*Bacillariophyceae*: single-celled, siliceous algae) are commonly used in such studies because they are sensitive to water quality changes and are therefore, good indicators of past lake conditions such as lake pH (Battarbee *et al.*, 1999, 2001) and total phosphorus (TP) concentrations (Hall and Smol, 1999). Diatoms represent components of both the phytoplankton and phytobenthos, but importantly shifts in the diatom community often correspond to changes in other biological groups (Kingston *et al.*, 1992). The diatom record is a potentially useful tool for assessing water quality and defining lake reference conditions, both chemical and ecological (Kauppila *et al.*, 2002; Bennion *et al.*, 2004). The use of palaeoecological techniques for determining reference conditions and assessing ecological change in lakes is well established, with many examples of their use in aquatic management and conservation (Battarbee, 1999; Stoermer and Smol, 1999; Bennion *et al.*, 1996, 2004).

The top and bottom approach involves the analysis of only two samples per site from a sediment core (Cumming *et al.*, 1992). This approach has been successfully used by the US Environmental Protection Agency's (USEPA) Environmental Monitoring and Assessment Program for Surface Waters (EMAP-SW) (Dixit *et al.*, 1999) and in south-eastern Ontario lakes of Canada (Reavie *et al.*, 2002). The approach assumes that the top and bottom samples represent the present day and reference conditions, respectively. The analysis of reference samples in this way removes the problem inherent in spatial-state schemes in which the lakes have been subject to different pressures and varying degrees of impact. For the UK, it is generally agreed that roughly 1850 AD is a suitable date against which to assess impacts for lakes, as this represents a period prior to major industrialisation and agricultural intensification (Battarbee, 1999; Fozzard *et al.*, 1999). However, it is accepted that aquatic systems have been subjected to anthropogenic impacts over much longer timescales than simply the last 100-150 years and, therefore, our reference conditions are unlikely to equate to the natural or pristine state. The diatom data from the surface (uppermost 0.5 or 1 cm) sample of each core is used to provide information on the current diatom assemblages of the lakes, since this represents the last few years accumulation of diatoms deposited from a variety of habitats within the lake. The bottom sample of the core is taken to represent the reference conditions, although in the absence of core chronologies, there are uncertainties as to whether the bottom samples represent true reference samples.

## 1.3 Report structure

The report is comprised of four main sections: 1) introduction, study rationale and background; 2) methodology; 3) presentation and discussion of the results; and 4) a summary of the findings and suggestions for further work.

## 2 Methods

### 2.1 Site selection

Short cores were collected by SEPA from 27 Scottish lochs in the summer of 2004, approximately 20 of which had never been subjected to palaeoecological analysis. Short cores were also collected by the ECRC on behalf of the Environment Agency from 100 lakes in England and Wales in the summer of 2004, of which approximately 50 lakes had not previously been the subject of down-core diatom analysis. Following elimination of most artificial reservoir sites, a total of 44 lakes in England, Wales and Scotland were shortlisted for inclusion in this project. Cores from five lakes were deemed too short and were excluded immediately, and following further screening, a subset of 30 sites (10 English, seven Welsh and 13 Scottish) underwent analysis. The site characteristics are given in Table 2.1 and details of the cores and analysis undertaken on each site in Table 2.2. The sites represent a range of lake types in the GB lake typology (Phillips, 2004), including peat (P) (four sites), low alkalinity (LA) (11 sites), medium alkalinity (MA) (six sites), high alkalinity (HA) (eight sites) and marl (one site) systems, and with examples of both shallow (Sh) and deep (D) waters (11 and 19 sites, respectively) (see Tables 2.1 and 2.2). Lakes of low alkalinity are best represented. Most of the lakes are circumneutral to alkaline and even the low alkalinity waters are only mildly acid. With the exception of Llyn Bodlyn, Llyn Egnant and Llyn Ogwen which are at altitudes above 200 m, the study lakes lie in relatively productive, lowland catchments and hence eutrophication is likely to be the key pressure. Further details of the sites and their chemistry are given in Table 2.1.

### 2.2 Field and laboratory methods

A sediment core was taken in the summer of 2004 from the deepest part of each lake using a Glew gravity corer which collects short cores of typically 20-40 cm in length. It might be expected, therefore, that at very productive sites, the short cores would not extend back as far as 100 years. Based on previous palaeolimnological research of dated sediment cores (Bennion, 2004), it is estimated that for low alkalinity lakes, where production is lower and sediment accumulation rates slower, a sediment depth of 20-30 cm dates back to around 1850. In higher alkalinity sites, where sediment accumulation rates are faster due to higher productivity, a sediment depth of around 50 cm dates back to 1850, and in the highly productive Cheshire-Shropshire Meres, the depth required to extend back to approximately 1850 is typically 60-70 cm. All cores were extruded in the field at either 0.5 cm or 1.0 cm intervals (depending on expected sediment accumulation rate).

Cores from five of the 44 lakes were deemed too short to provide reasonable core bottom samples to approximate reference conditions. This resulted in a suite of 39 shortlisted sites (see Table 2.2). Surface sediment samples had previously been prepared as part of *Development of a phytobenthos classification tool for lakes and lochs, DALES*, hence slides for diatom analysis were prepared from the bottom of each of the 39 cores (see Table 2.2), using standard methods (Battarbee *et al.*, 2001). Screening of the slides revealed that nine sites had extremely poor diatom preservation in the bottom samples, thus excluding these sites from analysis. Therefore, diatom analysis was carried out at only 30 lakes. Screening also revealed that some sites exhibited greater shifts in floristic composition than others. Therefore, for a subset of 20 of the 30 lakes, mid-core samples were also prepared for diatom analysis to allow the general trend in water quality to be determined. This resulted in a total of 50 samples for diatom analysis as part of this project, comprising 20 mid-core

samples and 30 core bottom samples. At least 300 valves (siliceous component of the cell wall bearing the taxonomic features) were counted from each sample using a Leitz research microscope with a 100x oil immersion objective and phase contrast. Principal floras used in identification were Krammer and Lange-Bertalot (1986-1991). All slides are archived at the ECRC and the data are stored in the Amphora database.

## 2.3 Data analysis

All diatom data were expressed as percentage relative abundance, and were screened and harmonised prior to data analysis. The 50 samples analysed here were harmonised with the surface samples of the 30 study lakes. The full dataset of 80 samples (30 cores, with two samples from 10 cores and three samples from 20 cores) comprised 398 diatom taxa. The most common 203 taxa (occurring at more than one per cent in more than two samples) are listed in Appendix 1. Summary diagrams of the diatom assemblages in the cores from each site (showing only those taxa present with a relative abundance of more than 5 per cent in at least one sample) were produced for each lake type (the one marl site is included in the HA diagram) using  $C^2$  (Juggins, 2003) – see Figures 2.1 to 2.4.

The degree of floristic change between the bottom (reference) sample and the surface (and mid) sample analysed in each core was assessed using the squared chord distance dissimilarity coefficient (Overpeck *et al.*, 1985) in the statistical software R (R Development Core Team, 2004). This is preferred to other dissimilarity measures as it maximises the signal to noise ratio, performs well with percentage data and has sound mathematical properties (Overpeck *et al.*, 1985). The scores range from zero to two. Scores less than 0.29, 0.39, 0.48 and 0.58 indicate insignificant floristic change at the first, 2.5th, fifth and 10th percentile, respectively (Simpson, 2003). The fifth percentile (score <0.475) is used here to define sites with low floristic change between the bottom (reference) sample and surface (and mid) sample. The scores are plotted for each lake in the form of bar graphs in Figures 2.1-2.4, to show how dissimilar the surface and mid samples are from the bottom (reference) sample. The actual values are shown in Appendix 2. The vertical line in Figures 2.1-2.4 is drawn at a squared chord distance dissimilarity score of 0.475 to illustrate which samples fall above and below this critical value.

For each lake type [P, LA, MA and HA (including one marl site)], detrended correspondence analysis (DCA) (Hill and Gauch, 1980) was performed using CANOCO version 4.5 (ter Braak and Smilauer, 2002) to assess the direction and magnitude of floristic change at each site. Only those 77, 127, 90 and 97 diatom taxa present with a maximum relative abundance of above one per cent in more than two samples, for P, LA, MA and HA lake types respectively, were included in the ordination analyses. The results are presented as biplots of Axis 1 and 2 sample scores and species scores in Figures 2.5-2.8. Samples with similar scores on the two axes lie in close proximity, reflecting similar diatom composition. For each core, lines connect the samples in a series from core bottom to core top (see Appendix 2 for sample codes). The direction of the line indicates the direction of floristic change and its length is a measure of species turnover in Hill's standard deviation units (Hill and Gauch, 1980). For species codes see Appendix 1.

The results are detailed below for each lake type. For each site, the major species shifts are described, the degree of floristic change is presented and an interpretation of the floristic changes is given.

**Table 2.1 Summary site characteristics of the 44 lakes shortlisted for analysis**

GB Lakes WBID	Name	Altitude (m.a.s.l.)	Surface area (ha)	<sup>1</sup> Max depth (m)	<sup>2</sup> GB Lake type		pH	Cond (µS/cm)	Alk (mg/L)	TP (µg/L)	SRP (µg/L)	TN (mg/L)	Chla (µg/L)	Si (mg/L)
12578	Loch an Lagain	136	27.7		P	D								
15316	Loch na Moracha	4	36.4		P	D								
18113	Loch Shnathaid	4	23.2		P	D								
8945	Loch Ascaig	135	27.1		P	D								
11611	Loch Brora	25	66.5		P	D								
29184	Grasmere	61	60.7	20.8	LA	D	6.99	45	12.74	20	3	0.41	7	1.31
35561	Llyn Bodlyn	385	16.5	20.0	LA	D	6.40	30	2.25	14	8	0.55	1	0.55
38409	Llyn Egnant	420	13.9	14.2	LA	D	6.12	32	1.88	23	9	4.13	3	0.43
33803	Llyn Ogwen	300	38.5	2.7	LA	D	7.03	25	3.65	11	6	0.36	2	0.58
33730	Llyn Padarn	105	97.6	27.0	LA	D	7.46	45	8.70	19	8	0.38	6	1.28
36405	Tal-y-llyn Lake	85	50.7	3.5	LA	D	7.34	42	8.44	29	9	0.55	6	1.20
29021	Thirlmere Reservoir	178	313.3	24.0	LA	D	7.15	32	5.00	19	1	37.01	3	1.46
11355	Loch Borralan	142	47.0		LA	Sh								
26257	Loch Skerrols	25	26.1		LA	Sh								
1138	Loch Ailsh	154	105.2		LA	Sh								
2257	Loch nan Gabhar	1	16.6		LA	Sh								
5714	Loch Rangag	117	31.6		LA	Sh								
16530	Loch Gowan	156	18.2		LA	Sh								
29321	Coniston Water	46	470.5	36.0	MA	D	7.19	61	13.54	22	1	0.49	5	0.69
29233	Windermere	37	1435.9	32.8	MA	D	7.96	66	16.76	23	6	0.47	9	0.76
11642	Loch Craggie	166	54.2		MA	D								
2490	Loch Hope	4	638.3		MA	D								
18682	Loch Druidbeag	7	256.5		MA	D								
29222	Elter Water	53	18.2	7.7	MA	Sh	6.98	48	12.46	18	2	0.43	-	1.85
22395	Lochan Lùnn Dà-Bhrà	156	26.0		MA	Sh								
4974	Loch Syre	122	44.0		MA	Sh								
32538	Llyn Alaw	42	308.4	3.8	MA	Sh	7.72	186	41.77	37	14	1.52	11	1.43

GB Lakes WBID	Name	Altitude (m.a.s.l.)	Surface area (ha)	<sup>1</sup> Max depth (m)	<sup>2</sup> GB Lake type		pH	Cond (µS/cm)	Alk (mg/L)	TP (µg/L)	SRP (µg/L)	TN (mg/L)	Chla (µg/L)	Si (mg/L)
28386	Talkin Tarn	128	25.5	12.5	Marl	D	8.12	122	43.97	51	9	1.71	25	1.51
15551	Loch Tormasad	8	21.1		HA	D								
26944	Loch Kinnabus	77	43.7		HA	D								
34990	The Mere, Ellesmere	98	43.4	18.0	HA	D	7.86	282	113.80	954	766	1.48	16	1.93
25899	Ardnave Loch	18	11.1		HA	D								
26178	Loch Ballygrant	77	26.6		HA	D								
43135	Blagdon Lake	45	164.6	9.0	HA	D	8.30	406	167.25	236	34	2.95	62	5.90
6405	Loch Meadie	116	39		HA	D								
2499	Loch Scarmclate	25	75.9		HA	D								
44518	Fonthill Lake	94	2.5	5.1	HA	Sh	8.06	453	200.67	31	16	5.20	30	6.44
43909	Shear Water	139	13.7	7.2	HA	Sh	8.75	235	70.63	31	8	2.42	78	10.56
32948	Llyn Dinam	8	9.7	1.5	HA	Sh	7.76	355	76.81	73	35	1.68	12	2.91
30244	Hornsea Mere	8	133.3	2.0	HA	Sh	8.22	509	170.42	500	-	2.01	26	4.26
33337	Llyn Coron	9	28.0	3.8	HA	Sh	8.05	311	97.00	106	56	3.45	9	6.75
32968	Llyn Penrhyn	8	22.3	2.6	HA	Sh	7.41	403	93.00	426	332	1.11	19	3.74
33627	Llyn Rhos-ddu	8	2.4	1.0	HA	Sh	7.43	348	134.37	54	28	1.22	22	8.93
2088	Loch of Mey	15	23		HA	Sh								

<sup>1</sup> Maximum depths given are those measured at the coring location. Note that these may not always be the absolute deepest point.

<sup>2</sup> GB lake type follows the scheme of Phillips (2004); LA, MA, HA = low, medium and high alkalinity, respectively; Sh = shallow, D = deep. Chemical data are given as annual means calculated from the Environment Agency WFD 2003-2004 dataset.

**Table 2.2** Details of cores and analysis undertaken at the 44 shortlisted lakes – only 30 sites included in the final analysis

GB lakes WBID	Name	Grid reference	<sup>1</sup> GB lake type		<sup>2</sup> Core code	Coring date	Core length (cm)	Sample intervals analysed for diatoms (cm)
12578	Loch an Lagain	NH658955	P	D	LAGN1	07.09.04	20	0, 10, 20
15316	Loch na Moracha	NF846663	P	D	MORA1	27.09.04	20	0, 10, 20
18113	Loch Shnathaid	NF826426	P	D	SHNA1	29.09.04	20	0, 10, 20
8945	Loch Ascaig	NC849255	P	D	ASCA1	24.08.04	20	0, 20
11611	Loch Brora	NC852078	P	D	BROR1	24.08.04	1	None**
29184	Grasmere	NY338065	LA	D	GRAS1	25.07.04	28	0, 10, 27
35561	Llyn Bodlyn	SH648239	LA	D	BODL1	14.09.04	23	0, 10, 22
38409	Llyn Egnant	SN792671	LA	D	EGNA1	24.09.04	31	0, 15, 30
33803	Llyn Ogwen	SH659604	LA	D	OGWE1	20.08.04	25	0, 10, 25
33730	Llyn Padarn	SH569614	LA	D	PADA1	21.08.04	25	0, 10, 25
36405	Tal-y-llyn Lake	ST850421	LA	D	TALY1	18.09.04	21	0, 20
29021	Thirlmere Reservoir	NY313162	LA	D	THIR1	22.07.04	31	0, 30
11355	Loch Borralan	NC262108	LA	Sh	BORL1	29.06.04	15	0, 10, 15
26257	Loch Skerrols	NR341638	LA	Sh	SKEL1	14.07.04	20	0, 10, 20
1138	Loch Ailsh	NC315109	LA	Sh	AILS1	30.06.04	40	0, 40
2257	Loch nan Gabhar	NM968632	LA	Sh	GABH1	18.08.04	15	0, 15
5714	Loch Rangag	ND177415	LA	Sh	RANG1	25.08.04	25	None*
16530	Loch Gowan	NH152564	LA	Sh	GOWA1	10.08.04	30	None*
29321	Coniston Water	SD301940	MA	D	CONI1	25.07.04	36	0, 20, 35
29233	Windermere	SD392958	MA	D	WIND1	26.07.04	31	0, 20, 30
11642	Loch Craggie	NC624074	MA	D	CRA4	11.07.04	30	0, 10, 30
2490	Loch Hope	NC463548	MA	D	HOPL1	07.07.04	20	0, 20
18682	Loch Druidbeag	NF789376	MA	D	DRUI1	28.09.04	10	None**
29222	Elter Water	NY333041	MA	Sh	ELTW1	26.07.04	21	0, 10, 20
22395	Lochan Lùnn Dà-Bhrà	NN087659	MA	Sh	LUNN1	17.08.04	20	0, 10, 20
4974	Loch Syre	NC661448	MA	Sh	SYRE1	08.07.04	25	None*
32538	Llyn Alaw	SH392866	MA	Sh	ALAW1	16.08.04	20	None*

GB lakes WBID	Name	Grid reference	<sup>1</sup> GB lake type		<sup>2</sup> Core code	Coring date	Core length (cm)	Sample intervals analysed for diatoms (cm)
28386	Talkin Tarn	NY545587	Marl	D	CZNYSSB1 (TALK)	22.07.04	31	0, 20, 30
15551	Loch Tormasad	NF820651	HA	D	TORM1	26.09.04	20	0, 10, 20
26944	Loch Kinnabus	NR301422	HA	D	KINB1	21.07.04	20	0, 20
34990	The Mere, Ellesmere	SJ406349	HA	D	SCM04B (ELLE)	11.08.04	35	0, 35
25899	Ardnave Loch	NR284727	HA	D	ARDN2	13.07.04	20	None*
26178	Loch Ballygrant	NR405662	HA	D	BALG3	22.07.04	20	None*
43135	Blagdon Lake	ST515596	HA	D	BLAG1	17.09.04	23	None*
6405	Loch Meadie	NC502410	HA	D	MEAD1	15.09.04	10	None**
2499	Loch Scarmclate	ND189596	HA	D	SCAM1	26.08.04	10	None**
44518	Fonthill Lake	ST937311	HA	Sh	FONT1	30.09.04	21	0, 10, 20
43909	Shear Water	ST850421	HA	Sh	SST84_1 (SHEA)	14.09.04	26	0, 15, 25
32948	Llyn Dinam	SH310775	HA	Sh	DINA2	16.08.04	20	0, 10, 20
30244	Hornsea Mere	TA190469	HA	Sh	HORN2	15.07.04	27	0, 26
33337	Llyn Coron	SH378700	HA	Sh	CORO2	16.08.04	20	0, 20
32968	Llyn Penrhyn	SH313768	HA	Sh	PERH2	16.08.04	30	None*
33627	Llyn Rhos-ddu	SH424648	HA	Sh	RHSD1	17.08.04	25	None*
2088	Loch of Mey	ND271736	HA	Sh	MEY1	14.09.04	10	None**

\* Diatom preservation too poor for analysis of core bottom sample (core tops analysed within DALES)

\*\* Cores too short for top and bottom analysis

<sup>1</sup> GB lake type follows the scheme of Phillips (2004); LA, MA, HA = low, medium and high alkalinity, respectively; Sh = shallow, D = deep.

<sup>2</sup> Core code is the AMPHORA core code. For some sites, simpler, alternative codes (noted in parentheses) have been used in all figures.



# 3 Results and discussion

## 3.1 Peat (P) lakes

A summary diagram of the common diatom taxa found in the samples from the peat lake type (occurring at more than five per cent relative abundance in more than one sample) is illustrated in Figure 3.1. DCA biplots (Axis 1 and 2) of the sample and species scores for the peat lakes are displayed in Figure 3.5.

The four peat lakes are all in Scotland and are deep. The sites have fairly similar circumneutral to acidophilous diatom assemblages, dominated by *Fragilaria exigua*. Other diatom taxa common in the P lakes include *Achnanthes minutissima*, *Brachysira vitrea*, *Cymbella graciis*, *Tabellaria flocculosa*, *Eunotia incisa*, *Frustulia rhomboides*, small *Fragilaria* spp. and small *Navicula* spp. (*vitiosa* and *arvensis*). Overall, the diatom assemblages of the four peat lakes do not show much change from core bottoms to core tops. Loch Ascaig has the lowest squared chord distance dissimilarity score at 0.318, with the distances of the other sites all being relatively low, lying close to the critical value of 0.475 and supporting the observed floristic stability.

Loch na Moracha shows the greatest change from core bottom to top, with the planktonic diatom, *Cyclotella comensis* decreasing from 20 per cent relative abundance in the bottom and mid-core samples, to below one per cent in the top. In Loch an Lagain's surface sediment sample, there is an increase in relative abundance of small benthic (sediment-dwelling) *Fragilaria* spp. at the expense of the epiphytic diatoms, *A. minutissima* and *B. vitrea*. This could indicate a decrease in aquatic macrophyte abundance. Loch Shnathaid may have experienced a slight reduction in acidity from core bottom to top. *F. exigua* and *E. incisa* (acidophilous taxa) decrease in relative abundance, whilst *A. minutissima*, *B. vitrea* and *T. flocculosa* (circumneutral-acidophilous taxa) increase in relative abundance. Caution should be taken in interpreting the data from the peat lakes, because all four cores were only 20 cm long and may be too short to represent reference conditions at the core base.

## 3.2 Low alkalinity (LA) lakes, deep and shallow

A summary diagram of the common diatom taxa found in the samples from the low alkalinity lake types (occurring at more than five per cent relative abundance in more than one sample) is illustrated in Figure 3.2. DCA biplots (Axis 1 and 2) of the sample and species scores for the low alkalinity lakes are displayed in Figure 3.6. The majority of lakes of the LA type are dominated by circumneutral, non-planktonic diatom taxa, although planktonic taxa are also common.

Grasmere and Llyn Padarn would appear to have experienced significant change from core bottom to top. In Grasmere, the dominant taxon in the core bottom is *A. minutissima*, whereas in the mid and surface samples, *A. formosa* constitutes a higher relative abundance. *C. comensis* only occurs in the bottom sample. An increasing abundance of *A. formosa* at the expense of *C. comensis* and *A. minutissima* is usually interpreted as indicating increased nutrient status, since *A. formosa* frequently appears in formerly oligotrophic lakes as a sign of enrichment. The changes also indicate a shift from a largely periphytic diatom community to a plankton-dominated community. The relatively high squared chord dissimilarity distance (0.65) (Figure 3.2) and the shift in sample scores (Figure 3.6) between the top and bottom samples of Grasmere support the inference of significant change in diatom species assemblages. In Llyn Padarn, the diatom assemblage appears to

have shifted from dominance of *C. comensis*, towards co-dominance of other planktonic diatom taxa such as *T. flocculosa*, *Aulacoseira subarctica*, *A. formosa* and *Cyclotella pseudostelligera*. Thirlmere Reservoir may tentatively be considered a reference site, since the diatom assemblage of *A. minutissima* and *C. comensis/rossii* has remained relatively constant from core bottom to top. In common with Grasmere, the appearance of *A. formosa* in the surface sediment sample may indicate a slight shift towards more mesotrophic conditions.

Three of the LA deep Welsh lakes, Llyn Bodlyn, Llyn Egnant and Llyn Ogwen have experienced significant changes in species composition from core bottom to top. These changes would appear to be indicative of increasing acidity (decrease in pH). For Llyn Bodlyn and Llyn Egnant, Figure 3.6 clearly illustrates the shift in sample scores from the right to the left of the diagram, moving from core bottom to top. This corresponds to a shift from predominantly circumneutral, non-planktonic taxa to an increase in relative abundance of circumneutral acidophilous, non-planktonic and planktonic taxa. In terms of diatom species, the community has changed from an *A. minutissima*-dominated community, to one dominated by *E. incisa* and *T. flocculosa* (Llyn Egnant), with *Cymbella perpusilla* (Llyn Egnant) and *Peronia fibula* (Llyn Bodlyn) also increasing in relative abundance. At Llyn Bodlyn and Llyn Egnant there has also been a loss of *C. comensis* and *Cyclotella rossii*, respectively. Care should be taken in interpreting the shift towards *T. flocculosa* in the surface sediments of both Llyn Bodlyn and Llyn Egnant, because *T. flocculosa* is a bloom-forming planktonic taxon and is a frequent component of autumn diatom blooms. Since cores from these sites were taken in September, it is likely that *T. flocculosa* is overrepresented in the surface sediments of the cores from these sites. It may be worth examining a sample from a depth of one to two cm to determine whether the increase in *T. flocculosa* in the core tops is merely a seasonal artefact.

Lochs Ailsh (LA, Sh), nan Gabhar (LA, Sh) and Tal-y-llyn Lake (LA, D) do not appear to have experienced significant diatom assemblage changes from core bottom to top and these sites are suggested to be good examples of LA reference sites. The sites are dominated by non-planktonic, circumneutral to acidic diatom taxa. Species diversity is high, with a broad range of periphytic taxa (*A. minutissima* usually dominating, with *F. exigua*, *B. vitrea*, *Synedra rumpens*, *Cymbella minuta* and *E. incisa* co-occurring in differing proportions in the different sites) and the presence of the planktonic diatom taxon, *T. flocculosa*. The high diversity of the diatom assemblages, likely brought about by the wide range of habitats, may explain the relative stability of these sites, in much the same way as high macrophyte species diversity plays a key structuring role in lakes (Carpenter and Lodge, 1986; Jeppesen *et al.*, 1997). The squared chord dissimilarity distances between the top and bottom samples of these sites all lie below 0.475, supporting the inference of little change in diatom species assemblages. Furthermore, the top and bottom sample scores (displayed in Figure 3.6) lie in close proximity, indicating similar species assemblages.

Visual interpretation of the diatom profile from Loch Skerrols indicates that the core is dominated by a high diversity of periphytic taxa (Figure 3.2). The record appears to be relatively stable, with *A. minutissima* at a consistent relative abundance throughout the core. The squared chord dissimilarity distance is highest (0.647) in the mid-core sample, probably due to a slight shift towards increased relative abundances of small *Fragilaria* spp. and *Gomphonema pumilum*. In the surface sediment sample, the epiphytic diatom, *Cocconeis placentula* is dominant. These shifts may merely indicate subtle changes in the availability of different periphytic diatom habitats within a clear-water lake, as opposed to a significant shift towards a different ecological state. The ongoing work of DALES may elucidate this observation through the analysis of seasonal periphytic diatom samples. The data suggest that Loch Skerrols is an example of a LA, Sh reference site.

Although Loch Borralan displays relatively little change in its diatom species assemblage and has low squared chord dissimilarity distances between its top and bottom samples, *A. formosa*, *Fragilaria crotonensis*, *Aulacoseira granulata* var. *angustissima* and *Stephanodiscus parvus* are consistent components of the diatom community in all core samples. These planktonic taxa are considered to be indicators of mesotrophic to eutrophic conditions and it is therefore unlikely that this site can be considered an example of a LA, Sh reference site. Furthermore, the apparent stability in the diatom assemblage of this site may arise because the core is only 15 cm long, and is probably insufficient to extend back to baseline conditions unless the sedimentation rate is extremely low. Alternatively, Loch Borralan may be a naturally meso-eutrophic LA, Sh lake.

### 3.3 Medium alkalinity (MA) lakes, deep and shallow

A summary diagram of the common diatom taxa found in the samples from the medium alkalinity lake types (occurring at more than five per cent relative abundance in more than one sample) is illustrated in Figure 3.3. DCA biplots (Axis 1 and 2) of the sample and species scores for the medium alkalinity lakes are displayed in Figure 3.7.

Lake Windermere (MA, D) has experienced the greatest change of all MA sites in this study. The diatom assemblage of this lake has shifted from dominance of the oligotrophic, planktonic taxon, *C. comensis* (and *C. krammeri*) in the bottom and mid-core samples, to the co-occurrence of the mesotrophic indicators, *A. formosa*, *A. subarctica*, *F. crotonensis*, *Aulacoseira islandica* and *S. parvus* in the surface sediment sample. In addition, the relative abundance of *A. minutissima* has decreased and *Cyclotella radiosia* has been eliminated. These shifts are indicative of nutrient enrichment, a phenomenon well documented for Windermere based on long-term monitoring data (Reynolds and Irish, 2000) and detailed palaeolimnological studies (Sabater and Haworth, 1995). The dissimilarity score between the bottom (30 cm) and mid-core (20 cm) sample is <0.475 and the DCA biplot sample scores are similar, indicating little change in the lower part of the profile. In contrast, the high squared chord distance dissimilarity score (1.17) between the bottom and top samples and the position of the top sample at a distance from the mid and bottom core samples in the DCA biplot (Figure 3.7) both indicate significant floristic change in recent times. Coniston Water (MA, D) has experienced similar floristic changes indicative of an increase in productivity but to a lesser extent than Windermere, with dissimilarity scores between the bottom sample and upper samples of around 0.7.

Elter Water (MA, Sh), appears to have experienced a shift from periphyton dominance to plankton dominance. *A. minutissima* has decreased in relative abundance and *A. formosa*, *C. stelligera/pseudostelligera* and *Cyclostephanos invisitatus* have increased in relative abundance (Figure 3.3). Since Elter Water is shallow, the change from periphyton to plankton may be indicative of a shift from the clear to turbid water state (the alternative stable states theory of Scheffer *et al.*, 1993). The species shifts are reflected in the squared chord distance dissimilarity scores. The dissimilarity scores between the bottom sample and the 20 cm and 10 cm samples are both in excess of the critical value. The sample scores in the DCA biplot (Figure 3.7) move from right to left, reflecting the compositional change in the sediment record and the large distances between data points reflect the large degree of change. Lochan Lùnn Dà-Bhrà is the other MA, Sh site included in this project and would appear to be a good example of a MA, Sh reference site. *A. minutissima* is dominant from core bottom to top, with a stable community of small *Fragilaria* spp., *B. vitrea*, *F. exigua* and *Synedra tenera/nana* occurring at low relative abundances (Figure 3.3). Although *A. formosa* does not appear in the surface sample, *C. comensis* is absent. The loch should be monitored in future years to determine whether the loss of *C. comensis* continues and whether this change represents an ecological shift.

Upon visual inspection of Figure 3.3, Loch Hope (MA, D) appears to be a potential reference site. This site's diatom assemblage has changed little between core bottom and top, displaying co-dominance of *A. minutissima* and *C. comensis/rossii* in both samples and co-occurrence of *T. flocculosa*, *B. vitrea* and *S. tenera/nana*. Loch Hope also displays a squared chord distance dissimilarity score of <0.475 and its sample scores are very similar, with the top and bottom samples lying in close proximity on the DCA biplot (Figure 3.7). However, the appearance of *A. formosa* in the surface sediment sample may provide evidence of a slight increase in nutrients. Loch Craggie (MA, D) also shows minimal change between core top and bottom (30 cm) samples, with *Fragilaria* spp. dominating throughout, alongside the small *Navicula* species, *Navicula vitiosa*. Any shifts in species abundances occur between different small *Fragilaria* spp. As in the case of Loch Hope, planktonic diatom taxa (including *A. formosa*, *C. pseudostelligera* and *A. subarctica*) are found at very low relative abundances in the upper core samples (10 cm and surface sediment), perhaps providing a subtle warning of the potential for nutrient enrichment at this site. Overall, however, both Loch Hope and Loch Craggie provide the best examples of MA, D reference lakes in this study.

### 3.4 High alkalinity (HA), deep and shallow lakes, and marl lakes

A summary diagram of the common diatom taxa found in the samples from the high alkalinity and marl lake types (occurring at more than five per cent relative abundance in more than one sample) are illustrated in Figure 3.4. DCA biplots (Axis 1 and 2) of the sample and species scores for the high alkalinity and marl lakes are displayed in Figure 3.8.

The one marl lake included in the current study, Talkin Tarn, appears to have experienced moderate change from core bottom to top, with nutrient enrichment the likely cause. The shift in the diatom species assemblage is from *C. radiosa*, a mesotrophic planktonic taxon dominant in the core bottom (30 cm) and mid (20 cm) samples, to the more nutrient-tolerant *S. parvus*, *Fragilaria capucina* var. *mesolepta*, *Cyclostephanos dubius*, *Stephanodiscus hantzschii* and *A. granulata* var. *angustissima* in the surface sediment sample (Figure 3.4 and Figure 3.8). The species shifts are reflected in the squared chord distance dissimilarity scores. Although there is little change between the bottom and mid-core samples (0.330), the dissimilarity between the bottom and top samples is considerably higher (0.784). Furthermore, there is little difference in the sample scores in the DCA biplot (Figure 3.8) between 30 and 20 cm, but between 30 and zero cm, the sample scores move from lower left to upper right reflecting the compositional change in the diatom species assemblage. These results indicate that the lake's diatom community was stable until recent times.

Of the eight high alkalinity lakes, three are deep (Loch Tormasad, Loch Kinnabus and The Mere, Ellesmere) and five are shallow (Fonthill Lake, Shear Water, Llyn Dinam, Hornsea Mere and Llyn Coron). The majority of the HA sites appear to have experienced significant change from core bottom to top. Fonthill Lake has seen a shift from a small *Fragilaria* spp. community to one in which the planktonic diatoms *Aulacoseira granulata* (and var. *angustissima*) and *F. crotonensis* all occur at increasing relative abundance. This species shift indicates nutrient enrichment and a switch from clear to turbid water conditions (the alternative stable states theory of Scheffer *et al.*, 1993). Planktonic diatoms have dominated Shear Water throughout the core with *C. dubius* being dominant and *S. hantzschii* being sub-dominant. The squared chord distance score of 0.848 between core bottom and top samples reflects changes in the planktonic diatom taxa, with a shift from *F. crotonensis* and *A. formosa* to *C. radiosa* and *A. subarctica* (Figures 3.4 and 3.8). These taxa are all considered mesotrophic indicators and therefore the shifts may simply reflect inter-annual variation. It is

recommended that a longer core is taken from Shear Water for a higher resolution investigation to determine ecological state, and in particular to determine whether the lake is a potential reference site. Llyn Dinam appears to have experienced an increase in the relative abundance of epiphytic (plant-dwelling) diatom taxa (*C. placentula* and *Rhoicosphenia abbreviata*) alongside a decreasing relative abundance of small benthic (sediment-dwelling) *Fragilaria* spp. and a corresponding increase in the relative abundance of small planktonic centric diatom taxa (*S. parvus*, *Cyclostephanos invisitatus* /*tholiformis*). Llyn Dinam may therefore have experienced a slight shift away from the clear-water state, increasing the available habitat for planktonic diatoms. In addition, this site may have seen an increase in the available habitat for the growth of *C. placentula* and *R. abbreviata*, namely an increase in the infestation of aquatic macrophytes or filamentous algae. Work being undertaken as part of DALES may shed light on the habitat preferences of *C. placentula* and *R. abbreviata*, enabling better interpretation of the observed shifts in Llyn Dinam's diatom species composition.

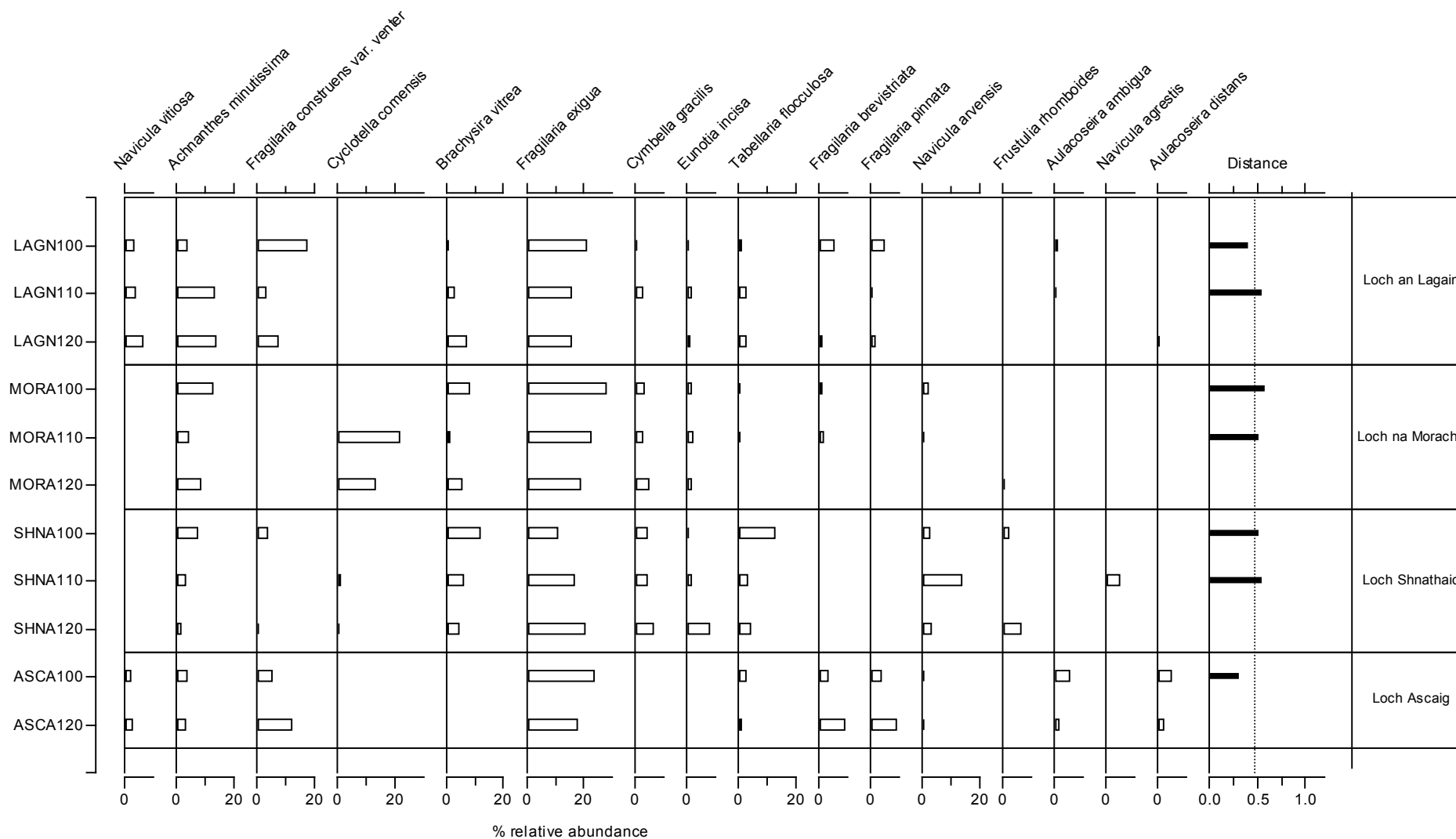
Loch Tormasad's diatom profile is difficult to interpret, since there are no major shifts in floristic composition, despite significant squared chord distance dissimilarity scores of 0.891 (core bottom to top) and 0.686 (core bottom to middle). There is a possibility that this site has seen a slight decrease in acidity from core bottom to top, since the relative abundance of *F. exigua* decreases as that of small *Fragilaria* spp. typically associated with more circumneutral to alkaline waters, and *A. minutissima* increase (Figure 3.4). To investigate the possibility of pH shifts at this site, it may be worth applying a pH transfer function to the diatom samples of Loch Tormasad. In terms of nutrients, this site could potentially be an example of a reference site, although further investigation is recommended using a longer sediment core to provide a deeper bottom sample.

The short distances between core bottom and top samples in the DCA biplot (Figure 3.8) show that the diatom communities of four of the HA lakes - Loch Kinnabus, Hornsea Mere, Ellesmere and Llyn Coron - remain relatively stable from core bottom to top, perhaps indicating that these lakes can be classified as reference sites. However, the data must be interpreted with caution because all cores were short (20 to 35 cm). For productive, HA lowland systems, where sediment accumulation rates are relatively rapid, it is unlikely that cores of only 20 to 35 cm in length are sufficient to extend back to 1850. Although Ellesmere has similar top and bottom diatom assemblages (Figure 3.4), it is recommended that bottom samples from longer sediment cores are investigated prior to making a decision as to the status of this lake. Hornsea Mere has been the subject of a previous palaeoecological study of a longer core. Nutrient-related shifts in diatom species assemblages were observed and the squared chord distance between core top and bottom (40 cm) was 0.893 (Bennion, 2004). Therefore, Hornsea Mere cannot be considered a reference site. Poor diatom preservation in Llyn Coron hindered data analysis and the results must be interpreted with caution. It is suggested that at this site, separate studies are conducted using indicators other than diatoms (such as cladocera, plant macrofossils), to further assess ecological status.

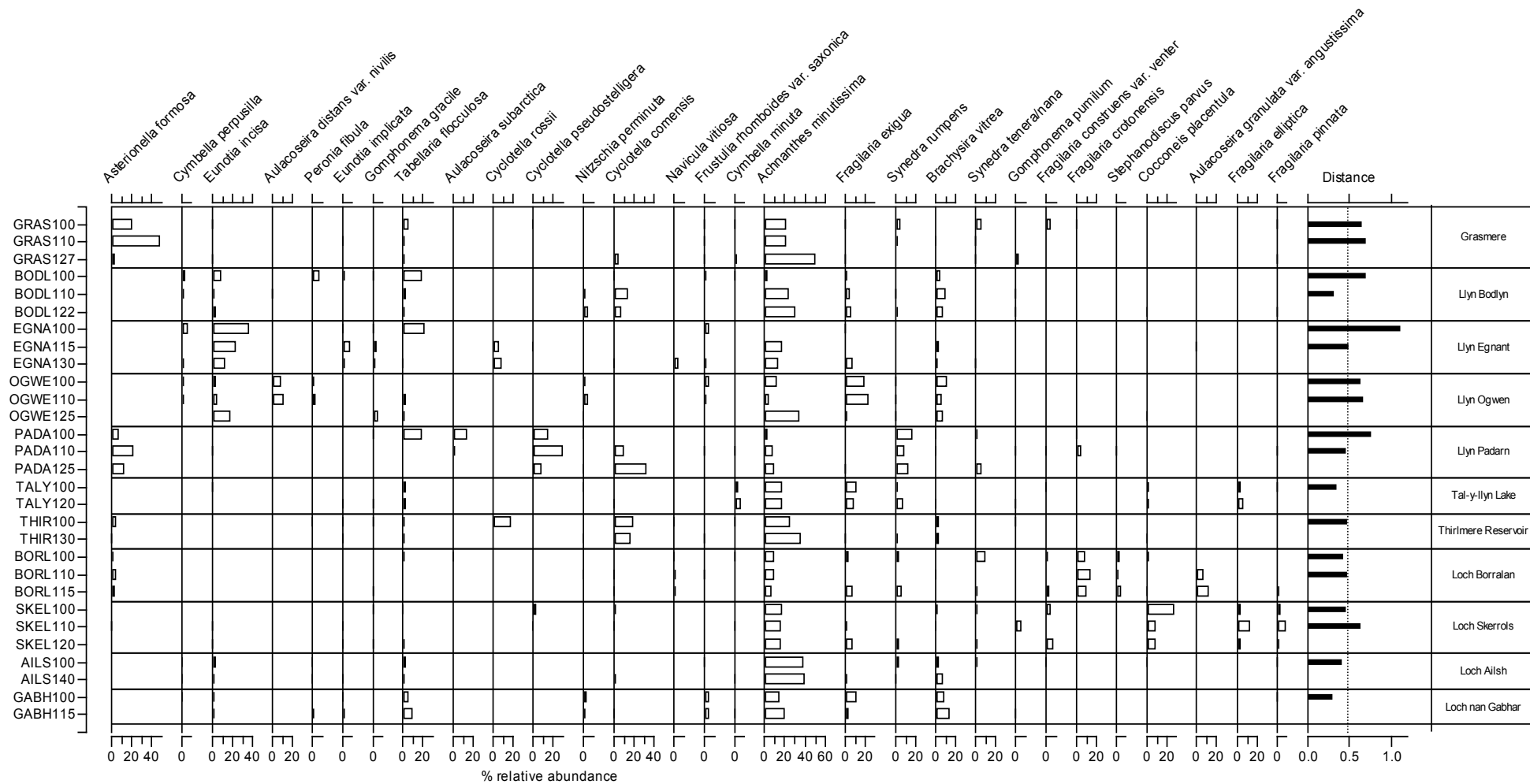
Given the concerns raised above, there appears to be only one potential reference lake within the HA group – Loch Kinnabus, where the diatom assemblage shows no significant change between top and bottom samples (squared chord distance = 0.418). The diatom assemblage of this site is diverse, dominated by *A. minutissima* throughout, with small *Fragilaria* spp. occurring alongside (Figure 3.4). These non-planktonic taxa are typical of moderate nutrient status and clear-water conditions. It is worth noting that the planktonic diatoms *C. radiosa*, *A. formosa*, *C. dubius* and *Stephanodiscus medius* occur at low relative abundance in the core top sample.

This species shift is indicative of mild eutrophication and may well provide an early warning that an ecologically important threshold has been crossed at Loch Kinnabus and that it is experiencing the early stages of nutrient enrichment. Nevertheless, this site provides the best example of a HA, D reference lake within this study, but monitoring and nutrient reduction measures should be put in place to ensure that this site remains in good condition.

**Figure 3.1 Summary diagram of diatom changes (% relative abundance) in cores from the peat (P), deep lake types. 'Distance' is the squared chord distance dissimilarity score between the core bottom and top (and mid) samples.**

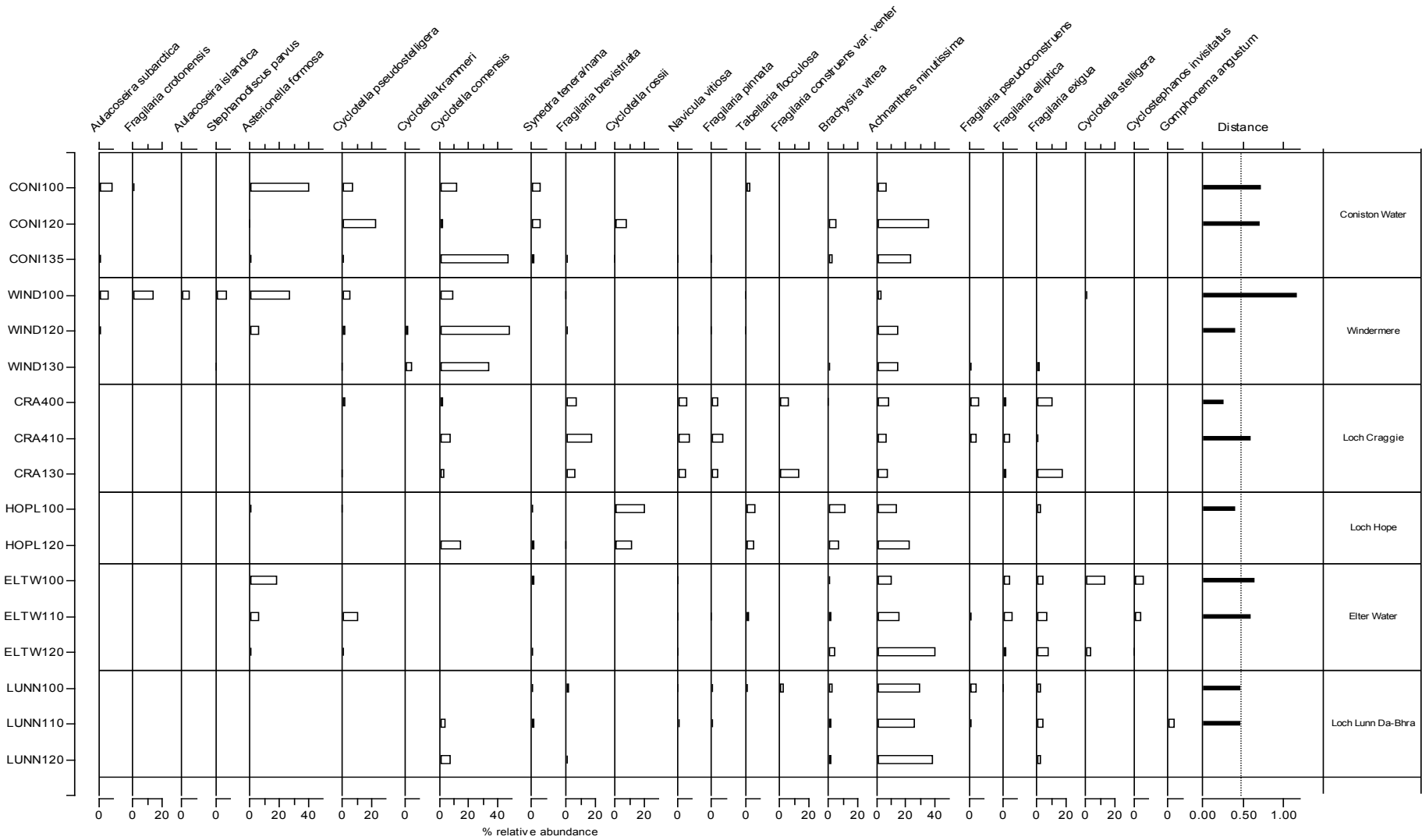


**Figure 3.2 Summary diagram of diatom changes (% relative abundance) in cores from the low alkalinity (LA), deep and shallow lake types. 'Distance' is the squared chord distance dissimilarity score between the core bottom and top (and mid) samples.**

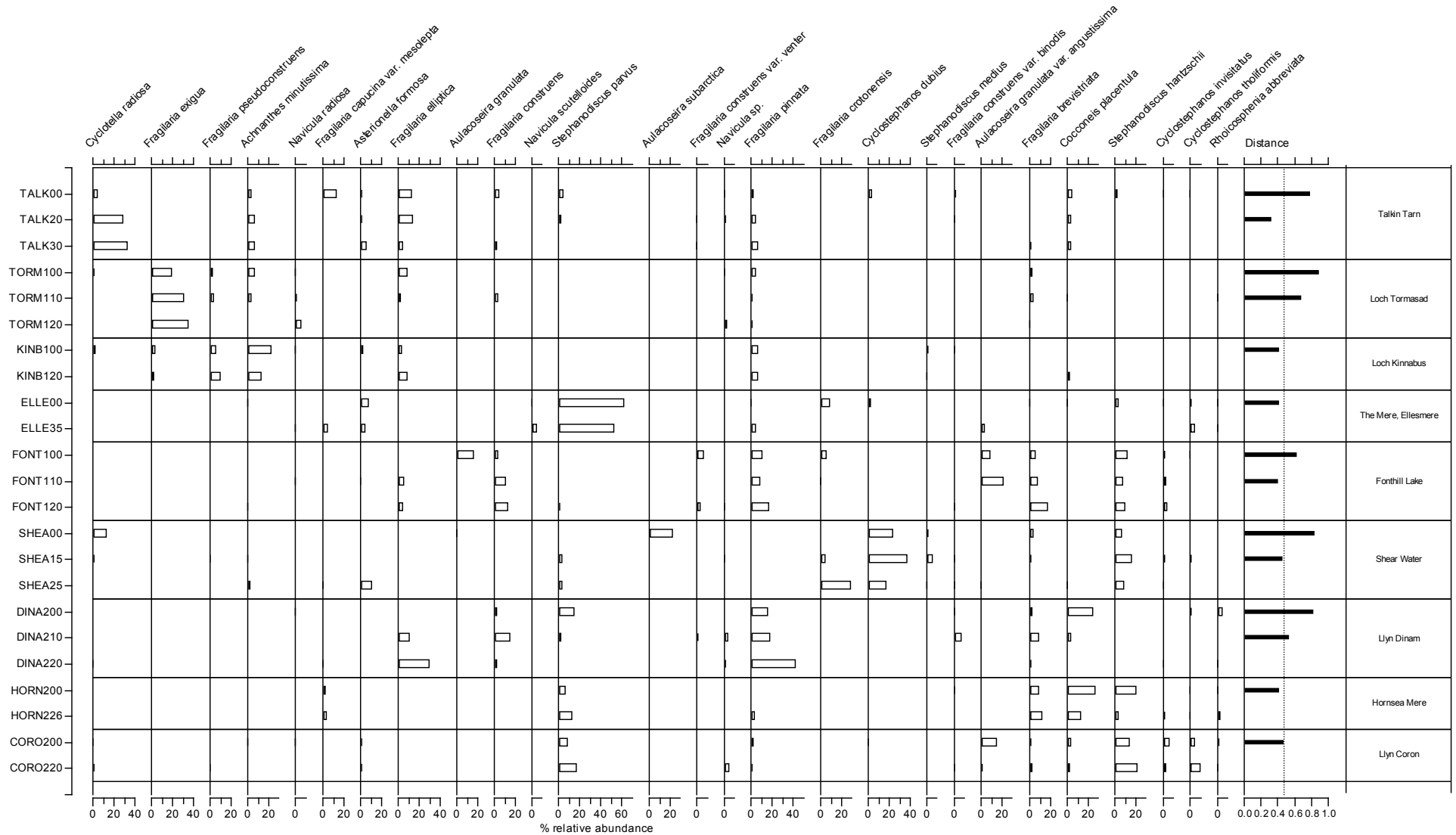




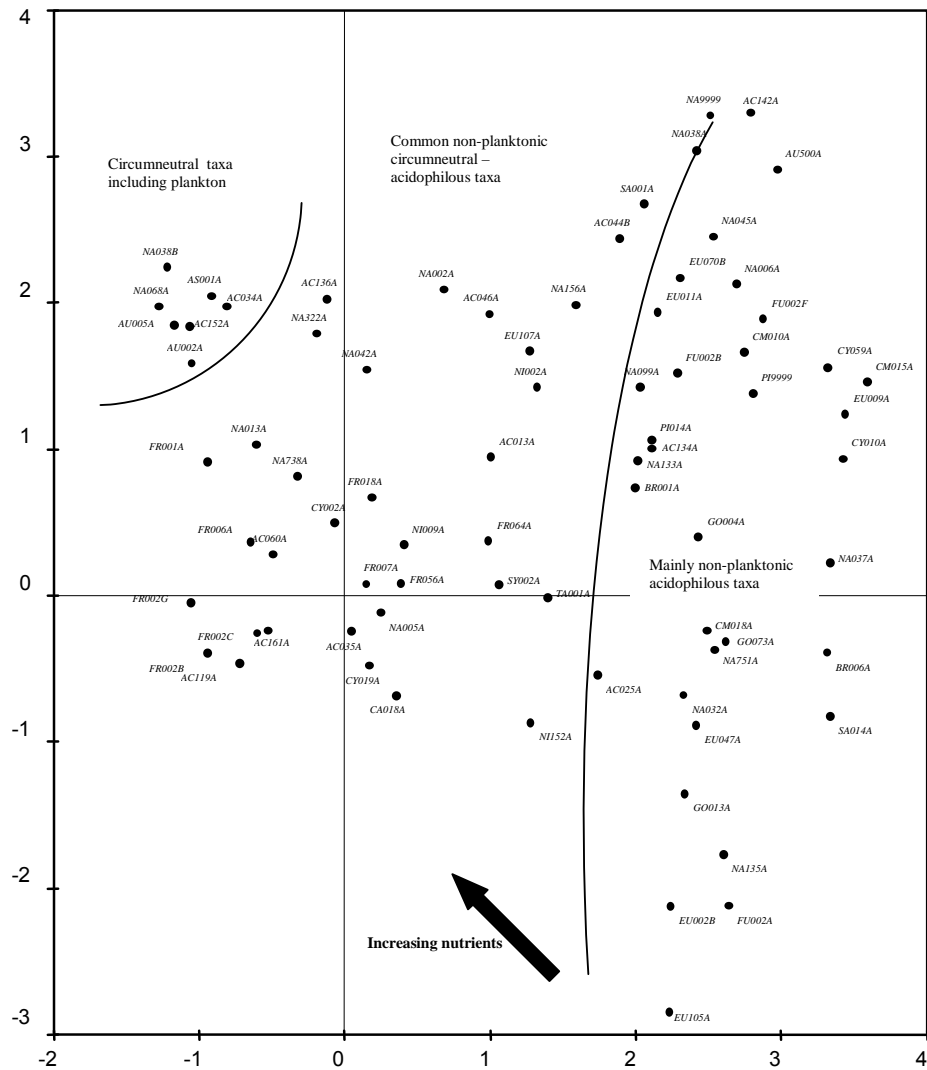
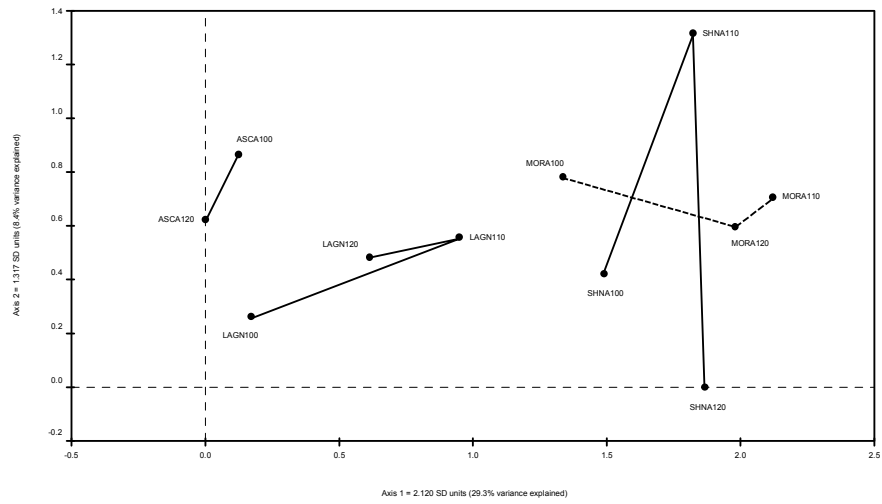
**Figure 3.3 Summary diagram of diatom changes (% relative abundance) in cores from the medium alkalinity (MA), deep and shallow lake types. 'Distance' is the squared chord distance dissimilarity score between the core bottom and top (and mid) samples.**



**Figure 3.4 Summary diagram of diatom changes (% relative abundance) in cores from the high alkalinity (HA), deep and shallow and marl (marl) lake types. 'Distance' is the squared chord distance dissimilarity score between the core bottom and top (and mid) samples.**

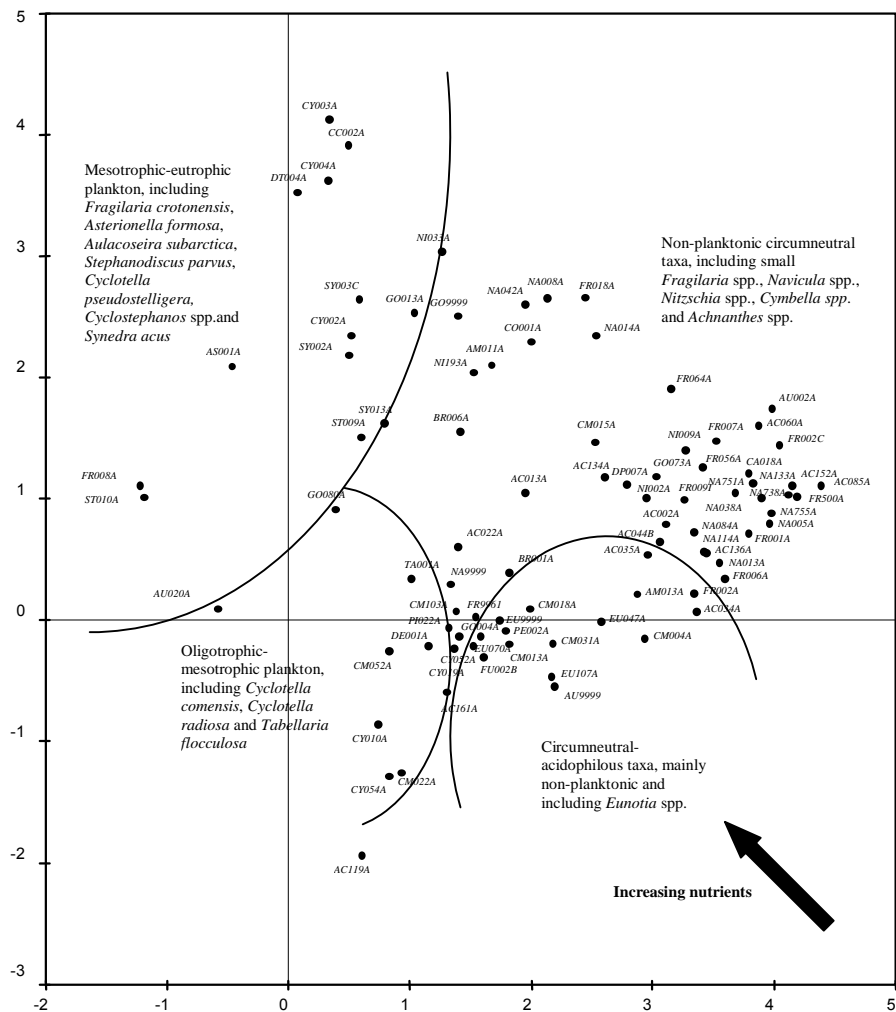
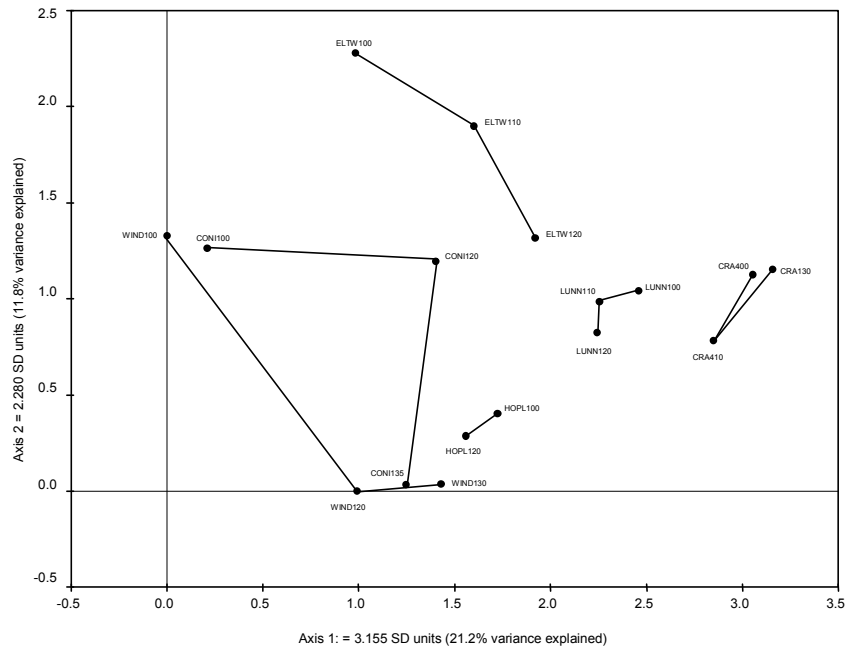


**Figure 3.5 DCA biplots (Axis 1 and 2) of the sample and species scores for peat lakes**

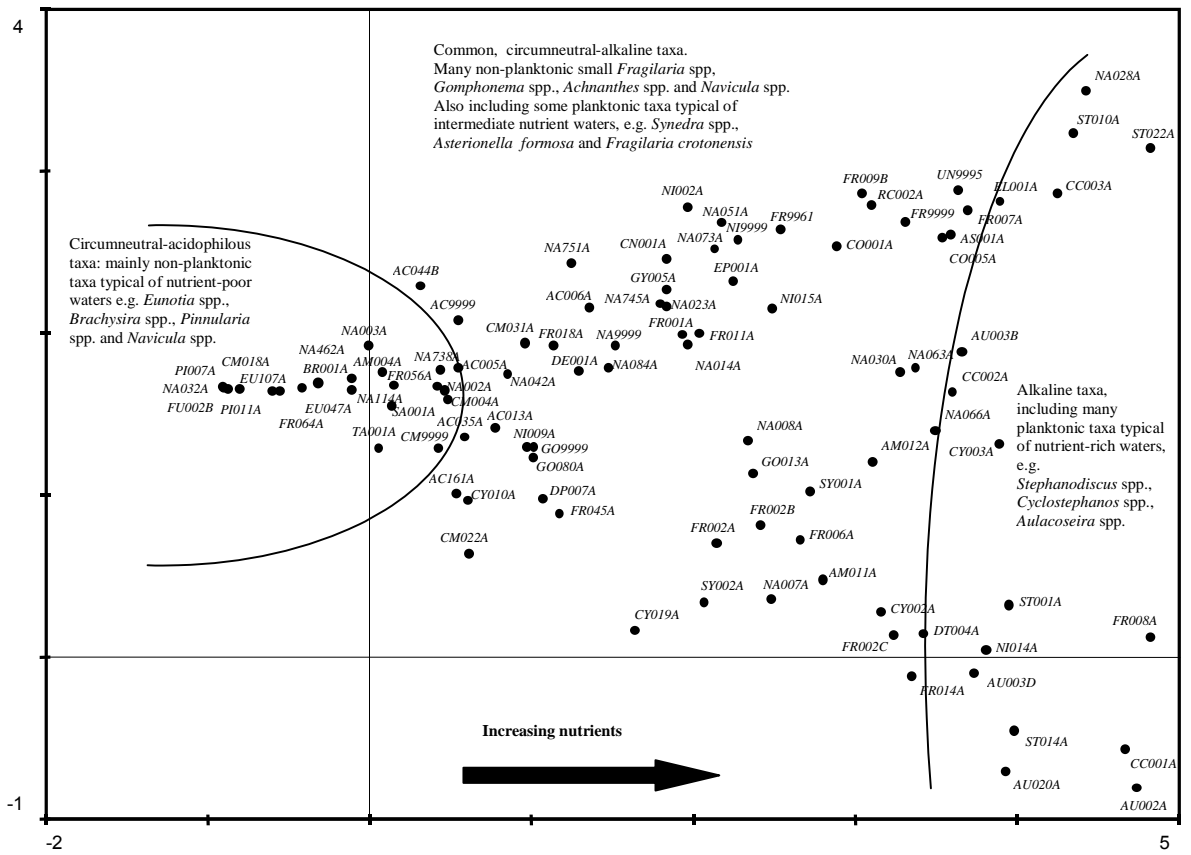
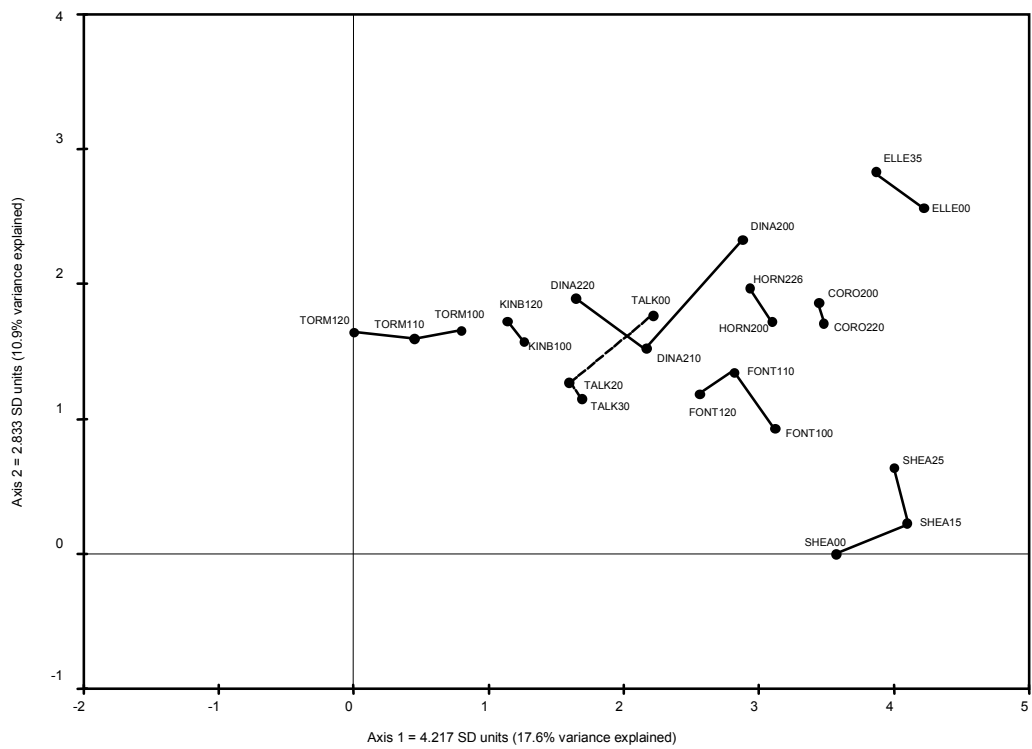




**Figure 3.7 DCA biplots (Axis 1 and 2) of the sample and species scores for MA lakes**



**Figure 3.8 DCA biplots (Axis 1 and 2) of sample and species scores for HA and marl lakes**



# 4 Summary of findings

Following screening for diatom preservation and the subsequent elimination of sites displaying diatom dissolution, core bottoms were analysed from 30 lakes (10 England, seven Wales and 13 Scotland). Mid-core samples were analysed from a sub-set of 20 of these lakes (seven England, five Wales and eight Scotland), resulting in the analysis of a total of 50 mid and bottom core samples. The degree of floristic change between the bottom and top/mid samples analysed in each core was estimated using the squared chord distance dissimilarity coefficient, and detrended correspondence analysis (DCA) was performed to assess the direction and magnitude of floristic change at each site. The results are summarised in Table 4.1 and an overview of the findings is discussed below.

**Table 4.1 Summary results from the diatom analysis of 30 cores**

Lake name	<sup>1</sup> GB lake type		<sup>1</sup> Significant floristic change?	<sup>2</sup> Trophic change?	Depth of reference sample (cm)	<sup>3</sup> Potential reference site?
Loch an Lagain	P	D	No (20-0) ... Yes (20-10)	No	20	Yes
Loch na Moracha	P	D	Yes	No	20	Possibly
Loch Shnathaid	P	D	Yes but relatively low score (~0.5)	↑ pH?	20	Possibly
Loch Ascaig	P	D	No	No	20	Yes
Grasmere	LA	D	Yes	↑	28	No
Llyn Bodlyn	LA	D	Yes (22-0) ... No (22-10)	↓ pH?	23	No
Llyn Egnant	LA	D	Yes	↓ pH?	31	No
Llyn Ogwen	LA	D	Yes	↓ pH?	25	No
Llyn Padarn	LA	D	Yes (25-0) ... No (25-10)	↑	25	No
Tal-y-llyn Lake	LA	D	No	No	21	Yes
Thirlmere Reservoir	LA	D	Yes but relatively low score (0.48)	?	31	Possibly
Loch Borralan	LA	Sh	No (15-0) ... Yes (15-10)	?	15	Possibly
Loch Skerrols	LA	Sh	No (20-0) ... Yes (20-10)	No	20	Yes
Loch Ailsh	LA	Sh	No	No	40	Yes
Loch nan Gabhar	LA	Sh	No	No	15	Yes
Coniston Water	MA	D	Yes	↑	25	No
Windermere	MA	D	Yes (30-0) ... No (30-20)	↑	30	No
Loch Craggie	MA	D	No (30-0) ... Yes (30-10)	No	30	Yes
Loch Hope	MA	D	No	No	20	Yes
Elter Water	MA	Sh	Yes	↑	21	No
Lochan Lùnn Dà-Bhrà	MA	Sh	No	No	20	Yes
Talkin Tarn	Marl	D	Yes (30-0) ... No (30-20)	↑	31	No
Loch Tormasad	HA	D	Yes	↑ pH?	20	Possibly
Loch Kinnabus	HA	D	No	No	20	Yes
The Mere, Ellesmere	HA	D	No	?	35	Possibly
Fonthill Lake	HA	Sh	Yes (20-0) .... No (20-10)	↑	21	No
Shear Water	HA	Sh	Yes (25-0)... No (25-15)	↑	26	Possibly
Llyn Dinam	HA	Sh	Yes	↑	20	No
Hornsea Mere	HA	Sh	No	?	27	No
Llyn Coron	HA	Sh	Yes but relatively low score (0.48)	?	20	Possibly

<sup>1</sup> Change was deemed significant where the squared chord distance dissimilarity scores between the core bottom and mid and/or top sample exceeded the critical value of 0.475. Numbers in parentheses indicate the depth (cm) of the two samples being compared.

<sup>2</sup> ↑ increase in trophic status; ↓ decrease in trophic status; ? uncertain. Assessment of trophic change is based on the diatom species shifts. Where floristic change is indicative of a shift in pH rather than trophic status, the symbol 'pH' is shown.

<sup>3</sup> For sites classed as 'Possibly', please see text.

In summary, 17 of the 30 sites appear to have experienced significant floristic change in diatom species assemblages and 13 sites displayed minimal floristic change. The majority of changes appear to relate to increases in trophic status, although at some sites, floristic changes suggest increasing acidity (Llyn Bodlyn, Llyn Egnant and Llyn Ogwen) or decreasing acidity (Lochs Tormasad and Shnathaid).

A total of 10 out of the 30 lakes are thought to be good examples of reference lakes, given the minimal change in their diatom species assemblages and low squared chord distance dissimilarity distances between core bottom and top samples. Tal-y-llyn Lake (LA, D) is the only non-Scottish example of a reference lake here. All other potential reference sites are Scottish lochs and examples for each lake type are present with the exception of marl lakes, for which there are no Scottish examples in this study. Lochs Lagain and Ascaig are examples of peat lake reference sites, Lochs Skerrols, Ailsh and nan Gabhar of LA, Sh reference lakes, Lochs Craggie and Hope of MA, D reference lakes, and Lochan Lùnn Dà-Bhrà of a MA, Sh reference lake; Loch Kinnabus is the only example of a HA, D reference lake. At some reference lakes such as Lochs Kinnabus, Hope, Craggie and Lochan Lùnn Dà-Bhrà, there may be early warning signs of slight increases in trophic status based on the appearance of nutrient-tolerant diatom taxa in the surface samples. We recommend that the water chemistry and ecology of these sites is monitored closely over the coming years, to ascertain whether a shift from reference conditions is occurring.

A further eight lakes showed relatively low floristic change throughout their cores and may therefore be potential reference lakes - Lochs na Moracha and Shnathaid (P, D), Thirlmere Reservoir (LA, D), Loch Borralan (LA, Sh), Loch Tormasad and Mere, Ellesmere (HA, D), Shear Water and Llyn Coron (HA, Sh). However, further investigation is required to confirm their status.

One limitation of this study is that none of the cores are dated and for some lakes where sediment cores are short, core bottom samples may not represent true 'reference' samples. This is of particular concern for the HA lakes since sediment accumulation rates can be rapid in these productive systems. The lack of a chronology is of less concern where the diatom assemblages remain stable throughout the core, but it becomes a greater limitation when interpreting the data from sites that exhibit floristic change, as we have no estimate of the time at which the changes occurred. Sediment accumulation rates are site specific and it is unlikely that all cores cover comparable time periods. Longer cores would need to be collected and radiometric or spheroidal carbonaceous particle (SCP) dating carried out to provide a more detailed assessment of the nature and timing of ecological changes at these sites over longer timescales. Analysis of remains of other biological elements preserved in lake sediment cores, such as plant macrofossils, cladocera and chironomids, would enable more holistic ecological reference conditions to be defined, and would provide valuable information on changes in ecological structure and function (Sayer et al., 1999; Bennion, 2001).

It is recommended that further palaeoecological work be carried out at those sites highlighted as potential reference lakes. In cases where sediment cores were short and core bottom samples are thought not to extend back far enough to represent reference conditions, it is suggested that longer sediment cores are taken. Diatom analysis of core bottom samples and dating using either radiometric or SCP methods to confirm suitability as reference samples is advised. In addition, sites such as Loch Borralan, where no change between top and bottom samples was seen but where the assemblages were comprised of many nutrient-tolerant diatom taxa, may represent naturally meso-eutrophic lakes and further analysis is recommended.



Nevertheless, this study shows that even low resolution analysis of sediment cores can produce valuable information for establishing reference conditions, selecting reference lakes, assessing ecological status and the extent of ecological change. Simple techniques such as dissimilarity scores and ordination analyses applied to palaeoecological data can be used to characterise lakes, and establish reference conditions and deviation from the reference state.

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# Acknowledgements

The authors would like to thank the following people for their contributions to this study:

- Numerous colleagues in the Environment Agency and Countryside Council for Wales for assistance in arranging site access.
- The Scottish Environment Protection Agency for collecting sediment cores from Scottish lochs.
- All landowners, estate managers and local wardens for their co-operation and for granting permission to carry out the work.
- Gavin Simpson at UCL for assistance with the squared chord distance dissimilarity data analysis.
- Ewan Shilland, James Shilland and Ben Goldsmith at UCL for technical support in the field.
- Liptrot, Sophie Theophile, Ellie Simon Turner and Liam Macrae at UCL for laboratory work.

**Appendix 1. List of the 203 common diatom taxa (more than one per cent in more than two samples) in the 30 cores with Diatcode and full name**

Diatcode	Name	Diatcode	Name
AC002A	Achnanthes linearis	FR008A	Fragilaria crotonensis
AC004A	Achnanthes pseudoswazii	FR009B	Fragilaria capucina var. mesolepta
AC005A	Achnanthes calcar	FR009I	Fragilaria capucina var. austriaca
AC006A	Achnanthes clevei	FR009L	Fragilaria capucina var. amphicephala
AC013A	Achnanthes minutissima	FR011A	Fragilaria lapponica
AC022A	Achnanthes marginulata	FR014A	Fragilaria leptostauron
AC023A	Achnanthes conspicua	FR018A	Fragilaria elliptica
AC025A	Achnanthes flexella	FR045A	Fragilaria parasitica
AC034A	Achnanthes suchlandtii	FR056A	Fragilaria pseudoconstruens
AC035A	Achnanthes pusilla	FR064A	Fragilaria exigua
AC037A	Achnanthes biasolettiana	FR500A	Fragilaria suboldenburgiana
AC044B	Achnanthes levanderi	FR9961	Fragilaria vaucheriae (fine)
AC046A	Achnanthes altaica	FR9999	Fragilaria sp.
AC060A	Achnanthes curtissima	FU002A	Frustulia rhomboides
AC083A	Achnanthes laevis	FU002B	Frustulia rhomboides var. saxonica
AC085A	Achnanthes lauenbergiana	FU002F	Frustulia rhomboides var. viridula
AC105A	Achnanthes petersenii	GO004A	Gomphonema gracile
AC119A	Achnanthes saccula	GO006A	Gomphonema acuminatum
AC134A	Achnanthes helvetica	GO013A	Gomphonema parvulum
AC136A	Achnanthes subatomoides	GO073A	Gomphonema angustum
AC142A	Achnanthes kuelbsii	GO080A	Gomphonema pumilum
AC143A	Achnanthes oblongella	GO9999	Gomphonema sp.
AC152A	Achnanthes carrisima	GY005A	Gyrosigma acuminatum
AC161A	Achnanthes ventralis	HN001A	Hannaea arcus
AC182A	Achnanthes rosenstockii	MR001A	Meridion circulare
AC9999	Achnanthes sp.	NA002A	Navicula jaernefeltii
AM004A	Amphora veneta	NA003A	Navicula radiosa
AM010A	Amphora fogediana	NA005A	Navicula seminulum
AM011A	Amphora libyca	NA006A	Navicula mediocris
AM012A	Amphora pediculus	NA007A	Navicula cryptopcephala
AM013A	Amphora inariensis	NA008A	Navicula rhynoccephala
AS001A	Asterionella formosa	NA013A	Navicula pseudoscutiformis
AS003A	Asterionella ralfsii	NA014A	Navicula pupula
AU002A	Aulacoseira ambigua	NA023A	Navicula gregaria
AU003B	Aulacoseira granulata var. angustissima	NA028A	Navicula scutelloides
AU003D	Aulacoseira granulata	NA030A	Navicula menisculus
AU005A	Aulacoseira distans	NA032A	Navicula cocconeiformis
AU005E	Aulacoseira distans var. nivilis	NA033A	Navicula subtilissima
AU020A	Aulacoseira subarctica	NA037A	Navicula angusta
AU031A	Aulacoseira alpigena	NA038A	Navicula arvensis
AU500A	Aulacoseira crassipuncta	NA038B	Navicula arvensis var. major
AU9999	Aulacoseira sp.	NA042A	Navicula minima
BR001A	Brachysira vitrea	NA045A	Navicula bryophila
BR006A	Brachysira brebbisonnii	NA051A	Navicula cari
CA003A	Caloneis silicula	NA063A	Navicula trivialis
CA018A	Caloneis tenuis	NA066A	Navicula capitata
CC001A	Cyclostephanos dubius	NA068A	Navicula impexa
CC002A	Cyclostephanos invisitatus	NA073A	Navicula placentula
CC003A	Cyclostephanos tholiformis	NA084A	Navicula atomus
CM004A	Cymbella microcephala	NA099A	Navicula bremensis
CM010A	Cymbella perpusilla	NA112D	Navicula minuscula var muralis
CM013A	Cymbella helvetica	NA114A	Navicula subrotundata
CM015A	Cymbella cesattii	NA128A	Navicula schoenfeldtii
CM017A	Cymbella hebridica	NA133A	Navicula schassmannii
CM018A	Cymbella gracilis	NA135A	Navicula tenuicephala
CM020A	Cymbella gaeumannii	NA156A	Navicula leptostriata
CM022A	Cymbella affinis	NA190A	Navicula agrestis
CM031A	Cymbella minuta	NA322A	Navicula detenta
CM035A	Cymbella angustata	NA462A	Navicula joubardii
CM052A	Cymbella descripta	NA738A	Navicula vitiosa
CM085A	Cymbella lapponica	NA745A	Navicula capitoradiata
CM103A	Cymbella silesiaca	NA751A	Navicula cryptotenella
CM9999	Cymbella sp.	NA755A	Navicula kuelbsii
CN001A	Cymbellonitzschia diluviana	NA9999	Navicula sp.
CO001A	Cocconeis placentula	NE004A	Nedium bisulcatum
CO005A	Cocconeis pediculus	NI002A	Nitzschia fonticola
CY002A	Cyclotella pseudostelligera	NI009A	Nitzschia palea
CY003A	Cyclotella meneghiniana	NI014A	Nitzschia amphibia
CY004A	Cyclotella stelligera	NI015A	Nitzschia dissipata

CY010A	<i>Cyclotella comensis</i>	NI017A	<i>Nitzschia gracilis</i>
CY019A	<i>Cyclotella radiosa</i>	NI033A	<i>Nitzschia paleacea</i>
CY052A	<i>Cyclotella rossii</i>	NI152A	<i>Nitzschia pusilla</i>
CY054A	<i>Cyclotella krammeri</i>	NI193A	<i>Nitzschia perminuta</i>
CY059A	<i>Cyclotella cyclopuncta</i>	NI9999	<i>Nitzschia</i> sp.
DE001A	<i>Denticula tenuis</i>	PE002A	<i>Peronia fibula</i>
DP007A	<i>Diploneis oblongella</i>	PI007A	<i>Pinnularia viridis</i>
DT004A	<i>Diatoma tenuis</i>	PI011A	<i>Pinnularia microstauron</i>
DT021A	<i>Diatoma mesodon</i>	PI014A	<i>Pinnularia appendiculata</i>
EL001A	<i>Ellerbeckia arenaria</i>	PI022A	<i>Pinnularia subcapitata</i>
EP001A	<i>Epithemia sorex</i>	PI9999	<i>Pinnularia</i> sp.
EU002B	<i>Eunotia pectinalis</i> var. <i>minor</i>	RC002A	<i>Rhoicosphenia abbreviata</i>
EU002D	<i>Eunotia pectinalis</i> var. <i>undulata</i>	RE001A	<i>Reimeria sinuata</i>
EU009A	<i>Eunotia exigua</i>	RH003E	<i>Rhopalodia rupestris</i>
EU011A	<i>Eunotia rhomboidea</i>	SA001A	<i>Stauroneis anceps</i>
EU013A	<i>Eunotia arcus</i>	SA014A	<i>Stauroneis gracilis</i>
EU025A	<i>Eunotia fallax</i>	SP006A	<i>Stenopterobia curvula</i>
EU040A	<i>Eunotia paludosa</i>	ST001A	<i>Stephanodiscus hantzschii</i>
EU047A	<i>Eunotia incisa</i>	ST009A	<i>Stephanodiscus alpinus</i>
EU070A	<i>Eunotia bilunaris</i>	ST010A	<i>Stephanodiscus parvus</i>
EU070B	<i>Eunotia bilunaris</i> var. <i>mucophila</i>	ST014A	<i>Stephanodiscus medius</i>
EU105A	<i>Eunotia subarcuatoides</i>	ST022A	<i>Stephanodiscus neoastraea</i>
EU107A	<i>Eunotia implicata</i>	ST9999	<i>Stephanodiscus</i> sp.
EU108A	<i>Eunotia intermedia</i>	SU001A	<i>Suriella angusta</i>
EU110A	<i>Eunotia minor</i>	SY001A	<i>Synedra ulna</i>
EU9999	<i>Eunotia</i> sp.	SY002A	<i>Synedra rumpens</i>
FR001A	<i>Fragilaria pinnata</i>	SY003C	<i>Synedra acus</i> var. <i>angustissima</i>
FR002A	<i>Fragilaria construens</i>	SY003F	<i>Synedra delicatissima</i>
FR002B	<i>Fragilaria construens</i> var. <i>binodis</i>	SY013A	<i>Synedra tenera/nana</i>
FR002C	<i>Fragilaria construens</i> var. <i>venter</i>	TA001A	<i>Tabellaria flocculosa</i>
FR002G	<i>Fragilaria construens</i> var. <i>pumilla</i>	UN9995	Unknown centric
FR006A	<i>Fragilaria brevistriata</i>	YH001A	<i>Ctenophora pulchella</i>
FR007A	<i>Fragilaria vaucheriae</i>		

**Appendix 2 Squared chord distance dissimilarity scores for the mid and surface samples in the 30 cores**

Lake name	<sup>1</sup> Sample code	<sup>2</sup> Squared chord distance dissimilarity score
Hornsea Mere	HORN200	0.420
	HORN226	
The Mere, Ellesmere	ELLE00	0.419
	ELLE35	
Thirlmere	THIR100	0.482
	THIR130	
Tal-y-llyn Lake	TALY100	0.359
	TALY120	
Llyn Coron	CORO200	0.479
	CORO220	
Loch Ailsh	AILS100	0.415
	AILS140	
Loch Hope	HOPL100	0.403
	HOPL120	
Loch nan Gabhar	GABH100	0.307
	GABH115	
Loch Kinnabus	KINB100	0.418
	KINB120	
Loch Ascaig	ASCA100	0.318
	ASCA120	
Grasmere	GRAS100	0.650
	GRAS110	0.698
	GRAS127	
Llyn Egnant	EGNA100	1.113
	EGNA115	0.503
	EGNA130	
Llyn Dinam	DINA200	0.830
	DINA210	0.544
	DINA220	
Coniston Water	CONI100	0.724
	CONI120	0.712
	CONI135	
Elterwater	ELTW100	0.649
	ELTW110	0.600
	ELTW120	
Shearwater	SHEA00	0.848
	SHEA15	0.466
	SHEA25	
Fonthill Lake	FONT100	0.636
	FONT110	0.404
	FONT120	
Talkin Tarn	TALK00	0.784
	TALK20	0.330
	TALK30	
Windermere	WIND100	1.170
	WIND120	0.408
	WIND130	
Llyn Bodlyn	BODL100	0.707
	BODL110	0.316
	BODL122	
Llyn Padarn	PADA100	0.761
	PADA110	0.465
	PADA125	
Llyn Ogwen	OGWE100	0.641



Lake name	<sup>1</sup> Sample code	<sup>2</sup> Squared chord distance dissimilarity score
	OGWE110 OGWE125	0.679
Loch Skerrols	SKEL100 SKEL110 SKEL120	0.465 0.647
Loch Craggie	CRA400 CRA410 CRA130	0.269 0.597
Loch an Lagain	LAGN100 LAGN110 LAGN120	0.418 0.559
Loch Shnathaid	SHNA100 SHNA110 SHNA120	0.516 0.555
Loch na Moracha	MORA100 MORA110 MORA120	0.583 0.528
Loch Borralan	BORL100 BORL110 BORL115	0.434 0.480
Lochan Lùnn Dà-Bhrà	LUNN100 LUNN110 LUNN120	0.474 0.466
Loch Tormasad	TORM100 TORM110 TORM120	0.891 0.686

<sup>1</sup> Last two digits of sample code indicate sample depth (cm).

<sup>2</sup> Squared chord distance dissimilarity score between the core bottom sample and each other sample in that core.

We are The Environment Agency. It's our job to look after your environment and make it **a better place** – for you, and for future generations.

Your environment is the air you breathe, the water you drink and the ground you walk on. Working with business, Government and society as a whole, we are making your environment cleaner and healthier.

The Environment Agency. Out there, making your environment a better place.

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