UKTAG Lake Assessment Method

Macrophytes and Phytobenthos

Phytobenthos – Diatoms for Assessing River and Lake Ecological Quality (Lake DARLEQ2)

by

Water Framework Directive – United Kingdom Technical Advisory Group (WFD-UKTAG)

Health and safety statement

WARNING. Working in or around water is inherently dangerous; persons using this standard should be familiar with normal laboratory and field practice. This published monitoring system does not purport to address all of the safety problems, if any, associated with its use. It is the responsibility of the user to establish appropriate health and safety practices and to ensure compliance with any national regulatory guidelines.

It is also the responsibility of the user if seeking to practise the method outlined here, to gain appropriate permissions for access to water courses and their biological sampling.

UKTAG Guide to Phytobenthos in Lakes
Diatoms for Assessing River and Lake Ecological Quality
(DARLEQ2)

1   Introduction

This classification method enables the assessment of phytobenthos in lakes according to the requirements of the Water Framework Directive (WFD).   Phytobenthos refers to a mostly microscopic group of organisms called algae found attached to submerged surfaces such as stones and plant stems. Assessment focuses on the diatoms, a large and diverse group of algae using a tool called "DARLEQ2" (**D**iatoms for **A**ssessing **R**iver and **L**ake **E**cological **Q**uality, based on a metric called the Trophic Diatom Index (TDI).

DARLEQ forms one part of the quality element "macrophytes and phytobenthos". Macrophytes are assessed separately with a method called LEAFPACS2. An earlier version of DARLEQ was evaluated and revised to ensure that in combination with the revised macrophyte tool this tool provides an appropriate assessment to the overall quality element of macrophytes and phytobenthos in lakes, and the revised method is known as DARLEQ2.

DARLEQ2 and LEAFPACS2 results are combined to produce an overall classification for macrophytes and phytobenthos in lakes, using the worst class of either sub-element.

1.1   Metrics
The metric used to classify phytobenthos in lakes is called Lake TDI (LTDI). It is based on the expert-derived riverine trophic diatom index which was re-calibrated and applied to lake diatom communities. The most recent index is LTDI2. Diatom taxa are each assigned a score from 1 (nutrient sensitive) to 5 (nutrient tolerant) and the computed LTDI2 scores range from 0 (very low nutrients) to 100 (very high nutrients). The WFD requires derivation of ecological status as an EQR (Ecological Quality Ratio). The LTDI2 EQR is calculated based on observed data and predicted reference values, resulting in an overall EQR representing an ecological status class of either High, Good, Moderate, Poor or Bad. The EQR scale ranges 0 (bad ecological status) to 1 (high ecological status).

1.2   Environmental pressures to which the method is sensitive
The method is used to detect the impact of nutrient enrichment, primarily indicating response to inorganic nutrient pressure, phosphorus and nitrogen. However, other factors such as grazing by invertebrates and hydromorphological regimes can influence diatom abundance and composition. These may influence the overall classification result but are not built into the classification method.

1.3   Geographic application
This assessment method is appropriate for all inland freshwaters in the UK (e.g. lakes, lochs and loughs), but does not cover peat and brackish systems.

1.4   Intercalibration
This is a process whereby all European Member States were required to compare WFD class boundary values for each biological quality element (e.g. phytobenthos, phytoplankton) to ensure similar levels are set across all countries. Once a classification method has been

intercalibrated, the method must be adhered to by Member States for the purposes of WFD assessment and reporting. Intercalibration mainly focussed on the EQRs which define the class boundaries between High and Good, and Good and Moderate. The current DARLEQ2 EQR boundaries for lakes are defined for three alkalinity types:

Alkalinity type (annual mean CaCO$_3$ mg/l)	Boundary			
	H/G	G/M	M/P	P/B
Low Alkalinity (<10)	0.92	0.70	0.46	0.23
Medium Alkalinity (10-50)	0.93	0.66	0.46	0.23
High Alkalinity (>50)	0.92	0.70	0.46	0.23

2   Data collection

2.1   Sample collection – location, frequency and sample volume

Samples of the biofilm which covers the upper surfaces of submerged cobbles or small boulders in lakes are collected by brushing or scraping with a clean toothbrush. Cobbles and small boulders are the preferred sampling substrate but if there are no cobbles or small boulders present at the sampling site, the submerged stems of emergent macrophytes, such as *Phragmites australis*, *Sparganium erectum*, *Glyceria maxima* or *Typha* species, or leaves and stems of submerged macrophytes such as *Ranunculus* species and *Potamogeton* species are sampled instead. The sampling method is fully detailed and conforms to European Standards (CEN, 2014). Samples should be collected from the littoral zones of lakes using a clean toothbrush, ensuring substrata have been submerged for at least 4 weeks prior to sampling.

2.1.1   Location

Samples must be representative of conditions in the lake being studied, and should be collected from littoral zone habitats with similar physical conditions at all sites, especially when collecting samples on repeat visits. Samples sites should be away from obvious human impacts and inflowing streams.

2.1.2   Timing and Frequency

Two samples per year should be collected, one during spring (between March and May) and one during autumn (September to November). If this is not possible, summer sampling (June to August) is an alternative option as analysis of results has demonstrated that seasonal effects are not significant. Samples should be collected not less than two months apart.

2.1.3   Sample volume

Vigorous brushing of the substrate with a clean toothbrush in a plastic tray removes the diatom film from surface of the substrate. In situations where submerged macrophytes have been sampled, cut random lengths are put into sampling bottles or bags and vigorously agitated to dislodge the attached diatoms. This process results in a nominal volume of sample collected that is easily transferred to a plastic container (volume 50-100mls). There is not a precise volume of sample required, but larger volumes should be avoided.

2.2   Sample analysis

If samples cannot be analysed soon after collection they should be preserved as soon as possible with Lugol's iodine (adding approximately 5-10% by volume). Samples are then digested to remove all internal contents of the diatom cell (the frustule), leaving clean 'valves' (digestion of the frustule generally results in separation of two valves). Permanent slides are prepared using Naphrax as a diatom mountant, and at least 300 undamaged valves of non-planktic taxa should be identified and counted using a high power microscope (x1000 magnification). The presence and number of valves is recorded of each diatom taxa present. The analytical method is fully detailed and conforms to European Standards (CEN, 2014).

2.3   Other data requirements

Alkalinity data is required as a predictor of reference (expected) conditions for the DARLEQ2 EQR. Data should be obtained from analysis of samples from the lake taken at monthly intervals over a period of a year, reported as mg/L $CaCO_3$.

It is recommended that the same alkalinity value is used as a predictor variable for both macrophyte and phytobenthos classifications.

2.4   Minimum data requirements

Confidence of classification will depend on the number of samples taken, and the proximity of the resulting EQR to a class boundary. Ideally a total of 6 samples (over 3 years) are recommended. In practice fewer are used to produce a classification, although with a reduced confidence of class.

2.5   Typology

Typology defines waterbodies by factors which have a strong influence on their ecology. In lake phytobenthos EQR calculations, lake types are defined by alkalinity - low alkalinity, <10 mg/L $CaCO_3$; moderate alkalinity, 10-50 mg/L $CaCO_3$; and high alkalinity, >50mg/L $CaCO_3$.

3 Procedures for calculating metric EQRs

The phytobenthos EQR is a ratio of observed to expected values. *Observed values* are taken from samples collected in the field and *expected values* (reference values) are predicted from alkalinity.

3.1 Calculating EQR for DARLEQ

3.1.1 Status class boundaries

The status class boundaries were derived during the Intercalibration process for three lake alkalinity types to give final values as:

Alkalinity type (annual mean CaCO$_3$ mg/l)	Boundary			
	H/G	G/M	M/P	P/B
Low Alkalinity (<10)	0.92	0.70	0.46	0.23
Medium Alkalinity (10-50)	0.93	0.66	0.46	0.23
High Alkalinity (>50)	0.92	0.70	0.46	0.23

3.1.2 Manual calculation of EQR and status class

Calculation of lake phytobenthos EQRs can be carried out manually as detailed below.

All diatom taxa are assigned a score from 1 (nutrient sensitive) to 5 (nutrient tolerant). The list of diatom taxa and their nutrient sensitivity scores is detailed in Appendix A.

The LTDI2 for each sample is calculated using equations 1 and 2:

$$\text{Observed value of lake trophic diatom index} = (W \times 25) - 25$$

Equation 1

where:

"W" is given by the equation:

$$W = \frac{\sum_{j=1}^{n} a_j \times s_j}{\sum_{j=1}^{n} a_j}$$

Equation 2

where:
"a_j" is the number of valves of taxon j, and
"s_j" is the nutrient sensitivity score in Appendix A corresponding to the taxon represented by j.

The expected (reference) value of the LTDI2 (eLTDI2) is predicted from a regression equation derived from a subset of reference sites characterized by very low levels of human pressure. Reference sites across the UK were selected from a database using a range of criteria including evidence of degree of change based on palaeolimnological data, and were used to develop a type-specific reference typology. The model uses alkalinity, an environmental variable reflecting background geology and fertility. The value for the expected LTDI2 at reference conditions applicable to the site was derived from the site typology as detailed below.

	Alkalinity $CaCO_3$ mg/l (annual mean)	Expected (Reference) value of LTDI2
Low Alkalinity (LA)	<10	22
Medium Alkalinity (MA)	10-50	35
High Alkalinity (HA)	>50	42

Marl lakes are included with HA types, but peat and brackish systems are not covered under the tool.

The ecological quality ratio for the parameter should be calculated using the following equation:

$$EQR = \left(100 - \text{observed value of Lake TDI2}\right) \div \left(100 - \text{reference value for Lake TDI2}\right)$$

<div align="right">Equation 3</div>

Calculated EQR is set to '1.0' in instances where calculates EQR is > 1.

3.1.3   Worked example

The following details a manual worked example from an analysis of a real sample collected from a site in a lake in northern England.

Taxon name	Abundance (a = number of valves)	Nutrient sensitivity score (s)	a x s
Achnanthidium minutissimum type	187	2	374
Brachysira styriaca	2	1	2
Brachysira vitrea	48	1	48
Caloneis silicula	1	3	3
Cyclotella sp.	3	0	0
Cymbella affinis	7	2	14
Cymbella cistula	1	3	3
Delicata delicatula	1	1	1
Denticula tenuis	13	2	26

Encyonema gracile	1	2	2
Encyonemopsis microcephala	7	2	14
Eucocconeis flexella	3	3	9
Eunotia pectinalis	5	1	5
Eunotia sp.	3	1	3
Fragilaria perminuta	8	3	24
Gomphonema acuminatum	1	3	3
Gomphonema gracile	4	3	12
Gomphonema parvulum	1	5	5
Gomphonema sp	1	3	3
Nitzschia frustulum	1	4	4
Nitzschia gracilis	1	3	3
Nitzschia sp.	2	4	8
Rossithidium lineare	1	3	3
Sellaphora pupula	1	4	4
Synedra tenera	3	3	9
Tabellaria flocculosa	2	1	2
Tryblionella acuminata	4	4	16
Sum of *a* =	312	Sum of *as* =	600

The observed value of lake trophic diatom index for this sample is calculated as follows:

sum *as* for all taxa in sample = 600

sum *a* for all taxa in sample = 312

Calculate W = sum *as* / sum *a* = 1.92

Calculate the observed value of LTDI2 for the sample using the equations 1 and 2:

$(W \times 25) - 25 = (1.92 \times 25) - 25 = 23.1$

The mean total alkalinity for the lake was 3.1 mg/l $CaCO_3$ = LA type (low alkalinity). The reference value for LTDI2 is therefore 22.

Using the equation 3, the ecological quality ratio for the sample is:

$(100 - 23.1) / (100 - 22) = 76.9 / 78 = 0.99$

An EQR of 0.99 gives a face-value status class of **High**.

An average of two EQRs obtained over a year provides an annual EQR.

3.1.4 Automated calculation of EQR and status class

An MS Excel spreadsheet has been produced with a series of worksheets for data input and calculation of all components of the classification, including Confidence of Class. Instructions for use are provided within the spreadsheet calculator. The most recent version of the spreadsheet calculator can be found on the UKTAG website. Automated data entry and calculation of diatom EQR using the DARLEQ2 calculator spreadsheet is recommended for large data sets.

NB As revisions are likely to be made and the calculator updated over time, it is important to check that the most recent version is being used.

4 Procedures for calculating statistical confidence in metric

The assessment of statistical error associated with each EQR entry is calculated within DARLEQ2 and is expressed as a "confidence of class", i.e. the statistical confidence we have of the metric falling into each of the five classes, from High to Bad. This also makes it possible to determine the statistical confidence of the river classifying as "worse than Good status".

5 References

CEN (2014). *Water quality - Guidance standard for the routine sampling and preparation of benthic diatoms from rivers and lakes.* EN 13946: 2014. Comité European de Normalisation, Geneva.

CEN (2014). Water quality – Guidance for the identification and enumeration of benthic diatom samples from rivers and lakes. EN 14407: 2014. Comité European de Normalisation, Geneva.

Appendix A

The following table lists the lake diatom taxa and their sensitivity scores (LTDI2) for use in DARLEQ2.

TaxonName	LTDI2	TaxonName	LTDI2
Achnanthes abundans	2	Achnanthes lanceolata subsp. frequentissima	5
Achnanthes altaica	2	Achnanthes lanceolata var. bimaculata	5
Achnanthes austriaca var. helvetica	3	Achnanthes lanceolata var. dubia	4
Achnanthes bahusiensis	4	Achnanthes lanceolata var. elliptica	4
Achnanthes biasolettiana	4	Achnanthes lanceolata var. elliptico-lanceolata	5
Achnanthes biasolettiana var. subatomus	3	Achnanthes lanceolata var. rostrata	5
Achnanthes bioretii	3	Achnanthes lapponica	3
Achnanthes calcar	4	Achnanthes laterostrata	4
Achnanthes carissima	3	Achnanthes lauenbergiana	5
Achnanthes chlidanos	3	Achnanthes levanderi	3
Achnanthes clevei	4	Achnanthes linearis	3
Achnanthes coarctata	3	Achnanthes linearis fo. curta	2
Achnanthes conspicua	5	Achnanthes lutheri	4
Achnanthes curtissima	3	Achnanthes marginulata	2
Achnanthes daonensis	2	Achnanthes microcephala	1
Achnanthes daui	3	Achnanthes minuscula	4
Achnanthes delicatula	5	Achnanthes minutissima	2
Achnanthes delicatula ssp. hauckiana	5	Achnanthes minutissima var. jackii	2
Achnanthes delicatula subsp. delicatula	5	Achnanthes nodosa	3
Achnanthes depressa	1	Achnanthes oblongella	2
Achnanthes detha	3	Achnanthes oestrupii	3
Achnanthes didyma fo. didyma	4	Achnanthes peragalli	4
Achnanthes elliptica	4	Achnanthes petersenii	3
Achnanthes exigua	4	Achnanthes ploenensis	5
Achnanthes exilis	2	Achnanthes ploenensis var. gessneri	5
Achnanthes flexella	3	Achnanthes pseudoswazi	3
Achnanthes flexella var. alpestris	3	Achnanthes pusilla	3
Achnanthes frigida	3	Achnanthes ricula	5
Achnanthes grana	4	Achnanthes rosenstockii	3
Achnanthes helvetica	3	Achnanthes rossii	3
Achnanthes helvetica var. alpina	3	Achnanthes rostrata	5
Achnanthes holsatica	3	Achnanthes saccula	3
Achnanthes hungarica	3	Achnanthes saxonica	2
Achnanthes ingratiformis	4	Achnanthes scotica	2
Achnanthes joursacense	4	Achnanthes semiaperta	3
Achnanthes kriegeri	3	Achnanthes silvahercynia	3
Achnanthes kryophila	4	Achnanthes sp.	3
Achnanthes kuelbsii	3	Achnanthes stolida	3
Achnanthes laevis	3	Achnanthes straubiana	3
Achnanthes lanceolata	5	Achnanthes subatomoides	3

TaxonName	LTDI2
Achnanthes subsalsa	3
Achnanthes suchlandtii	3
Achnanthes ventralis	3
Achnanthes ziegleri	4
Achnanthidium affine	2
Achnanthidium biasolettiana var. subatomus	3
Achnanthidium biasolettianum	2
Achnanthidium caledonicum	1
Achnanthidium eutrophilum	2
Achnanthidium exilis	2
Achnanthidium microcephalum	1
Achnanthidium minutissima var. gracillima	1
Achnanthidium minutissima var. saprophila	2
Achnanthidium minutissimum	2
Achnanthidium minutissimum type	2
Achnanthidium pyrenaicum	4
Achnanthidium saprophilum	2
Achnanthidium sp	2
Achnanthidium subatomus	3
Adlafia bryophila	3
Adlafia minuscula	5
Adlafia minuscula var. muralis	5
Adlafia suchlandtii	4
Amphipleura kriegerana	2
Amphipleura pellucida	2
Amphipleura rutilans	2
Amphipleura sp.	1
Amphora delicatissima	5
Amphora dusenii	4
Amphora fogediana	5
Amphora inariensis	5
Amphora libyca	5
Amphora montana	4
Amphora normanii	4
Amphora ovalis	5
Amphora ovalis var. libyca	5
Amphora ovalis var. pediculus	5
Amphora pediculus	5
Amphora pediculus	5
Amphora pediculus type	5
Amphora sp.	5
Amphora veneta	5
Amphora veneta var. capitata	4
Aneumastus tuscula	3

TaxonName	LTDI2
Anomoeoneis brachysira	1
Anomoeoneis follis	2
Anomoeoneis vitrea	1
Bacillaria paradoxa	5
Bacillaria paxillifer	5
Brachysira brebissonii	1
Brachysira brebissonii fo. thermalis	1
Brachysira follis	1
Brachysira neoexilis	2
Brachysira procera	2
Brachysira serians	2
Brachysira sp.	1
Brachysira styriaca	2
Brachysira vitrea	1
Brachysira vitrea type	1
Caloneis bacillum	4
Caloneis bacillum var. lancettula	4
Caloneis silicula	3
Caloneis silicula var. alpina	3
Caloneis sp.	2
Caloneis tenuis	3
Caloneis undulata	3
Caloneis ventricosa	3
Cavinula cocconeiformis	3
Cavinula jaernefeltii	3
Cavinula pseudoscutiformis	3
Cavinula scutelloides	5
Cavinula variostriata	4
Chamaepinnularia soehrensis	1
Chamaepinnularia soehrensis var. hassica	2
Cocconeis diminuta	2
Cocconeis disculus	4
Cocconeis disculus var. diminuta	2
Cocconeis neodiminuta	3
Cocconeis neothumensis	4
Cocconeis pediculus	5
Cocconeis placentula	4
Cocconeis placentula agg	3
Cocconeis placentula var. euglypta	3
Cocconeis placentula var. lineata	3
Cocconeis placentula var. pseudolineata	3
Cocconeis pseudothumensis	3
Cocconeis sp.	3
Cosmioneis pusilla	4

TaxonName	LTDI2	TaxonName	LTDI2
Craticula accomoda	3	Cymbella minuta	4
Craticula ambigua	3	Cymbella minuta var. silesiaca	3
Craticula cuspidata	4	Cymbella naviculiformis	2
Craticula halophila	4	Cymbella perpusilla	2
Craticula halophilioides	2	Cymbella prostrata	4
Craticula minusculoides	5	Cymbella pusilla	2
Craticula molestiformis	4	Cymbella reichardtii	4
Craticula vixivisiblilis	3	Cymbella reinhardtii	4
Ctenophora pulchella	3	Cymbella rupicola var. rostrata	1
Cymatopleura solea	4	Cymbella silesiaca	3
Cymatopleura solea agg	4	Cymbella sinuata	4
Cymatopleura solea var. apiculata	4	Cymbella sp.	1
Cymbella aequalis	2	Cymbella subaequalis	4
Cymbella affinis	2	Cymbella tumida	2
Cymbella amphicephala	3	Cymbella turgidula	3
Cymbella ancyli	2	Cymbellonitzschia diluviana	4
Cymbella aspera	3	Cymbopleura cuspidata	4
Cymbella brehmii	3	Cymbopleura incerta	2
Cymbella caespitosa	4	Cymbopleura lapponica	2
Cymbella cesatii	1	Cymbopleura naviculiformis	2
Cymbella cistula	3	Cymbopleura reinhardtii	4
Cymbella cistula var. maculata	3	Cymbopleura subaequalis	4
Cymbella cuspidata	3	Decussata placenta	3
Cymbella cymbiformis	2	Delicata delicatula	1
Cymbella delicatula	1	Denticula elegans	3
Cymbella descripta	3	Denticula kuetzingii	4
Cymbella elginensis	3	Denticula sp	3
Cymbella falaisensis	2	Denticula tenuis	2
Cymbella gaeumannii	2	Diadesmis contenta	4
Cymbella gracilis	2	Diadesmis perpusilla	3
Cymbella hebridica	1	Diadesmis sp.	3
Cymbella helvetica	2	Diatoma ehrenbergii	3
Cymbella hustedtii	2	Diatoma elongatum	2
Cymbella incerta	2	Diatoma elongatum var. tenue	2
Cymbella lacustre	3	Diatoma hyemale var. mesodon	2
Cymbella lacustris	3	Diatoma mesodon	2
Cymbella laevis	2	Diatoma moniliformis	3
Cymbella lanceolata	3	Diatoma problematica	2
Cymbella lapponica	2	Diatoma sp.	2
Cymbella leptoceras	3	Diatoma tenue	2
Cymbella leptoceros var. angusta	5	Diatoma tenue var. elongatum	2
Cymbella lunata	2	Diatoma vulgare	5
Cymbella mesiana	2	Diatoma vulgare agg	4
Cymbella microcephala	2	Diatoma vulgare var. ehrenbergii	3

TaxonName	LTDI2
Diatoma vulgaris fo. morphotype constricta	5
Diatoma vulgaris fo. morphotype linearis	5
Diatoma vulgaris fo. morphotype ovalis	5
Diploneis elliptica	3
Diploneis marginestriata	3
Diploneis modica	4
Diploneis oblongella	3
Diploneis oblongella var. gibbosa	3
Diploneis oculata	4
Diploneis ovalis	3
Diploneis parma	4
Diploneis sp.	2
Ellerbeckia arenaria	5
Ellerbeckia sp.	5
Encyonema brehmii	3
Encyonema caespitosum	4
Encyonema elginense	3
Encyonema gaeumannii	2
Encyonema gracile	2
Encyonema hebridicum	1
Encyonema hebridicum	1
Encyonema lacustre	3
Encyonema minutum	4
Encyonema perpusillum	2
Encyonema prostratum	4
Encyonema reichardtii	4
Encyonema silesiacum	3
Encyonema sp.	2
Encyonopsis aequalis	2
Encyonopsis cesatii	1
Encyonopsis falaisensis	2
Encyonopsis microcephala	2
Eolimna minima	4
Eolimna rotunda	5
Eolimna subminuscula	5
Eolimna submuralis	5
Eolimna utermoehlii	4
Epithemia adnata	3
Epithemia argus	3
Epithemia sorex	3
Epithemia sp.	3
Epithemia turgida	2
Eucocconeis depressa	1
Eucocconeis flexella	3

TaxonName	LTDI2
Eucocconeis laevis	3
Eunotia arculus	2
Eunotia arcus	1
Eunotia bactriana	1
Eunotia bidentula	1
Eunotia bilunaris	1
Eunotia bilunaris var. linearis	1
Eunotia bilunaris var. mucophila	1
Eunotia curvata	1
Eunotia curvata var. linearis	1
Eunotia curvata var. subarcuata	1
Eunotia denticulata var. denticulata	2
Eunotia diodon	1
Eunotia elegans	1
Eunotia exigua	1
Eunotia exigua var. tridentula	1
Eunotia faba	1
Eunotia fallax	1
Eunotia flexuosa	3
Eunotia formica	1
Eunotia glacialis	1
Eunotia implicata	1
Eunotia incisa	1
Eunotia intermedia	1
Eunotia islandica	1
Eunotia meisteri	1
Eunotia microcephala	1
Eunotia minor	1
Eunotia monodon fo. monodon	1
Eunotia monodon var. bidens	1
Eunotia muscicola	1
Eunotia muscicola var. tridentula	2
Eunotia naegelii	1
Eunotia nymanniana	1
Eunotia paludosa	1
Eunotia paludosa var. trinacria	2
Eunotia pectinalis	1
Eunotia pectinalis fo. impressa	1
Eunotia pectinalis var. minor	1
Eunotia pectinalis var. minor fo. impressa	1
Eunotia pectinalis var. minor fo. intermedia	1
Eunotia pectinalis var. undulata	1
Eunotia pectinalis var. ventralis	1
Eunotia pectinalis var. ventricosa	1

TaxonName	LTDI2
Eunotia pirla	2
Eunotia praerupta	1
Eunotia praerupta var. bidens	1
Eunotia rhomboidea	1
Eunotia rhyncocephela	2
Eunotia septentrionalis	3
Eunotia serra	2
Eunotia serra var. diadema	2
Eunotia serra var. tetraodon	2
Eunotia silvahercynia	1
Eunotia soleirolii	1
Eunotia sp.	1
Eunotia subarcuatoides	1
Eunotia sudetica	2
Eunotia tenella	1
Eunotia valida	1
Eunotia vanheurckii	1
Fallacia helensis	5
Fallacia insociabilis	4
Fallacia lenzii	5
Fallacia lucinensis	3
Fallacia monoculata	4
Fallacia pygmaea	3
Fallacia sp.	5
Fallacia subhamulata	5
Fallacia sublucidula	4
Fallacia tenera	5
Fistulifera saprophila	4
Fragilaria bicapitata	1
Fragilaria bidens	3
Fragilaria brevistriata	4
Fragilaria capucina	2
Fragilaria capucina agg	1
Fragilaria capucina subsp. intermedia	1
Fragilaria capucina var. amphicephala	2
Fragilaria capucina var. austriaca	2
Fragilaria capucina var. capitellata	2
Fragilaria capucina var. distans	3
Fragilaria capucina var. gracilis	2
Fragilaria capucina var. mesolepta	3
Fragilaria capucina var. perminuta	3
Fragilaria constricta fo. constricta	2
Fragilaria constricta fo. stricta	2
Fragilaria construens	4

TaxonName	LTDI2
Fragilaria construens var. binodis	4
Fragilaria construens var. exigua	2
Fragilaria construens var. pumila	3
Fragilaria construens var. subsalina	4
Fragilaria construens var. venter	4
Fragilaria elliptica	4
Fragilaria exigua	2
Fragilaria fasciculata	5
Fragilaria incognita	3
Fragilaria intermedia	1
Fragilaria karelica	2
Fragilaria lapponica	3
Fragilaria leptostauron	5
Fragilaria leptostauron var. dubia	5
Fragilaria mesolepta	3
Fragilaria nanana	3
Fragilaria nitzschioides	2
Fragilaria oldenburgiana	3
Fragilaria pararumpens	2
Fragilaria parasitica	3
Fragilaria parasitica var. subconstricta	4
Fragilaria perminuta	3
Fragilaria pinnata	4
Fragilaria pinnata var. lancettula	4
Fragilaria pinnata var. trigona	4
Fragilaria pseudoconstruens	3
Fragilaria robusta	4
Fragilaria sp.	3
Fragilaria tenera	3
Fragilaria vaucheriae	4
Fragilaria vaucheriae var. capitellata	2
Fragilaria virescens	1
Fragilaria virescens var. capitata	1
Fragilaria virescens var. exigua	2
Fragilariforma bicapitata	1
Fragilariforma constricta	2
Fragilariforma exigua	2
Fragilariforma sp.	1
Fragilariforma virescens	1
Fragilariforma virescens var. capitata	1
Frustulia amphipleuroides	1
Frustulia crassinveria	1
Frustulia erifuga	1
Frustulia krammeri	1

TaxonName	LTDI2
Frustulia rhomboides	1
Frustulia rhomboides fo. capitata	1
Frustulia rhomboides type	1
Frustulia rhomboides var. amphipleuroides	1
Frustulia rhomboides var. crassinervia	1
Frustulia rhomboides var. crassinervia	1
Frustulia rhomboides var. saxonica	1
Frustulia rhomboides var. saxonica fo. undulata	1
Frustulia rhomboides var. viridula	1
Frustulia saxonica	1
Frustulia sp.	1
Frustulia vulgaris	1
Frustulia vulgaris var. capitata	1
Geissleria acceptata	3
Geissleria decussis	5
Geissleria palustris	5
Geissleria schoenfeldii	3
Gomphocymbella ancyli	2
Gomphoneis olivaceoides	3
Gomphoneis olivaceum	4
Gomphoneis quadripunctatum	3
Gomphonema "intricatum" type	2
Gomphonema acidoclinatum	3
Gomphonema acuminatum	3
Gomphonema acuminatum var. coronatum	3
Gomphonema affine	3
Gomphonema angustatum	3
Gomphonema angustatum var. obtusatum	3
Gomphonema angustatum var. productum	3
Gomphonema angustatum var. sarcophagus	3
Gomphonema angustum	3
Gomphonema anoenum	3
Gomphonema augur	5
Gomphonema bavaricum	3
Gomphonema clavatum	4
Gomphonema clevei	3
Gomphonema constrictum	3
Gomphonema cuneolus	3
Gomphonema dichotomum	3
Gomphonema exiguum var. minutissimum	3
Gomphonema gracile	3
Gomphonema grovei	4
Gomphonema hebridense	2
Gomphonema intricatum var. pumilum	3

TaxonName	LTDI2
Gomphonema lateripunctatum	3
Gomphonema micopus	4
Gomphonema minusculum	3
Gomphonema minutum	4
Gomphonema olivaceoides	3
Gomphonema olivaceum	4
Gomphonema olivaceum agg	3
Gomphonema olivaceum var. calcarea	4
Gomphonema olivaceum var. minutissima	3
Gomphonema olivaceum var. olivaceoides	3
Gomphonema parvulum	5
Gomphonema parvulum var. exilissimum	3
Gomphonema procerum	3
Gomphonema pseudoaugur	4
Gomphonema pseudotenellum	4
Gomphonema pumilum	3
Gomphonema sarcophagus	4
Gomphonema sp.	3
Gomphonema staurophorum	3
Gomphonema subtile	3
Gomphonema tergestinum	3
Gomphonema truncatum	3
Gomphonema utae	3
Gomphonema vibrio	3
Gomphonema vibrio var. intricatum	3
Gomphonema vibrio var. pumilum	3
Gomphosphenia grovei	4
Gyrosigma acuminatum	5
Gyrosigma attenuatum	5
Hannaea arcus	2
Hannaea arcus var. amphioxys	1
Hantzschia amphioxys	4
Hantzschia amphioxys fo. capitata	4
Hantzschia virgata var. capitellata	4
Karayevia clevei	4
Karayevia laterostrata	4
Karayevia ploenensis	5
Karayevia sp	4
Kobaysiella subtilissima	2
Krasskella kriegerana	2
Lemnicola hungarica	3
Luticola cohnii	2
Luticola goeppertiana	4
Luticola mutica	4

TaxonName	LTDI2
Luticola saxophila	5
Luticola sp.	3
Luticola ventricosa	4
Martyana martyi	5
Mastogloia elliptica	2
Mastogloia smithii	3
Mastogloia smithii var. amphicephala	2
Mastogloia sp.	3
Mayamaea agrestis	5
Mayamaea atomus	5
Mayamaea atomus var. permitis	5
Melosira sp.	3
Melosira varians	5
Meridion circulare	3
Meridion circulare var. constrictum	2
Navicella pusilla	3
Navicula accomoda	3
Navicula agrestis	5
Navicula angusta	2
Navicula antonii	5
Navicula aquaedurae	2
Navicula arcus	2
Navicula arvensis	3
Navicula atomus	5
Navicula atomus var. permitis	3
Navicula bacillum	4
Navicula bryophila	3
Navicula capitata	4
Navicula capitata var. hungarica	4
Navicula capitata var. lueneburgensis	5
Navicula capitatoradiata	4
Navicula cari	5
Navicula cari var. cincta	4
Navicula caroliniana	3
Navicula caterva	3
Navicula cincta	4
Navicula claytonii	4
Navicula clementis	4
Navicula cocconeiformis	3
Navicula concentrica	4
Navicula constans var. symmetrica	5
Navicula contenta fo. contenta	4
Navicula cryptocephala	4
Navicula cryptotenella	5

TaxonName	LTDI2
Navicula cryptotenella agg	4
Navicula cryptotenelloides	5
Navicula cuspidata	4
Navicula decussis	5
Navicula detenta	2
Navicula dicephala	5
Navicula difficillima	2
Navicula digitoradiata	4
Navicula elginensis	4
Navicula exilis	4
Navicula gallica var. perpusilla	3
Navicula gastrum	3
Navicula germainii	4
Navicula goeppertiana	4
Navicula gracilis	5
Navicula graciloides	5
Navicula gregaria	5
Navicula halophila fo. halophila	4
Navicula halophiloides	2
Navicula heimansioides	3
Navicula helensis	5
Navicula hungarica	4
Navicula ignota var. acceptata	3
Navicula ignota var. palustris	5
Navicula integra	3
Navicula jaernefeltii	3
Navicula lanceolata	4
Navicula lanceolata	4
Navicula laterostrata	4
Navicula lenzii	5
Navicula leptostriata	2
Navicula libonensis	5
Navicula mediocris	2
Navicula menisculus	5
Navicula menisculus var. grunowii	5
Navicula menisculus var. upsaliensis	5
Navicula minima	5
Navicula minuscula	5
Navicula minuscula var. muralis	4
Navicula minusculoides	5
Navicula modica	4
Navicula molestiformis	4
Navicula muralis	4
Navicula mutica	4

TaxonName	LTDI2
Navicula mutica	4
Navicula mutica var. ventricosa	4
Navicula oblongella	3
Navicula phyllepta	3
Navicula placenta	3
Navicula placentula	5
Navicula porifera var. opportuna	3
Navicula pseudoanglica	4
Navicula pseudobryophila	4
Navicula pseudolanceolata	4
Navicula pseudolanceolata var. densilineolata	4
Navicula pseudoscutiformis	3
Navicula pseudotuscula	4
Navicula pupula	4
Navicula pusilla	4
Navicula pygmaea	4
Navicula radiosa	3
Navicula radiosa var. tenella	5
Navicula radiosafallax	4
Navicula reichardtiana	5
Navicula reinhardtii	5
Navicula rhynchocephala	4
Navicula rotunda	5
Navicula salinarum	5
Navicula saprophila	4
Navicula saxophila	5
Navicula schmassmannii	3
Navicula schoenfeldii	3
Navicula scutelloides	5
Navicula seminuloides	4
Navicula seminulum	5
Navicula slesvicensis	4
Navicula soehrensis	1
Navicula soehrensis var. hassiaca	2
Navicula soehrensis var. muscicola	2
Navicula sp.	4
Navicula stroemii	4
Navicula subatomoides	3
Navicula subhamulata	2
Navicula subminuscula	5
Navicula submuralis	4
Navicula subrotundata	4
Navicula subtilissima	2
Navicula suchlandtii	4

TaxonName	LTDI2
Navicula tantula	4
Navicula tenelloides	5
Navicula tenuicephala	3
Navicula tripunctata	5
Navicula trivialis	4
Navicula tuscula	3
Navicula utermoehlii	4
Navicula variostriata	4
Navicula veneta	4
Navicula ventralis	4
Navicula viridula	4
Navicula viridula var. germainii	4
Navicula viridula var. linearis	4
Navicula vitabunda	4
Navicula vixvisibilis	2
Navicula(dicta) schmassmannii	3
Navicula(dicta) sp.	5
Naviculoid (small undiff)	5
Neidium affine	2
Neidium affine var. amphirhynchus	2
Neidium affine var. humerus	2
Neidium ampliatum	3
Neidium binodis	4
Neidium bisulcatum	2
Neidium hercynicum	2
Neidium productum	4
Neidium sp.	2
Nitzschia abbreviata	5
Nitzschia acicularis	4
Nitzschia acidoclinata	2
Nitzschia acula	2
Nitzschia agnita	3
Nitzschia alpina	3
Nitzschia amphibia	5
Nitzschia amphibia var. acutiuscula	5
Nitzschia angustata	3
Nitzschia angustatula	3
Nitzschia angustiforaminata	4
Nitzschia apiculata	4
Nitzschia archibaldii	4
Nitzschia bacillum	3
Nitzschia brevissima	4
Nitzschia capitellata	4
Nitzschia clausii	4

TaxonName	LTDI2	TaxonName	LTDI2
Nitzschia commutata	4	Nitzschia sinuata var. delognei	5
Nitzschia constricta	3	Nitzschia sinuata var. tabellaria	3
Nitzschia debilis	3	Nitzschia sociabilis	5
Nitzschia disputata	3	Nitzschia solita	3
Nitzschia dissipata	5	Nitzschia soratensis	4
Nitzschia draveillensis	4	Nitzschia sp.	4
Nitzschia epithemioides	4	Nitzschia subacicularis	3
Nitzschia epithemoides var. disputata	3	Nitzschia sublinearis	4
Nitzschia filiformis	3	Nitzschia supralitorea	4
Nitzschia flexa	3	Nitzschia tryblionella var. debilis	3
Nitzschia fonticola	4	Nitzschia tubicola	5
Nitzschia frustulum	4	Nitzschia valdestriata	3
Nitzschia frustulum var. bulnheimiana	4	Nitzschia vermicularis	3
Nitzschia gracilis	3	Opephora olsenii	4
Nitzschia hantzschiana	3	Opephora sp.	3
Nitzschia heufleriana	4	Oxyneis binalis	1
Nitzschia hungarica	5	Peronia fibula	1
Nitzschia incognita	3	Pinnularia abaujensis	1
Nitzschia inconspicua	5	Pinnularia appendiculata	1
Nitzschia intermedia	3	Pinnularia biceps	1
Nitzschia lacuum	4	Pinnularia borealis	2
Nitzschia levidensis	3	Pinnularia borealis var. rectangularis	2
Nitzschia liebetruthii	4	Pinnularia brebissonii	2
Nitzschia linearis	4	Pinnularia divergens	1
Nitzschia linearis var. subtilis	4	Pinnularia gibba	1
Nitzschia linearis var. tenuis	4	Pinnularia intermedia	3
Nitzschia microcephala	4	Pinnularia interrupta	1
Nitzschia obtusa	3	Pinnularia irrorata	1
Nitzschia obtusa var. brevissima	3	Pinnularia lundii	1
Nitzschia obtusa var. scalpelliformis	4	Pinnularia major	1
Nitzschia palea	5	Pinnularia mesolepta	1
Nitzschia palea var. debilis	3	Pinnularia microstauron	1
Nitzschia palea var. tenuirostris	5	Pinnularia microstauron var. brebissonii	1
Nitzschia paleacea	4	Pinnularia rupestris	1
Nitzschia paleaeformis	3	Pinnularia sp.	1
Nitzschia perminuta	3	Pinnularia subcapitata	1
Nitzschia perminuta	3	Pinnularia subcapitata var. hilseana	1
Nitzschia pumila	4	Pinnularia undulata	3
Nitzschia pura	4	Pinnularia viridis	1
Nitzschia pusilla	5	Placoneis clementis	4
Nitzschia recta	4	Placoneis dicephala	5
Nitzschia sigma	3	Placoneis elginensis	4
Nitzschia sigmoidea	3	Placoneis gastrum	3
Nitzschia sinuata	4	Placoneis placentula	5

TaxonName	LTDI2
Placoneis porifera var. opportuna	2
Placoneis pseudoanglica	4
Placoneis symmetrica	5
Planothidium calcar	4
Planothidium daui	3
Planothidium delicatulum	5
Planothidium ellipticum	4
Planothidium engelbrechtii	5
Planothidium frequentissimum	5
Planothidium granum	4
Planothidium haukianum	5
Planothidium lanceolatum	5
Planothidium oestrupii	3
Planothidium peragalli	4
Planothidium rostratum	5
Planothidium sp.	5
Planothidium subatomoides	3
Platessa conspicua	5
Psammothidium abundans	2
Psammothidium bioretii	3
Psammothidium chlidanos	3
Psammothidium didymum	4
Psammothidium grishunun fo. daonensis	2
Psammothidium helveticum	3
Psammothidium kuelbsii	3
Psammothidium lauenburgianum	5
Psammothidium levanderi	3
Psammothidium marginulatum	2
Psammothidium rossii	3
Psammothidium scoticum	2
Psammothidium sp.	3
Psammothidium subatomoides	3
Pseudostaurosira brevistriata	4
Pseudostaurosira pseudoconstruens	3
Pseudostaurosira robusta	4
Pseudostaurosira sp.	2
Reimeria sinuata	4
Reimeria sp.	3
Rhizosolenia sp.	3
Rhoicosphenia abbreviata	5
Rhoicosphenia curvata	5
Rhopalodia brebissonii	4
Rhopalodia gibba	2
Rhopalodia gibberula	4

TaxonName	LTDI2
Rhopalodia gibberula var. rupestris	3
Rossithidium linearis	3
Rossithidium nodosum	3
Rossithidium petersenii	3
Rossithidium pusillum	3
Rossithidium sp	1
Sellaphora bacillum	4
Sellaphora minima	4
Sellaphora pupula	4
Sellaphora seminulum	5
Sellaphora sp.	4
Sellaphora vitabunda	4
Simonsenia delognei	5
Skeletonema sp.	4
Stauroforma exiguiformis	2
Stauroneis acuta	5
Stauroneis anceps	3
Stauroneis anceps fo. gracilis	3
Stauroneis anceps fo. linearis	3
Stauroneis anceps var. var. rhomboidalis	3
Stauroneis kriegeri	3
Stauroneis kriegeri fo. undulate	3
Stauroneis obtusa	3
Stauroneis palustris	4
Stauroneis smithii	4
Stauroneis sp.	3
Staurosira construens	4
Staurosira construens fo. subsalina	4
Staurosira construens var. binodis	4
Staurosira construens var. exigua	1
Staurosira construens var. venter	4
Staurosira constuens var. pumila	3
Staurosira elliptica	4
Staurosira oldenburgiana	3
Staurosira sp.	3
Staurosirella lapponica	3
Staurosirella leptostauron	5
Staurosirella leptostauron var. dubia	5
Staurosirella martyi	5
Staurosirella pinnata	4
Staurosirella pinnata var. lancettula	4
Staurosirella sp.	4
Stenopterobia curvula	1
Stenopterobia sigmatella	1

TaxonName	LTDI2
Surirella amphioxys	3
Surirella angusta	3
Surirella bifrons	3
Surirella biseriata	3
Surirella brebissonii	4
Surirella brebissonii var. kuetzingii	4
Surirella crumena	3
Surirella delicatissima	1
Surirella elegans	5
Surirella linearis	4
Surirella minuta	4
Surirella ovalis	3
Surirella roba	3
Surirella sp.	3
Synedra acus	3
Synedra acus var. angustissima	3
Synedra acus var. delicatissima	3
Synedra capitata	3
Synedra delicatissima	3
Synedra delicatissima var. angustissima	3
Synedra famelica	3
Synedra fasciculata	5
Synedra nana	3
Synedra parasitica	3
Synedra parasitica var. subconstricta	4
Synedra pulchella	3
Synedra rumpens	2
Synedra rumpens var. familiaris	2
Synedra sp.	3

TaxonName	LTDI2
Synedra tabulata	5
Synedra tenera	3
Synedra tenera / nana type	1
Synedra ulna	3
Synedra ulna var. aequalis	3
Synedra ulna var. biceps	3
Synedra ulna var. danica	3
Synedra ulna var. oxyrhynchus	3
Synedrella parasticia	3
Synedrella subconstricta	4
Tabellaria binalis	1
Tabellaria fenestrata	2
Tabellaria flocculosa	1
Tabellaria kuetzingiana	2
Tabellaria quadriseptata	1
Tabellaria sp.	3
Tabellaria ventricosa	2
Tabularia fasciculata	4
Tetracyclus emarginatus	2
Tetracyclus lacustris	1
Thalassiosira pseudonana	5
Tryblionella acuminata	3
Tryblionella angustata	4
Tryblionella apiculata	4
Tryblionella debilis	3
Tryblionella hungarica	5
Tryblionella levidensis	5
Tryblionella sp.	4