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Water Framework Directive Intercalibration Technical Report

Northern Lake Macrophyte ecological assessment methods

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Introduction

The European Water Framework Directive (WFD) requires the national classifications of good ecological status to be harmonised through an intercalibration exercise. In this exercise, significant differences in status classification among Member States are harmonized by comparing and, if necessary, adjusting the good status boundaries of the national assessment methods.

Intercalibration is performed for rivers, lakes, coastal and transitional waters, focusing on selected types of water bodies (intercalibration types), anthropogenic pressures and Biological Quality Elements. Intercalibration exercises were carried out in Geographical Intercalibration Groups - larger geographical units including Member States with similar water body types - and followed the procedure described in the WFD Common Implementation Strategy Guidance document on the intercalibration process (European Commission, 2011).

In a first phase, the intercalibration exercise started in 2003 and extended until 2008. The results from this exercise were agreed on by Member States and then published in a Commission Decision, consequently becoming legally binding (EC, 2008). A second intercalibration phase extended from 2009 to 2012, and the results from this exercise were agreed on by Member States and laid down in a new Commission Decision (EC, 2013) repealing the previous decision. Member States should apply the results of the intercalibration exercise to their national classification systems in order to set the boundaries between high and good status and between good and moderate status for all their national types.

Annex 1 to this Decision sets out the results of the intercalibration exercise for which intercalibration is successfully achieved, within the limits of what is technically feasible at this point in time. The Technical report on the Water Framework Directive intercalibration describes in detail how the intercalibration exercise has been carried out for the water categories and biological quality elements included in that Annex.

The Technical report is organized in volumes according to the water category (rivers, lakes, coastal and transitional waters), Biological Quality Element and Geographical Intercalibration group. This volume addresses the intercalibration of Lake Northern macrophytes methods.

Contents

Annexes

1. Introduction

In the Northern Lake Macrophyte GIG:

- Five Member States (Finland, Ireland, Norway, Sweden and UK) compared and harmonised their national lake macrophyte assessment systems;
- All methods address eutrophication and follow a slightly different assessment principles (as Finland includes helophytes and only 2 countries measure macrophyte abundance), still IC was considerers as feasible;
- Intercalibration "Option 2" was used - indirect comparison of assessment methods using a standardized common metrics; a multiple regression model was developed to derive a lake-specific reference value for ICCM;
- The comparability analysis show that methods give a closely similar assessment (in agreement to comparability criteria defined in the IC Guidance), so only Norway did a slight boundary adjustment;
- The final results include EQRs of lake macrophyte assessment systems of Finland, Ireland, Norway, Sweden and UK for low and moderate alkalinity types (all countries), high alkalinity types (only UK and IE).

2. Description of national assessment methods

Five Member States compared and harmonised their national lake macrophyte assessment systems (more detailed descriptions in Annex A):

- Finnish macrophyte classification system (FINMAC) ;
- Ireland - Free Macrophyte Index;
- Norway - National macrophyte index (Trophic Index – TIc);
- Sweden - Trophic Macrophyte Index (TMI);
- UK - LEAFPAC macrophyte lake classification tool.

2.1. Methods and required BQE parameters

All macrophyte assessment systems include taxonomic composition metrics while abundance metrics included in IE and UK methods, not included in NO and SE methods, FI method included only relative abundance.

As a conclusion the NGIG makes the case that it is neither possible nor necessary to include abundance metrics because:

- Lack of suitable data across all MS (no common coverage data, colonisation depth (Cmax) not collected by all countries);
- IE are the only MS with a metric based on Cmax and, since the IE EQR vs ICM relationship is not significantly different from other MS, it could be argued that the omission of Cmax is not critical;

- Further, our own ICCM which is based purely on composition has a highly significant correlation with Secchi depth (r^2 =0.41). In this sense it can be argued that the common metric will integrate macrophyte abundance through the effect of changing productivity on species composition;
- Norway evaluated carefully the relationship between species composition based index and maximum colonisation index. The major conclusion was that species composition-based index had a better response to eutrophication than maximum colonisation depth due to several reasons explained in Annex A.

Table 2.1 Overview of the metrics included in the national macrophyte assessment methods. MP- macrophytes

MS	Full BQE method	Taxonomic composition	Abundance a	Combination rule of metrics
FI	MP	Proportion of type specific taxa (PTST) Percent Model Affinity (PMA) Reference index (RI)	Relative abundance is a part of one metrics (PMA)	Average
IE	MP	%RF (% relative frequency) Chara %RF Elodeids Plant trophic score %RF Tolerant taxa	Zc – maximum depth of colonisation Average depth of presence	Average
NO	MP	Species composition index based on presence/ absence of taxa	Not included	Only one metric
SE	MP	Trophic Macrophyte Index	Not included	Only one metric
UK	MP	Lake Macrophyte Nutrient Index (LMNI) Number of functional groups of macrophyte taxa (NFG) Number of macrophyte taxa (NTAXA)	Mean percent cover of hydrophytes (COV) Relative percent cover of filamentous algae (ALG)	Weighted average

Three member states, UK, FI and IE have accounted for abundance within their tool but not necessarily as a metric per se. However, NO and SE have not used abundance. All of the member state methods tend to be, either a single metric or multiple metrics based on sensitive and / or tolerant taxa because they are best correlated (responsive) with the pressure of enrichment (inferred from TP concentrations).

In that light, it is not considered necessary to account for abundance because

- It is not the strongest response and it is difficult to quantify,

- Only 3 countries actually account for abundance but it is not measured in the same way by these member states;
- Coverage can also vary considerably from year to year and is highly dependent on effort and method of sampling and weather conditions at time of survey.

See Annex B for the analysis supporting this conclusion .

The WISER report section referring to Cmax reported very complex results:

- Cmax relationships were stronger with Secchi depth than TP (usually not evident or weak, normally TP is used to check the relationship with pressure), colour or chlorophyll. Secchi is not normally used as measure of a pressure because it is a response to a pressure as a result of many factors and will largely reflect chlorophyll concentrations which are already considered separately by classifications of the biological quality element phytoplankton;
- The relationships of Cmax with Secchi varied with latitude, varied from year to year (Danish lakes) and differed according to species. No clear outcome on how to implement Cmax.as a metric was suggested and this is also evident from the fact sheet. Factsheet: http://www.wiser.eu/download/WISER-CommonMetric_C_max_March2011.pdf

Sampling and data processing

All countries use quite similar sampling strategies but macrophyte abundance is measured in different ways (Table 2.2).

Table 2.2 Overview of the sampling and data processing of the national phytoplankton assessment methods

Member State	Sampling device	Surveyed compartment/ Habitat / ecotope	How is abundance measured
FIN	Rake	Belt-transect method based on 5 m wide transects. Number of transects are depending on lake size starting from 8 transects (0.5 km2) to 25 transects (large lake). 2/3 of transects are situated at representative sites (not too open or sheltered), 1/3 of transects are on sheltered shallow sites. Restored sites are not monitored.	Frequency and coverage (percent scale: 0,5, 1, 2, 3, 5, 7, 10, 15, 20, 30?90, 100) of all detected species estimated.
IE	Doubleheader rake, bathyscope	The sampling was carried out at a minimum of 4 x 100m transects located perpendicular to the shore each with 9 sampling positions at set distances from shore using a double headed rake thrown 4 times at each sampling	Abundance is expressed as % relative frequency

		position. Observations using a bathyscope were also included.	
NO	Dredge, aquascope	The survey extend from the shore to the maximum depth of colonisation of macrophytes, at different localities/habitats	Semi-quantitative scale where 1 = rare and 5 = dominant
SE	Dredge, grapnel, rake	Whole-lake survey by mainly raking from boat. The survey is stopped when all macrophyte species are recorded	Presence/absence scale
UK		4 - 8 lake sectors should be surveyed depending on lake area. A sector should comprise a 100 metre length of shoreline. It should extend from the shore to the centre of the lake or to the maximum depth of colonisation of macrophytes. The sectors should be arranged to give an approximately equal spread around the perimeter of the lake.	Each indicator taxon present in the lake should be assigned a value (0 -100 %) which is an estimate of the percentage cover of the taxon in the area of the lake surveyed.

National reference conditions

Table 2.3 summarizes the methodology used to derive the reference conditions for lake macrophyte assessment systems in the NGIG countries.

Table 2.3 Overview of the methodologies used to derive the reference conditions for the national macrophyte assessment methods

MS	Finland	Ireland	Norway	Sweden	UK
Key source to derive RC	Existing near-natural reference sites,	Existing near-natural reference sites	Expert knowledge (from WISER) No ref sites ???	Existing near-natural reference sites, Historical data, Least Disturbed Conditions	Existing near-natural reference sites, Historical data, Modelling (extrapolating model results)
Geographical scope of reference definition	Covering whole Finland	Lakes in 11 counties out of 26 in the Republic of Ireland, the majority on the Western side of the country	Covering Norway	Entire country, however only few reference lakes from the Swedish mountain range	Whole of UK (England, Wales, Scotland & Northern Ireland)
Number of ref sites	177 lakes	46	109	49	Ca 600 surveys (mixture of historic and contemporary surveys)
Ref criteria	Mostly based on pressure criteria. < 10% agriculture (in total catchment area), no major point sources, mainly judged from	Expert opinion and also for some lakes through paleo validation*	Expert judgement and water chemistry data	Proportion of clear-cuts within the lakes catchments <10%, that of agricultural land <10% and that of	Sites selected by iterative application of biological and physicochemical criteria. <15% total cover of pressure tolerant taxa, highly pressure sensitive specie present, cover of highly tolerant species <10% total cover, number of aquatic taxa and functional groups > 25th percentile of type specific richness

MS	Finland	Ireland	Norway	Sweden	UK
	visual observation of GIS land-use and population data. Further experts from local environmental centre were used in final determination.			urban areas <0.1%. No lowering of water level. Tot-P <12.5 microgram, Tot-N <300 microgram, pH >6.0	after allowance for lake area, mean cover score per species within global mean interquartile range. No established invasive alien or translocated species, dominant acid tolerant taxa <30% relative cover (the 75th percentile cover of lakes where acid deposition below critical load), filamentous green algae < 10% relative cover. Mean annual concentration of TP < Good/Moderate boundary value determined by UK MEI approach (site specific, details in µ, No evidence of hydromorphologial modification, impacted land cover <20% of catchment area.
Time period	1970-2004		1970 - 2005	1926-2006	1830-2005

*Leira, M., P. Jordan, D. Taylor, C. Dalton, H. Bennion, N. Rose & K. Irvine 2006. Assessing the ecological status of candidate reference lakes in Ireland using palaeolimnology. Journal of Applied Ecology 43: 816?827

National boundary setting

Table 2.4 summarizes the methodology used to derive ecological boundaries.

Table 2.4 Overview of the methodology used to derive ecological class boundaries for lake macrophyte assessment systems.

MS	Finland	Ireland	Norway	Sweden	UK
Pressure assessed	Eutrophication, hydromorphological change	Eutrophication	Eutrophication	Eutrophication	Eutrophication, hydromorphological change
Rationale/ technique of quality class boundary setting	HG – from ref sites, others - equidistant division of continuum	HG – from ref sites, others - ecological changes	HG -– from ref sites, GM - ecological changes	HG – from reference sites, GM - ecological changes	HG – from reference sites, GM - using paired metrics that respond in different ways to the influence of the pressure
H/G boundary	Lower quartile of reference lake index values	From reference lakes where they exist	Lower quartile of reference lake index values	From metric variability at near-natural reference sites (5th percentile of the reference lakes)	Lower 5th percentile of the multimetric EQR in reference sites
G/M boundary	Other boundaries were calculated by dividing rest of the values evenly in four groups for every lake types.	Boundaries were based on points of ecological change for a number of metrics along the pressure gradient TP.	Good/moderate: where stands of the large isoetids, Littorella, Lobelia, Isoetes (in low alkalinity lakes) or Chara spp. (high alkalinity alkes) decrease (“sudden drop”)	Class boundaries are based on the occurrence of species along the P-gradient.. The species used were those showing “sudden drops” in their occurrence beyond the 75% percentile.	Using logistic regressions for each lake type the LMNI scores at which tolerant taxa composed 15, 35, 65 and 90% of the community were identified. The mean of these type-specific EQRs were used to set the HG, GM, MP and PB boundaries.

Results of WFD compliance checking

The table below lists the criteria from the IC guidance and compliance checking conclusions

General conclusion of the compliance checking:

- All methods are WFD compliant;
- The only problem is lack of abundance metrics in SE and NO methods (FI – only relative abundance), this problem was extensively discussed (see above) and conclusion was that abundance metrics are "neither possible nor necessary to include abundance".

Table 2.5 List of the WFD compliance criteria and the WFD compliance checking process and results

Compliance criteria	Compliance checking conclusions
1. Ecological status is classified by one of five classes (high, good, moderate, poor and bad).	FI, IE, NO, UK – compliant Sweden - compliant, however, no distinction between poor and bad ecological status due to lack of data.
2. High, good and moderate ecological status are set in line with the WFD's normative definitions (Boundary setting procedure)	FI, IE, NO, SE, UK – compliant See table above
3. All relevant parameters indicative of the biological quality element are covered (see Table 1 in the IC Guidance) and combination rules defined.	IE, UK – includes both composition and abundance of macrophytes FI – includes composition and relative abundance of macrophytes NO, SE – only composition Explanations provided above
4. Assessment is adapted to intercalibration common types that are defined in line with the typological requirements of the WFD Annex II and approved by WG ECOSTAT	FI, IE, NO, SE, UK – compliant
5. The water body is assessed against type-specific near-natural reference conditions	FI, IE, NO, SE – compliant See table above UK = assessed against site-specific natural reference conditions where predictors include the same variables used to define lake types
6. Assessment results are expressed as EQRs	FI, IE, NO, SE, UK – compliant
7. Sampling procedure allows for representative information about ecological status in space and time	FI, IE, NO, SE, UK – compliant
8. All data relevant for assessing the biological parameters specified in the WFD's normative definitions are covered by the sampling procedure	FI, IE, NO, SE, UK – compliant

9. Selected taxonomic level achieves adequate confidence and precision in classification	FI, IE, NO, SE, UK – compliant

3. Results IC Feasibility checking

Typology

Intercalibration feasible in terms of typology - all assessment methods are appropriate for the common types (except that high alkalinity Atlantic types - 301a and 302a are shared only by UK and IE).

Table 3.1 Evaluation if IC feasibility regarding common IC types

Common IC type	Type characteristics	MS sharing
101	Low alkalinity, clear. Expanded with some lakes less deep than 3 meters (mean depth), also deep lakes might be included Altitude not counted.	All
102	Low alkalinity, humic.	All
201	Moderate alkalinity, clear. Expanded with some lakes less deep than 3 meters (mean depth). Altitude not counted.	All
202	Moderate alkalinity, humic. Altitude not counted	All
301a	High alkalinity clear Atlantic	UK, IE
302b	High alkalinity humic Atlantic	UK, IE

Pressures addressed

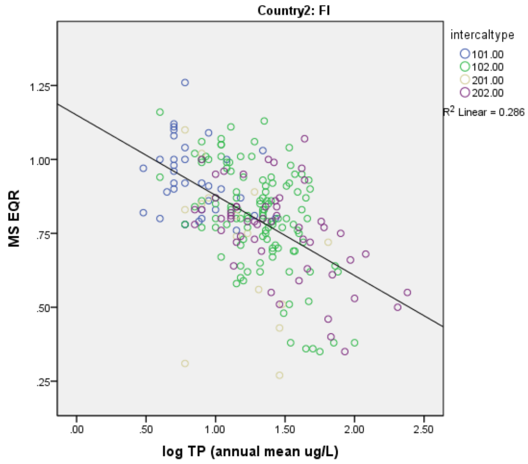
Intercalibration is feasible in terms of pressures addressed by the methods - all are describing eutrophication pressures with high significance (see Table below and Figure 3.1).

Table 3.2 Evaluation if IC feasibility regarding pressures addressed by assessment systems

Method	Pressure	Indicators, strength of the relationship
FI	Eutrophication	TP*: r^2 = 0.29, p = 0.0001
IE	Eutrophication	TP*: r^2 = 0.56, p = 0.0001
NO	Eutrophication	TP*: r^2 = 0.65, p = 0.0001
SE	Eutrophication	TP*: r^2 = 0.15, p = 0.0001 (note: constrained TP gradient compared with other MS)
UK	Eutrophication	TP*: r^2 = 0.48, p = 0.0001

FI

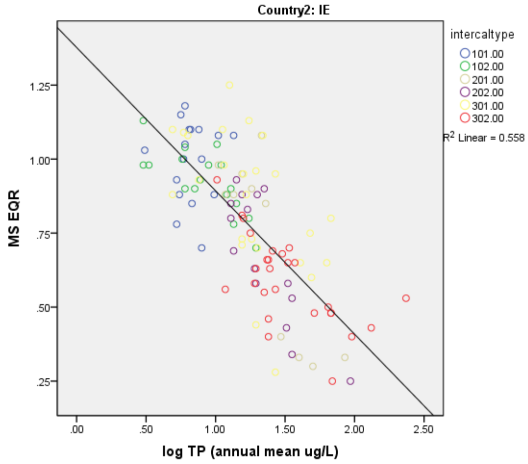
Eutrophication, hydromorphological degradation



TP*: r^2 = 0.29

IE

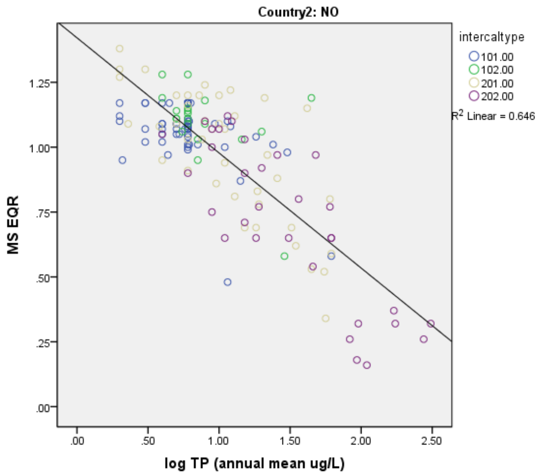
Eutrophication



TP*: r^2 = 0.56

NO

Eutrophication



TP*: r^2 = 0.65

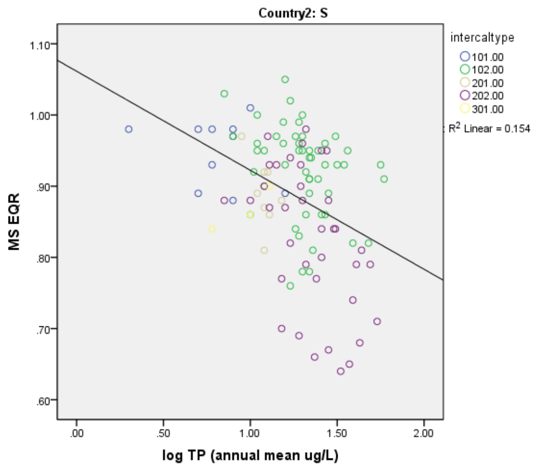
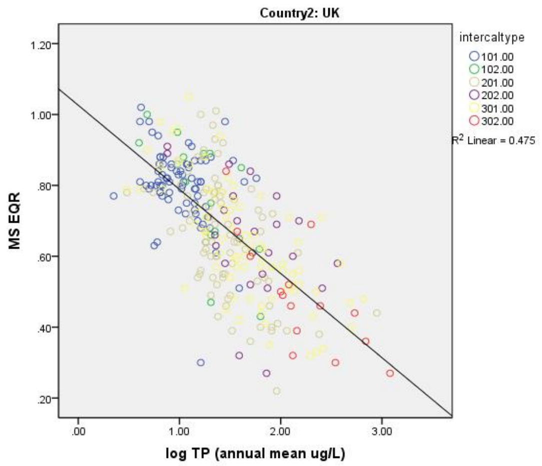
SE	Eutrophication	 TP*: r^2 = 0.15 (note constrained TP gradient compared with other MS)
UK	Eutrophication	 TP*: r^2 = 0.48

Figure 3.1 Pressure-response relationships of NGIG national lake macrophyte assessment systems: a) Finland, b) Ireland, c) Norway, 4) Sweden, 5) UK.

Assessment concept

All national methods follow a similar assessment concept (see table below) although Finland includes helophytes in community analysis.

Table 3.3 Evaluation if IC feasibility regarding assessment concepts

Method	Assessment concept	Remarks
FI	Focus on littoral zone, all macrophyte species are included in assessment (also helophytes)	Helophytes are traditionally included in list of macrophytes
IE	Submerged and Floating-leaved aquatic plants only are used in the assessment. The sampling uses a minimum of 4 transects perpendicular to the shore with 9 sampling position covering a distance of 100m at each transect.	Helophytes are noted but not used in the assessment.
NO	All aquatic macrophytes (incl isoetids, elodeids, nymphaeids, lemnids, and charophytes are sampled.	Helophytes are listed in the field, but not included in the Trophic Index.
SE	All aquatic macrophytes (including isoetids, elodeids, nymphaeids, lemnids, and charophytes are sampled.	Helophytes are listed in the field, but not included in the TMI
UK	• Submerged and floating-leaved aquatic plants are used in the assessment. Shoreline and littoral are surveyed. Metrics include LMNI trophic index • Number of functional groups • Number of Taxa • Mean %cover hydrophytes • Relative cover of macroalgae	Helophytes are not used in the assessment

4. IC dataset collected

Huge dataset was collected within the Northern macrophyte GIG (Table 4.1 and Table 4.2).

Table 4.1 Overview of the Northern GIG macrophyte IC dataset

Type /MS	FI	IE	NO	SE	UK	Total
101	36	18	71	12	91	228
102	125	18	20	53	29	245
201	19	12	44	12	92	**179**
202	55	19	37	39	30	**180**
301	0	38	30	5	97	170
302	0	34	22	0	17	73

Table 4.2 List of the data acceptance criteria used for the data quality control and the data acceptance checking

Data acceptance criteria	Alkalinity, altitude, lake area and TP, plus at least one other relevant pressure indicator (secchi depth, chl_a or catchment land cover) needed for all sites
Data requirements (obligatory and optional)	During the first IC-round environmental data was checked and some outliers were removed from analysis. Biological survey data required >3 ICM scoring taxa for inclusion.
The sampling and analytical methodology	All members are using applied transect method which means that all depth zones must be sampled
Level of taxonomic precision required and taxalists with codes	Species level of identification is used, species list is based on Rebecca and Wiser coding
The minimum number of sites/ samples per intercalibration type	OK
Sufficient covering of all relevant quality classes per type	Quality gradient per MS maximized by using a global dataset with IC lake type as a fixed effect.

5. Common benchmarking

Common approach for setting reference conditions

The general approach used by the GIG was to establish a common method for the estimation of the reference values for the macrophyte common metric (ICCM). Rather than develop a type-specific reference value for the ICCM the GIG agreed to develop a lake-specific reference value using a multiple regression model. This has the benefit of removing the artefact of discontinuities between types and by converting the Observed ICCM to an EQR allows all lakes to be analysed in a single analysis. Thus when the EQR for the common metric was determined for all lakes in the common data set each country used the same model (with an appropriate country effect), thus ensuring the harmonisation of reference conditions for the common metric.

Reference sites

True reference sites were selected according to specific national criteria. The criteria used consisted of pressure data, impact data, knowledge of biology and chemistry, land-use data in conjunction with expert judgement, and in some cases confirmation by palaeo-data.

Due to the high number of lakes in the NGIG area, it was not possible to quantify the pressure criteria for every single lake. Drainage basin borders were not available for all lakes. It is important to note that because the GIG applied the boundary setting protocol to a common data set, substantial differences between MS identification of reference sites would become obvious during the boundary setting procedure as the common metric was applied to the whole database.

The validity of the global population of reference sites was also checked as part of the modelling procedure. There was no significant within-type relationship between the metric and TP (as a surrogate global pressure) and TP was not a significant predictor of variation in the ICM within the global population of benchmark sites.

Additionally SE analysed some lakes with land use data (Annex C). There was no significant correlation with general land use of drainage basins and species composition index. This confirmed the difficulty to utilise land use data for reference condition identification.

Reference criteria

The term 'Member State' has a highly significant fixed effect in the TP - alkalinity relationship, which means that screening based on fixed criteria or continuous benchmarking is not valid due to typological, methodological or biogeographical reasons (Figure 5.1). Although it is arguable that this relationship might simply arise simply because lakes in different countries are differentially impacted the country effect remains highly significant if the analysis is based solely on reference sites (and indeed also on benchmark sites).

Typological reasons include different geological background (e.g. hard limestone versus more easily weathered calcareous rock) whereas methodological influences might include differences in instrumentation, frequency and depth of water sampling, lower limits of measurement resolution. Biogeographical differences might be related to differences in average water temperatures or duration of ice cover and their effect on thermal stratification which will effect nutrient concentrations in surface waters as well as weathering rates in the surrounding catchment. More than 2000 km separate the most northerly and southerly sites in the common dataset and it is unrealistic to expect the same nutrient concentration for a given alkalinity across all lakes.

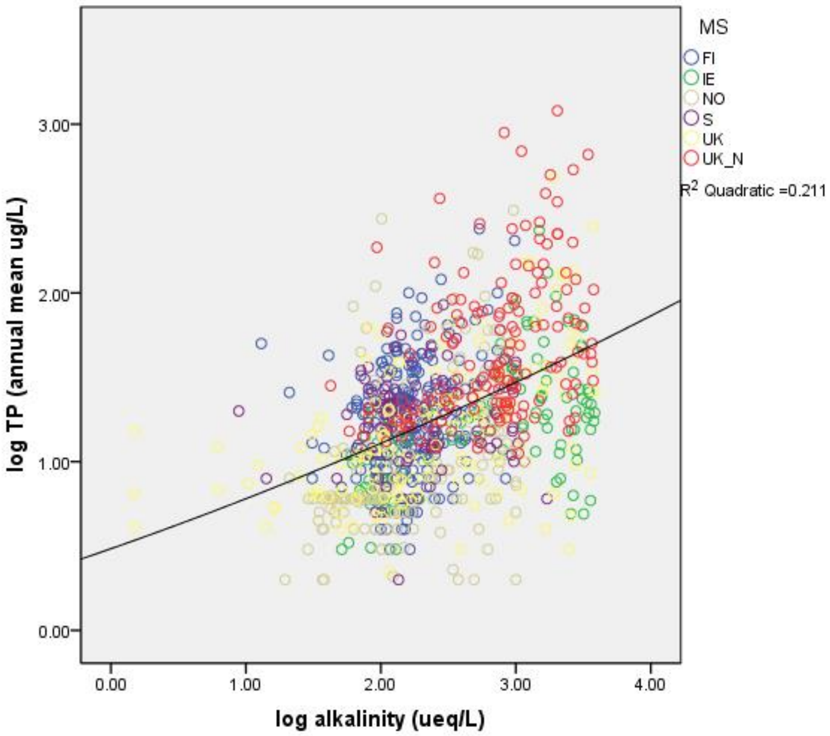


Figure 5.1 Relationship between log TP and log alkalinity in the global GIG dataset.

An iterative approach was used in the screening of reference sites. In the first step the observed ICM values were converted to an ICM EQR by modeling the expected ICM from alkalinity, altitude and colour alone. National reference sites were then screened on a country by country basis based on the pressure v ICM EQR relationships. Where there was a statistically significant inverse relationship between ICM EQR and pressure the most pressurized reference sites were excluded until the relationship was non-significant at $p = 0.05$. The use of a relatively high probability threshold imposes stricter acceptance criteria. After manual screening the global correlation between pressure and ICM is non-significant (Figure 5.2).

These screened reference sites are henceforth referred to as benchmark sites. In the second step a new set of models were then developed based only on benchmark sites to allow calculation of the ICM EQR where the expected ICM value is modeled from the population of benchmark sites. In these models country was included as a fixed effect.

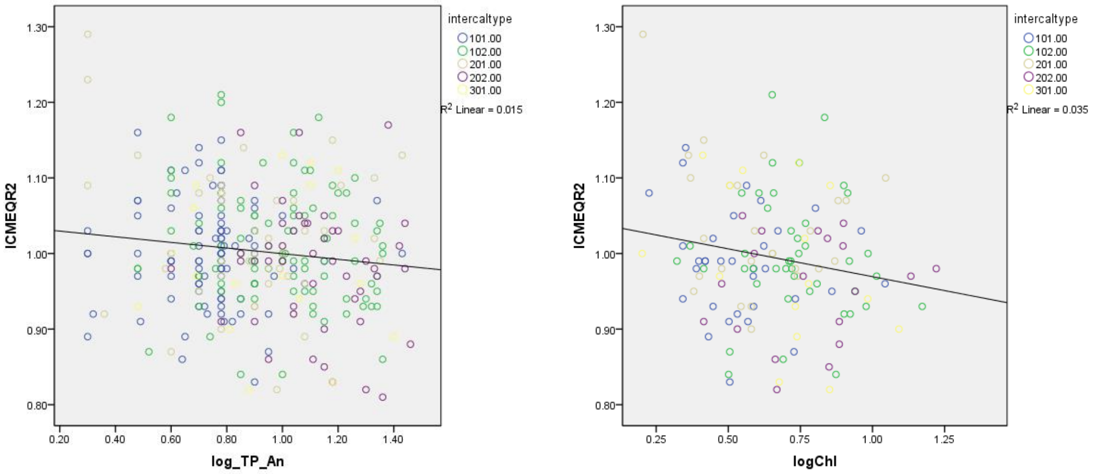


Figure 5.2 Regression between ICMEQR2 and TP (left)/Chl-a (right) at benchmark sites (non-significant at p = 0.05)

Table 5.1 Total number of benchmark sites is a following:

MS/ IC type	101	102	201	202	301	302	Total
FI	31	54	5	26			116
IE	7	10	2	3	9		31
NO	56	16	25	17	8	4	126
SE	9	8	6	3	3		29
UK	5	3	8	0	12	0	28
Total	108	91	46	49	32	4	330

Screening of reference sites:

- We have started with 450 putative reference sites,
- The number was reduced to 432 in phase 1 and further reduced to 394 by removing sites with outlying TP, chl-a or Secchi depth;
- Finally reduced to 330 partly due to revisions of national methods;
- Remaining sites screened at global and within-country level based on ICM EQR v pressure relationships. No evidence of within-IC type pressure-response relationship within these benchmark sites.

Description of setting reference conditions

Reference sites are identified originally by using the chemical screening criteria.

FI: In national macrophyte classification the upper quartile metric value for reference sites was used as a reference. Reference sites were checked against pressure criteria and especially all heavily modified lakes were removed. Also some impacted sites of large lakes were removed. In general it follows WFD determination " The taxonomic composition corresponds totally or nearly totally to undisturbed conditions", which means that type specific species are present and only very few species indicating disturbance have arrived.

SE: For all reference lakes a trophic index based on indicator values of present macrophyte species was calculated. A trophic index ≥ the 75th percentile index determined reference conditions. Reference conditions were calculated separately for the three typology groups.

For Norway, reference lakes have been identified from existing chemical data and expert opinion. All regulated or acidified lakes are removed, as are lakes influenced by other pressures. The trophic index is calculated for all reference lakes, and the median of these values represent the reference conditions. Reference conditions were calculated separately for each lakes type.

UK reference sites were identified at a type-specific level using individual species-pressure relationships indicated by empirical analysis, historical macrophyte records and expert opinion. Finally all reference sites remaining were checked against land cover and total P data where available. Any remaining outliers or sites with known hydromorphological modifications were removed.

IE reference lakes were initially identified using existing chemical data and expert opinion and in some cases validated by palaeolimnology. Originally 63 lakes were considered as candidate reference lakes in Free *et al.* 2006. This list was revised using expert judgement and palaeolimnological results. This list was further refined using a chl cut off value of 7 ug/L and a TP cut off value of 10 ug/L. The former is the highest value of all the HG boundaries for IE types -i.e. did not use type specific HG boundary values. The TP value is indicative of the HG boundary. The remaining 33 lakes, data was used to further define reference condition and calculate the reference values for the Index. Of the lakes remaining, 12 had been examined palaeolimnogically and 11 were shown to be in reference condition.

The range of values in benchmark sites for the common metric (ICM) is shown for each lake type and member state in Figure 5.3 The values show an expected progressive increase along a natural fertility gradient expressed by alkalinity and humic content. It should be noted that the reference ICM is higher in Finland and Sweden for the majority of lake types. This is not an indication of differences in the criteria for selecting reference sites in these countries but a reflection of differences in biological conditions caused by biogeographical and possibly methodological factors. The primary reason for the increased reference values is a W-E biogeographic gradient of increasing taxonomic richness for reference lakes due to increased proximity to continental species pool, decreasing geographical isolation and rate of post-glacial recovery. Survey effort may also be a contributing factor since intensive boat-based whole lake surveys will record a greater number of taxa in a given lake than partial surveys or transects. Observed ICM values will generally increase as richness increases.

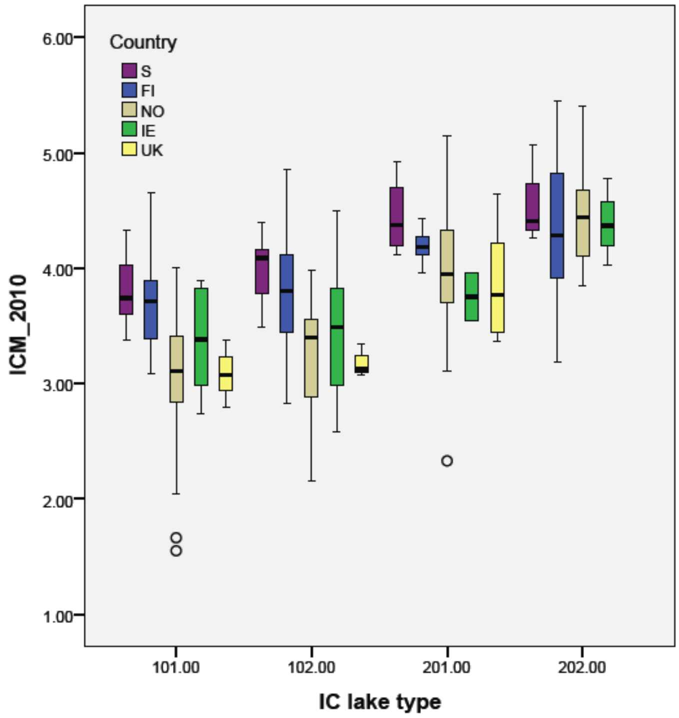


Figure 5.3 Range of observed ICCM values in GIG benchmark lakes

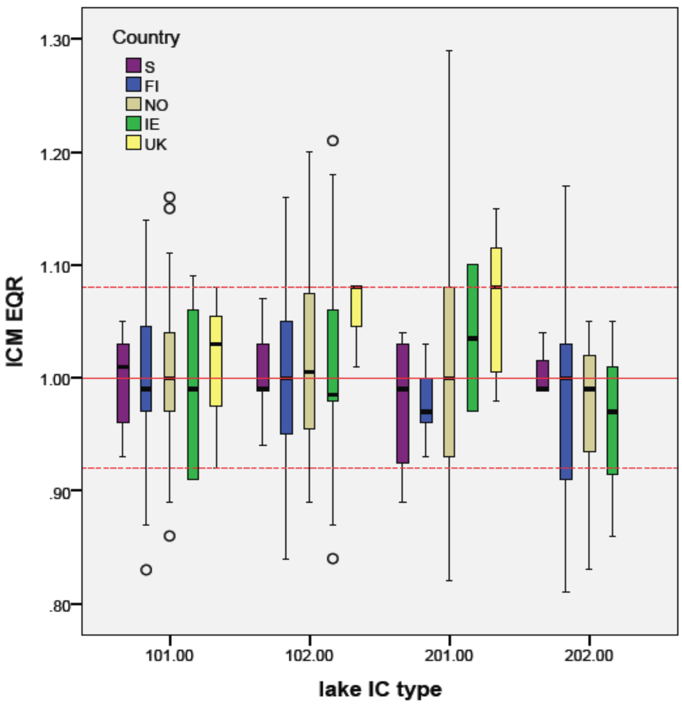


Figure 5.4 Range of intercalibration common metric EQRs in NGIG benchmark sites split by lake type and country. The solid red line shows the global mean ICM EQR and the dashed red lines indicate upper and lower standard deviation.

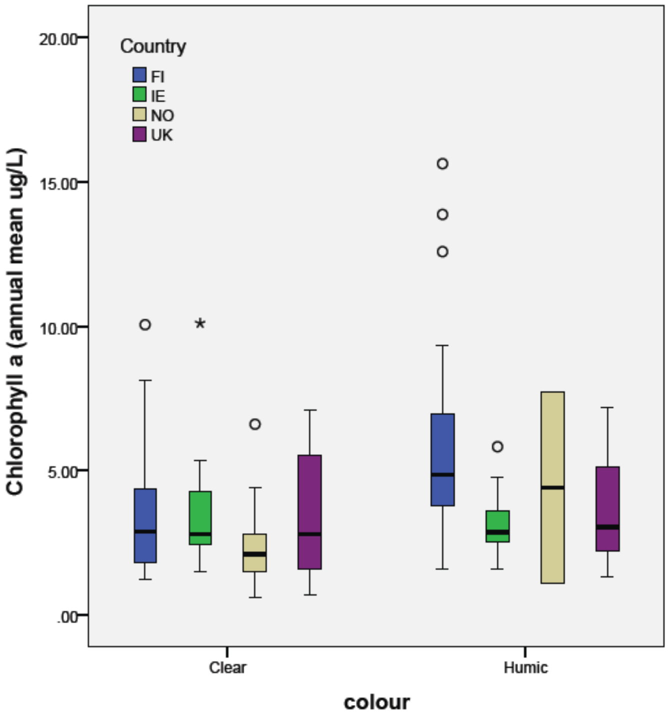


Figure 5.5 Range of chlorophyll a values in N-GIG low and moderate alkalinity benchmark lakes, split by colour type and country.

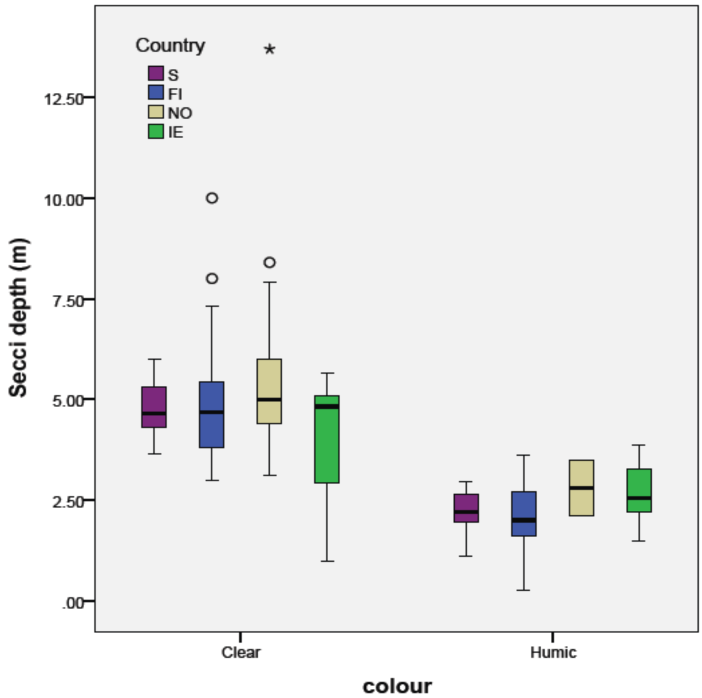


Figure 5.6 Range of water transparency (mean Secchi depth m) for low and moderate alkalinity benchmark lakes in NGIG divided by colour type and country

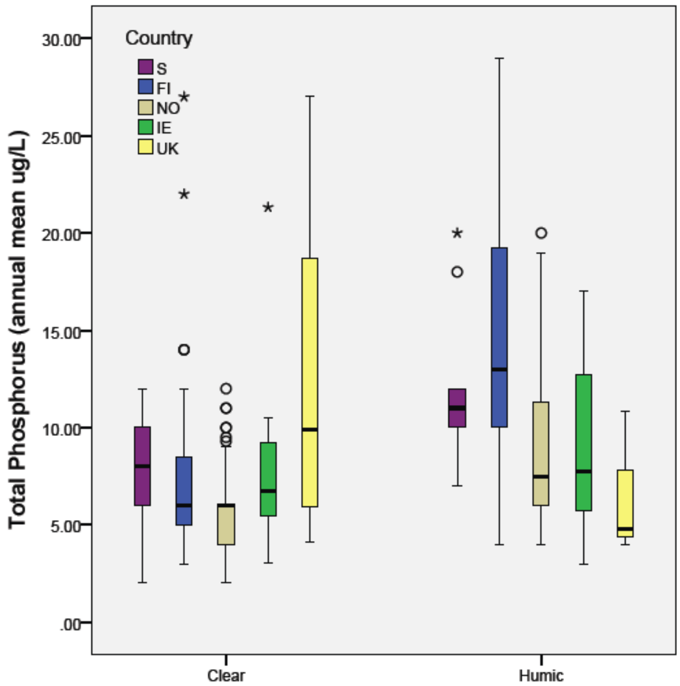


Figure 5.7 Range of total phosphorus (Tot Pug/l) for low and moderate alkalinity benchmark lakes in NGIG divided by colour type and country

If the model for predicting expected ICM at benchmark sites is adequate to remove the country effect then the range of ICM EQRs (Figure 5.4) should be similar for each country. . This is confirmed by Figure 5.4. The global mean ICM EQR is 1±0.08.

A comparison of other impact indicators, phytoplankton biomass (chlorophyll a) and water transparency (Secchi depth) in reference sites do not reveal any clear differences between Finland and Sweden in comparison to other countries in the NGIG (Figure 5.5 and Figure 5.6). Further tot P values are quite the same (Figure 5.7) and we conclude that the GIG selection of reference sites is broadly consistent within the GIG

Benchmark standardisation

The fixed effect of country was considered in a global model to derive the expected ICM value for any reference site in any MS. The country effect was highly significant (alongside the effect of alkalinity, colour, and altitude) and provided a set of country-specific intercept deviates for use in the model (see Table below). Thus, based on the model below for a given alkalinity, colour and altitude, relative to the UK (arbitrary baseline), SE has an ICM value in benchmark sites that is on average 0.223 units higher whilst the value for IE is on average 0.332 units lower.

Table 5.2 Country-specific intercept deviates for use in the model

MS	Country offset
FI	-0.04
IE	-0.332
NO	-0.327
SE	0.223
UK	0

6. Comparison of methods and boundaries

IC Option and Common Metrics

IC Options 2 was used: Country specific indices and their calculation principles were different in different countries. Due to lack of relevant data the full methods of UK, IE and FI could only be applied to their own data, or at best, also that of one additional country.

The differences in data acquisition:

- Finnish data included results from several surveys between years 1980-2004, mainly done by transect method or as whole lake survey, where also depth distribution of different plants was taken into account. Maximum colonisation depth was not calculated and indices used also helophytes and bryids in calculations.
- The Irish dataset was generated from a research project involving the surveying of over 200 lakes in a 3 year period. 4 research fellows were involved and all were trained prior to undertaking sampling. The sampling was carried out throwing a double headed rake 4 times at each sampling position –9 in all at set

distances from shore - along 100m transects. The number of transects varied with lake area with a min. of 4. BSP at the national level was determined using this dataset. Max. col. depth was included in analysis, but helophytes were not analysed.

- The Swedish dataset was heterogeneous regarding e.g. purpose of study, date of sampling and applied field method. It contained macrophyte data collected 1929-2005. The data were collected using different methods, but mainly a whole lake survey method was applied sampling the species at a present-absence scale. No quality assurance procedures were applied. Max. col. depth was not included nor helophytes.
- The Norwegian dataset is based on several whole lake surveys from 1920-2005, however, homogeneous sampling methods were utilised including all depth zones. Max. col. depth was not included nor helophytes.
- The UK dataset was extracted from a master dataset constructed from all known sources within the UK to facilitate the development of the UK assessment method. Surveys used were those from Universities, surveys carried out for conservation and regulatory agencies by qualified surveyors. The dataset was checked by national experts during development of the UK WFD methods and there are no significant data quality issues. Max. col. depth was not included nor helophytes. Relative cover of macroalgae and mean coverage of all aquatic taxa is needed.

Taking into account all of these facts it was not possible to robustly apply all indices/all countries approach in a reciprocal manner to the data of all other MS. An Option 2 approach was therefore dictated.

IC common metrics

New combined N-CB GIG lake macrophyte common metric was implemented:

- It was derived from cross-GIG dataset (1600 lake macrophyte surveys in 15 member states in NW and Central Europe);
- Calculated arithmetic mean [TP] for each taxa based on lakes occupied (total 174 scoring taxa);
- TP values were log transformed and rescaled to continuous gradient from 1 (ultraoligotrophic) to 10 (hypertrophic);
- ICM value per lake was calculated based on unweighted average scores of all taxa present;
- Linear correlation (Figure 6.1) between ICM and mean log TP (internal calibration) is $r^2 = 0.51$ ($p < 0.0001$);

Observed ICM values were then converted to an EQR using a GLM of benchmark sites with alkalinity, colour, altitude and country to predict expected ICM values

EQR = (Obs ICM-10)/(Exp ICM-10).

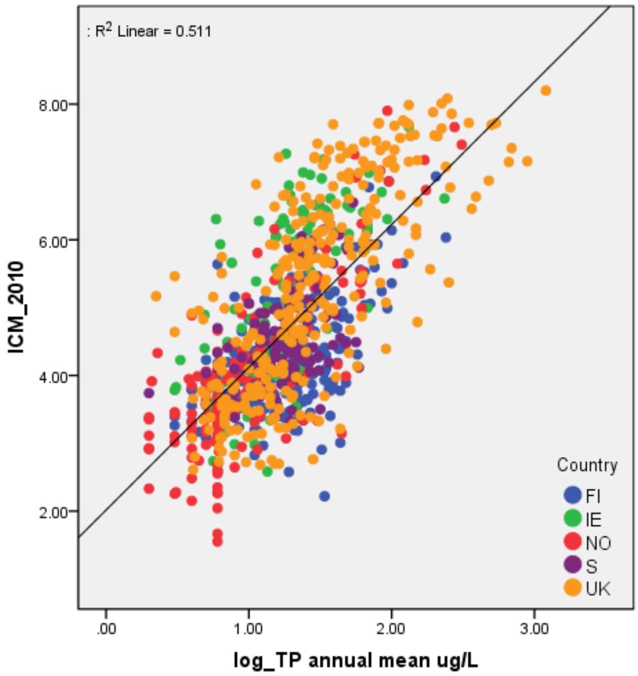


Figure 6.1 Relationship between ICM and log TP annual mean (annual mean value of total phosphorus concentration)

Common metric was modelled in new population of reference sites:

- Alkalinity main predictor, weak effect of altitude and colour;
- Mean lake depth not available for all reference sites and did not significantly improve model based on those sites for which depth was available;
- Country effect highly significant ;
- SE and FI tend to have higher ICM values for a given set of conditions, probably reflecting higher numbers of taxa in these lakes as noted in Phase 1 analyses;
- Essentially there is a more 'continental' part of NGIG represented by SE and FI and an 'atlantic' part represented by IRL, NO and UK.

EQR values for all sites in dataset (1076 lakes) were derived based on observed and expected ICM values

Results of the regression comparison

All countries (FI now included with revised method) have highly significant relationships between MS EQR and the common metric EQR for the sites in their own countries**.** Therefore comparison of methods is feasible using the common metric (i.e. Option 2 approach). A graphical representation of the relationship between MS EQR and the common metric, expressed as an EQR is provided in Figure 6.2

Using the frequency distribution of EQR values of reference sites an independent 'classification' of all sites in the dataset was created with the HG boundary set at 0.89 (equivalent to the lower 10th percentile of the EQRs of the benchmark sites), and the GM boundary set at 0.78 (equivalent to the lower 1 percentile of the EQRs of the benchmark sites). This gives a useful impression of the quality of MS sites from an international perspective based on the common metric and can be used for guidance.

All methods have significant regressions to the common metrics (see table below)

The correlation coefficients (r) and the probability (p) for the correlation of each method with the common metric (analyses based on all types combined)

Table 6.1 The correlation coefficients (r) and the probability (p) for the correlation of each method with the common metric (PCM)

Member State/Method	r	p
FI	0.62	<0.001
IE	0.72	<0.001
NO	0.91	<0.001
SE	0.67	<0.001
UK	0.87	<0.001

The outcomes of the regression complied with the following characteristics according to the IC Guidance:

- All relationships were highly significant p<=0.001;
- Assumptions of normally distributed error and variance (homoscedasticity) of model residuals must be met;
- Common metric must represented all methods (r2>0.5);
- Observed minimum r2 was > half of the observed maximum r2;(not fulfilled 0.39(FI)> 0.82/2 (NO)
- Slope of the regression should lie between 0.5 and 1.5.

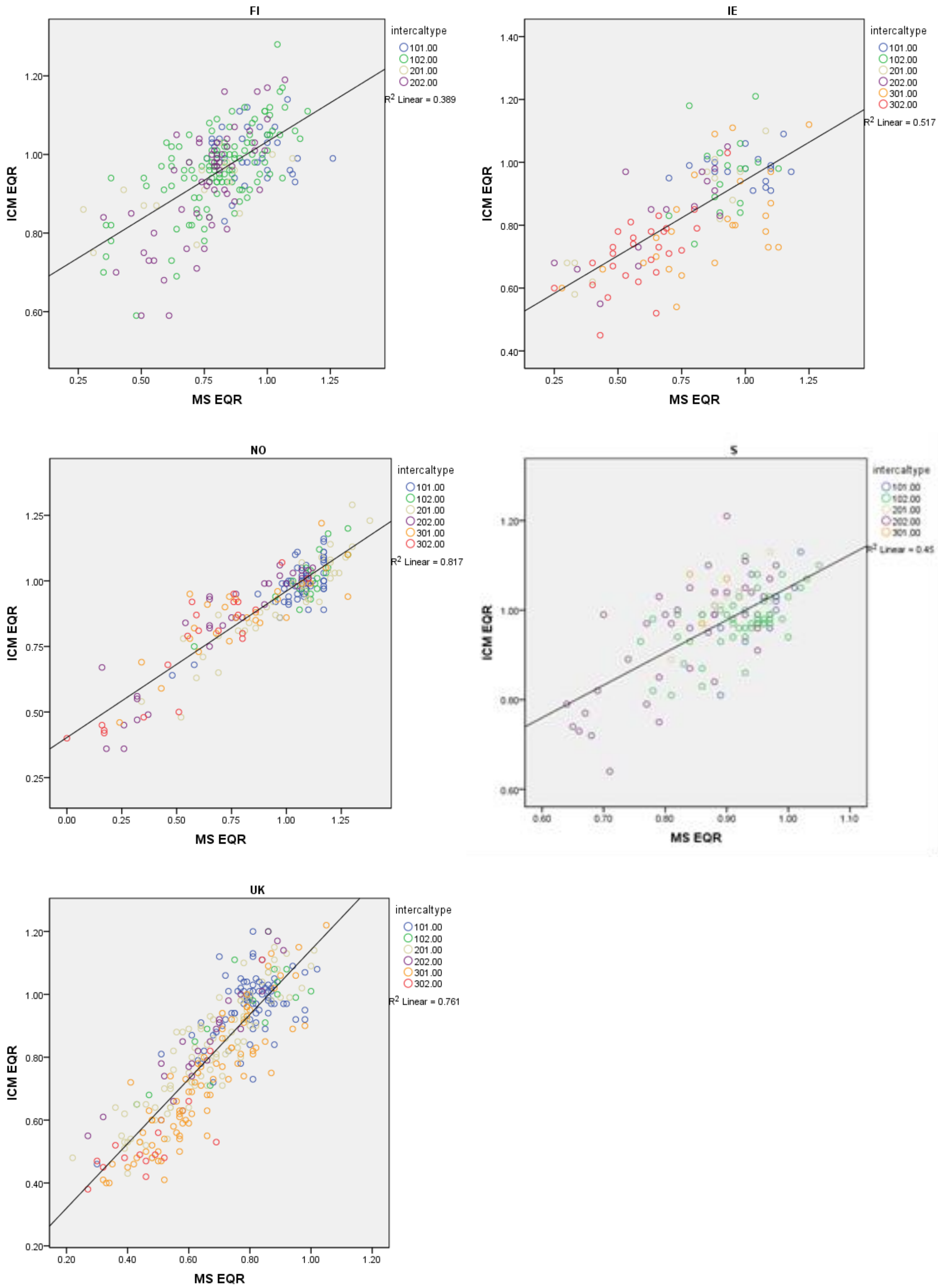


Figure 6.2 Summary of common metric EQR v MS EQR relationships for Low, Moderate and High Alkalinity lakes: a) FI, b) IE, c) NO, d) SE, e) UK.

Conclusion: All national methods included. FI and SE did not consider high alkalinity (types 301 and 302 lakes).

By comparing MS EQR with a site specific benchmark standardised ICM EQR all commonly compared IC types are considered in a single analysis – more powerful because gradient length is maximised and precision of model coefficients is increased. But possible effect of IC type needs to be considered. which is significant in case of IE. S and UK (NO)

Evaluation of comparability criteria

- An Option 2 approach using the common metric EQR has been used to establish the relative positions of MS HG and GM boundaries on the ICM EQR scale.
- The comparison of boundaries has been undertaken at the level of low and moderate alkalinity lakes combined and the outcomes averaged for each MS across the respective lakes types.
- Bias of national methods has been compared on the basis of class equivalents.

Comparability criteria are acceptable according to the Annex V requirements:

- All boundary biases for MS methods are above -0.250 ;
- SE boundaries precautionary so > 0.25.

The results of the boundary comparison are shown In Table 6.2 and Table 6.3.

Table 6.2 provides a comparison of the bias in national methods expressed as class widths **prior to adjustment**. In general the boundaries of Sweden are precautionary relative to the global view.

Table 6.2 Preliminary results of methods comparison in low and high alkalinity lakes (class boundary biases)

	NO	UK	IE	S	FI
HG	-0.10	0.09	-0.13	0.39	-0.03
GM	-0.22	-0.16	-0.15	0.33	0.22

Iterative adjustments of the necessary class boundaries have then been undertaken to achieve a position where all **boundaries lie within 0.25 class units of the global mean on the national scale**:

- Sweden opted to remain precautionary but is only just outside the quarter of a class harmonisation band.
- The changes made by NO reflect the use in this national method of lake-type specific class boundaries which were observed to produce an unduly relaxed view of the GM boundary in type 201 lakes. A tightening of the NO boundaries for this lake type thus produces a more harmonised view of the GM boundary.

The final biases are summarised in Table 6.3 below.

Table 6.3 Final IC result of methods comparison in low-high alkalinity lakes (class boundary biases)

	NO	UK	IE	S	FI
HG	-0.06	0.09	-0.13	0.39	-0.03
GM	-0.12	-0.16	-0.15	0.33	0.22

To investigate the differences in macrophyte communities inside high alkalinity lakes detrended correspondence analysis was performed (Figure 6.3). It seems to be that high alkalinity lakes differ significantly depending on geographical position (Figure 6.3). Atlantic subtypes in IE and UK have different species composition compared to more continental subtypes of NO and SE. Species pool in these relatively small lakes is more isolated than the species pool in other lakes usually part of larger lake chains.

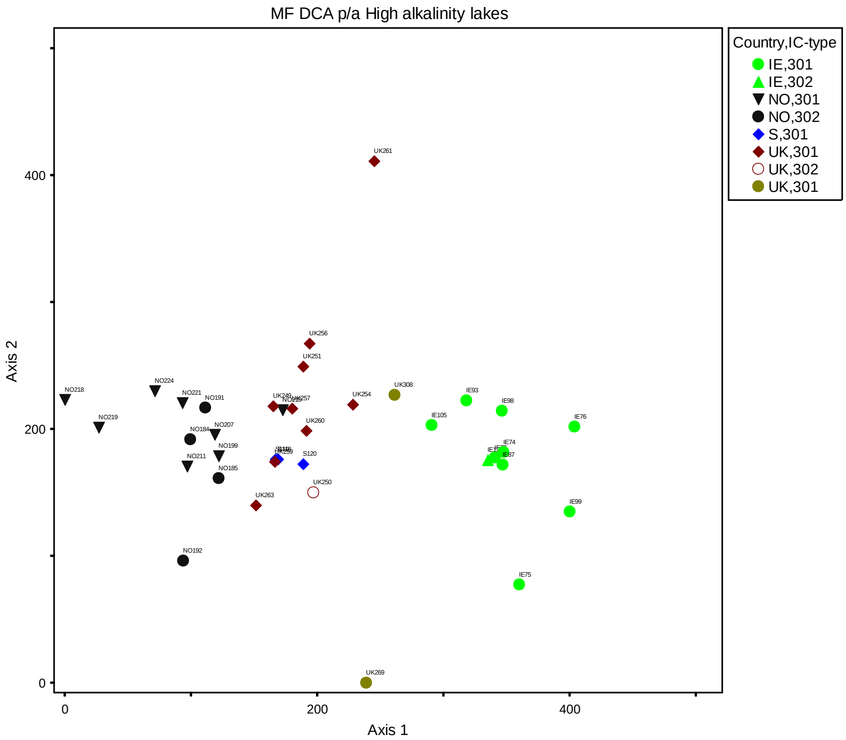


Figure 6.3 DCA ordination of high alkalinity lake types (301. 302).

Therefore, a bilateral comparison was undertaken based only on IE and UK (only for Atlantic subtype of high alkalinity lakes, shared by UK and IE), results in the table below

Table 6.4 Final result in high alkaline lakes based on bilateral comparison of IE and UK.

Boundary	UK	IE
HG	0.09	-0.08
GM	-0.06	0.08

The final boundaryvalues are shown in Table 6.5.

Table 6.5 Class boundaries to be included in the IC Decision

Member State	Classification	Ecological Quality Ratios	
	Method	High-good boundary	Good-moderate boundary
FI	Finnmac	0.8 (all types)	0.6 (all types)
IE	Free Macrophyte Index	0.9 (all types)	0.68 (all types)
NO	National macrophyte index (Trophic Index – TIc)	Type 101: 0.98 Type 102: 0.96 Type 201: 0.95 Type 202: 0.99	Type 101: 0.85 Type 102: 0.87 Type 201: 0.75 Type 202: 0.77
SE	Trophic Macrophyte Index (TMI)	Type 101: 0.93 Type 102: 0.93 Type 201: 0.89 Type 202: 0.91	Type 101: 0.80 Type 102: 0.83 Type 201: 0.78 Type 202: 0.78
UK	LEAFPACS lake macrophyte classification tool	0.8 (all types)	0.66 (all types)

7. Description of biological communities

7.1. Reference communities

Typical in soft water lakes consists of isoetids whereas in more alkaline lakes charophytes and elodeids are more common. Due to large geographical gradient pool of species was different in continental side and Atlantic side of region.

In general. the typical species are the following:

At the continental side of GIG (Finland. Sweden. partly Norway) in low alkalinity clear water lakes (101) reference communities consists of large isoetids such as *Isoetes lacustris*. *I. echinospora*. *Lobelia dortmanna* and further elodeids like *Myriophyllum alterniflorum*. Also helophytes such as *Equisetum fluviatile* and *Phragmites australis* are common. By increasing humic substances floating leaved species such as *Nuphar pumila* and *Nymphaea alba* become more common and also soft bottom elodeids like *Potamogeton perfoliatus* and *P. gramineus* are abundant.

Increasing alkalinity brings more *Potamogeton* species such as *Potamogeton berchtoldii* and *P. praelongus* whereas isoetids are more rare at the continental side of GIG. Also fine leaf species such as *Myriophyllum sibiricum* are more common. Moderate alkalinity humic lakes does not differ very much of clear water ones. but vegetation is more limited and favours soft bottom species.

At the Atlantic side of GIG (Ireland. UK) the core component of *Juncus bulbosus*. *Littorella uniflora*. *Potamogeton polygonifolius*. *P. natans*. *Isoetes lacustris*. *Myriophyllum alterniflorum*. *Menyanthes* and *Sparganium angustifolium* are all highly likely to occur in clear water lakes. However. richness is likely to be somewhat higher than the equivalent peaty lakes due to the frequent presence of taxa such as *Eleogiton fluitans*. *Subularia aquatica* and various species of *Utricularia*. *Nitella* and *Callitriche*.

Moderate alkalinity lakes: most of the species that characterize reference conditions in low alkalinity lakes are still present and form the dominant or co-dominant component of the vegetation. However. they are also associated with a high diversity of other taxa which now include several of the more nutrient sensitive charophytes (e.g. *Chara aspera*. *Chara virgata*) plus a range of pondweeds that would normally indicate increased enriched in low alkalinity lakes.

High alkalinity reference lakes are dominated by charophytes but also contain a high proportion of the more mesotrophic element of the flora that is found in reference moderate alkalinity lakes (e.g. *Myriophyllum alterniflorum*. *Potamogeton alpinus*. *P. gramineus*).

Reference lake communities are described in more detail in Annex C

7.2. Description of “good” status communities

Type 101: Low alkalinity. clear lakes

A graphical representation of functional changes in type 101 lakes along the ICM EQR gradient is shown in Figure 7.1. The overall pattern with decreasing ecological quality is for a marked reduction in isoetids. and a weaker reduction ij the contribution of myriophyllids and charids. accompanied by an expansion in nymphaeids. elodeids. callitrichids and lemnids.

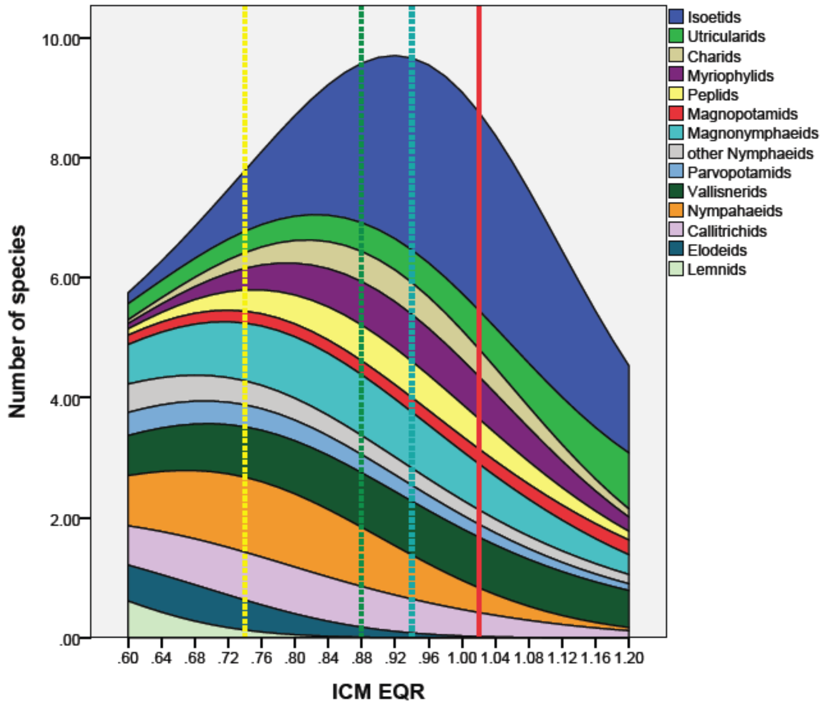


Figure 7.1 Functional changes in the composition of aquatic vegetation along a global ecological gradient in low alkalinity. clear lakes. Red = reference. Blue =H/G. Green=G/M. Yellow=M/P.

Figure 7.1 incorporates the global mean position of the class boundaries of the five participating member states. High status is mainly distinguished from Good status by the absence of the most stress tolerant groups (elodeids and lemnids) and a greatly superior contribution of isoetids (typically accounting for about one third of the community). Good status is differentiated from Moderate status by only a very small contribution of lemnids and elodeids at good status. and marked excess of isoetids + myriophyllids over nymphaeids + callitrichids which shifts to an excess of the latter at moderate status.

Type 102: Low alkalinity. humic lakes

A graphical representation of functional changes in type 102 lakes along the ICM EQR gradient is shown in Figure 6.2. The overall pattern with decreasing ecological quality is for a marked reduction in isoetids and to a lesser extent peplids. accompanied by an expansion in nymphaeids and magnonymphaeids. In the more impacted lakes these latter growth forms are increasingly replaced by elodeids. hydrocharids. callitrichids and lemnids.

Figure 7.2 incorporates the global mean position of the class boundaries of the five participating member states. High status is mainly distinguished from Good status by a more or less complete absence of the most stress tolerant groups (elodeids. lemnids and hydrocharids) and a superior contribution of isoetids compared to nymphaeids. Good status is differentiated from Moderate status by only a very small contribution of lemnids and elodeids at good status and a balance of isoetids + peplids versus nymphaeids + magonymphaeids which shifts strongly in favour of the latter at moderate status.

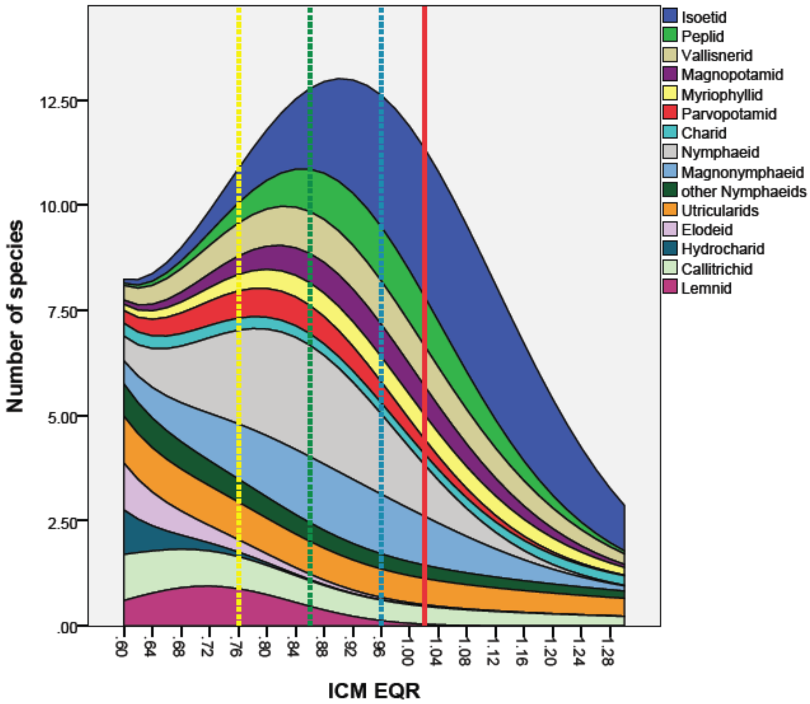


Figure 7.2 Functional changes in the composition of aquatic vegetation along a global ecological gradient in low alkalinity. humic lakes. Red = reference. Blue =H/G. Green=G/M. Yellow=M/P.

Additionally, Figure 7.3 shows distribution of mean annual TP, number of scoring species and Secchi depth for lakes classified by common metric boundaries for different NGIG lake types.

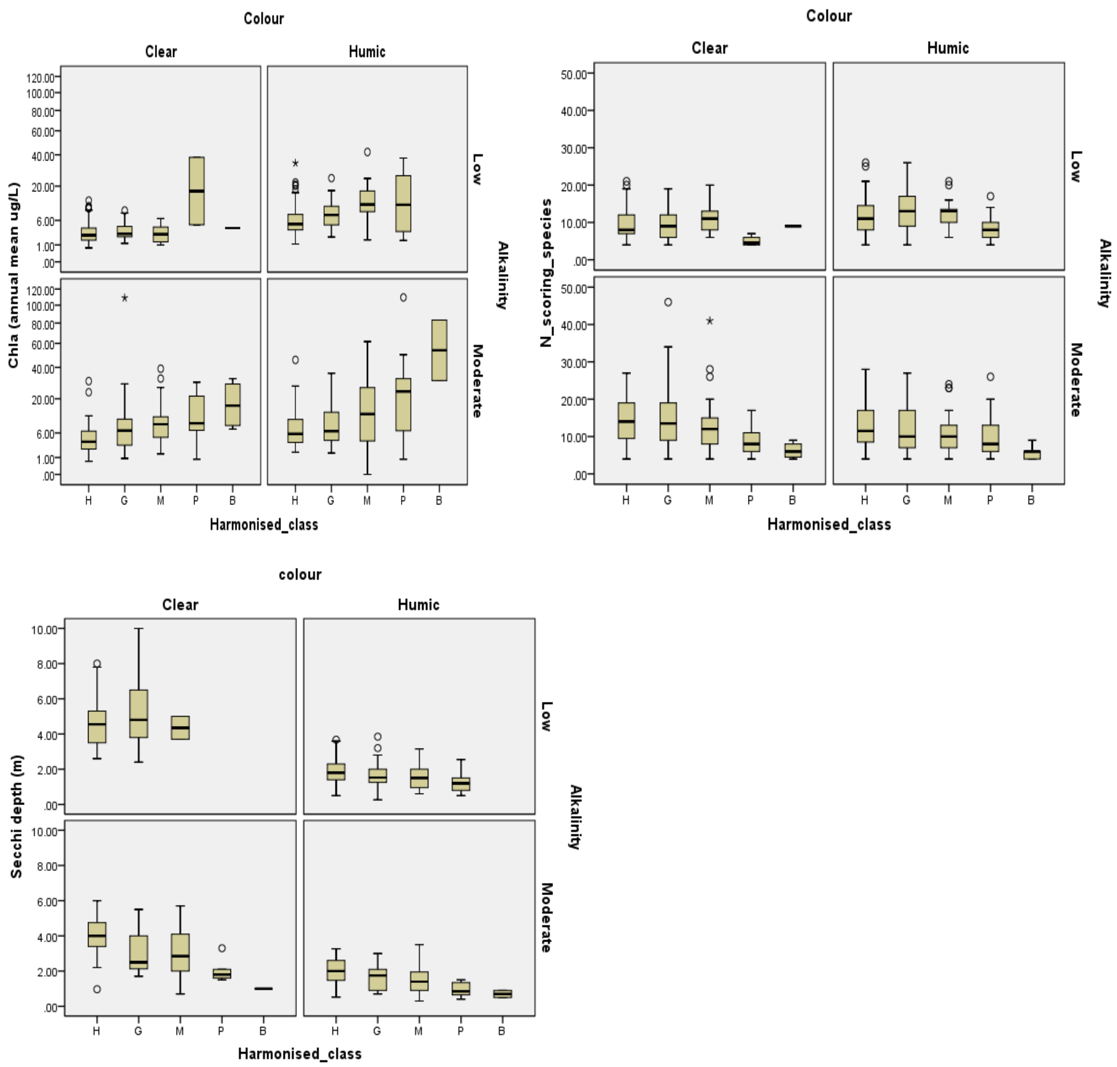


Figure 7.3 Distribution of mean annual TP, number of scoring species and Secchi depth for lakes classified by common metric boundaries for NGIG lakre types (clear low alkalinity, clear moderate alkalinity, humic low alkalinity, humic moderate alkalinity lakes)

Annexes

A. Descriptions of Lake Macrophyte assessment methods

A.1 Lake macrophyte assessment system in Finland

Introduction

Finnish classification system for lake macrophytes was developed to take into account multiple pressures and humic rich waters. It is based on a multimetric index consisting of three metrics (Proportion of type specific taxa PTST, Percent Model Affinity PMA and Reference Index RI) supposed to meet the normative definitions of WFD. Reference values for each metric are calculated using a reference lake typology consisting of 12 lake types based mainly on lake area, depth and water colour. Further reference values are given separately for lakes of northern and southern geographical region due to strong latitudinal patterns of aquatic macrophyte distribution (Heino & Toivonen 2008).

Field methods

Aquatic macrophyte data was collected in various surveys between 1970 - 2004 (see full list in Leka et al. 2008). The majority of older data was sampled by using phytolittoral inventories (PI), but latest data is collected by main belt transects method (TS). Both methods include visits at several sites and with survey ranging from uppermost littoral (helophyte zone) to deepest growing points of submerged plants.

Macrophyte data includes both real hydrophytes and helophytes (Kuoppala et al. 2008). Currently used data consisted of 177 reference lakes and 131 impacted lakes with impaired status (Table A.1). Research lakes were relatively evenly distributed along lake rich area of Finland although majority of reference lakes (68 %) were situated in the northern part of country and further 73 % of impacted lakes were situated in southern Finland. Division between south and north Finland was done by using biogeographical division for littoral plants (Eurola 1999). North-south boundary is situated at Oulujoki river basin, where all lakes situated more than 120 m a.s.l belong to northern types and lakes situated less than 120 m a.s.l. belong to southern types.

Reference lakes

Reference lakes have been selected mostly based on pressure criteria. The main pressure criteria are: < 10% agriculture (in total catchment area), and no major point sources, mainly judged from visual observation of GIS land-use and population data. Further experts from local environmental centre were used in final determination. Current lake typology of Finland follows largely A-typology of WFD with additional division to northern and southern lakes (Table A.1).

*Table A.1 Total number of different lakes and lake types included in Finnish macrophyte data. R = reference lakes, I = impacted lakes. S = southern part of Finland, N = northern part of Finland. *Finnish lake typology has following criteria: size; large > 40 km^2, middle sized = 5 – 40 km^2, small = < 5 km^2, depth; shallow < 3 metres, other depth categories, water colour; clear < 30 mgPtl^{-1}, humic 30 – 90 mg Pt l^{-1}, polyhumic > 90 mg Pt l^{-1} . **lakes situated above tree line, ***naturally eutrophic southern lakes, ****calcerous lakes.*

Lake type*	Size	Depth	Water colour	Reference status		Impacted status	
				South	North	South	North
Vh	Small/middle		Clear	9	12	1	2
SVh	Large		Clear	8	8	15	
Ph	Small		Humic	10	12	7	7
Kh	Middle		Humic	7	9	6	2
SKh	Large		Humic	7	0	14	5
Rh	All		Polyhumic	3	3	2	2
MVh	All	Shallow	Clear	0	6	3	1
MKh	All	Shallow	Humic	10	35	7	13
MRh	All	Shallow	Polyhumic	6	11	11	15
Pola**	All				19		
Rr***	All			7		17	
Rk****	All				14		1

Classification method

Finnish macrophyte classification system is based on a multimetric index consisting of three metrics (Proportion of type specific taxa, Percent Model Affinity and Reference index). Leka et al. (2008) analysed a large dataset consisting of macrophyte data of 773 lakes and ponds. One tested indicator was proportion of type-specific species, which is based on the method developed by Hämäläinen et al. (2002) for benthic invertebrates of rivers. In principle it follows general determination of WFD, where type specific communities of macrophytes are emphasized. Basic assumption is that typical, type specific species are present in 50 % of reference lakes of each type (Table 2).

The presence of type specific species is calculated as follows. The probability of occurrence (p) for each species (i) in any reference lakes (j) belonging to each lake type (k) is first estimated as

$$P_{kj*1} = \frac{\sum kji}{\sum kj}$$

i.e. the ratio between the number of reference sites of the type k occupied by the species i and the total number of reference sites belonging to that type. Species with $P_{kj*1} \geq 0.5$

are considered to be type-specific. The observed number of type-specific species in each lake (j) belonging to a type k is obtained simply as

$$O_{kij} = \sum kji \big| p_{kj*i} \geq 0.5$$

Method is proved to be efficient for benthic invertebrates, but for macrophytes the interpretation is difficult because of a large number of generalist species (Vallinkoski et al. 2004). The index does not take into account the new species arriving with the course of eutrophication. Therefore more advanced index, proportion of type-specific species of total number of species (PTST), was developed.

The share of type specific species in a reference lake was:

$$Sts = \frac{\sum kji}{\sum kji \big| p_{kj*i} \geq 0.5}$$

In other words type specific (reference) species are replaced by other species indicating eutrophication, for example typical soft water isoetids communities are replaced by nymphaeids or lemnids, which are usually rich in biodiversity.

A list of type specific species at different types is presented as Table A.2 and Table A.3.

Ecological quality ratios of relative model similarity (PMA, Percent Model Affinity, Novak & Bode 1992) were counted from values of vegetation indices (Ilmavirta & Toivonen 1986). Method is originally developed for diatom communities, but further applied for benthic invertebrates (Tolonen ym. 2005) and aquatic macrophytes (Vallinkoski ym. 2004, Leka ym. 2003). PMA takes both species composition and their abundance into account by comparing relative abundance values for average values of reference communities. PMA is measured as percentage similarity:

PMA = 100-0,5 ∑│ai + bi│= ∑(ai , bi),

where

ai = relative share (%) of taxa i in reference lake.

bi = share of same taxa in lake under comparison.

Trophic index (RI) originates from reference index developed originally for Bavarian rivers to measure precision of reference status (Schaumburg et al. 2004, Stelzer et al. 2005). Basic principle is division of different taxa according to their indicator value related to sensitivity and tolerance against eutrophication. Method was further developed and modified by Penning et al. (2009a,b) by using Finnish national datasets.

The following description of sensitive, tolerant and indifferent species was applied:

1. Sensitive species: species which are most apparent or only appear in reference lakes, often in high abundance. Frequency and abundance decreases (and often the species disappears entirely) with increased eutrophication pressure; the 75th percentile of observations is chosen as the phosphorous limit above which a

species is no longer considered sensitive. Based on expert judgement this value in 30 μg P/l.

2. Tolerant species: species occurring at an increased frequency and abundance at higher eutrophication pressure. Often rare or with lower abundance in reference lakes; the 75th percentile of observations lies above the upper phosphorous limit and the 25th percentile lies above the lower phosphorous limit. Upper limit is 30 μg P/l and lower limit (15 μg P/l).
3. Indifferent species: species with wide preference, common not only in reference lakes but also in slightly eutrophic lakes; the 75th percentile of observations is more than the upper phosphorous limit and the 25th percentile is less than the lower phosphorous limit. These species disappear in hypertrophic lakes. Upper limit is 30 μg P/l and lower limit (15 μg P/l).

List of species belonging to different groups is presented in Table A.4.

The trophic indices (RI) subtract the number or abundance of tolerant species from the number or abundance of sensitive species either based on the count of species using presence/absence data (Eq. 1):

$$RI = \frac{N_S - N_T}{N} \times 100$$

where NS is the number of Sensitive species found in the lake, NT is the number of Tolerant species and N is the total number of species, including Indifferent species.

Both indices include all aquatic macrophyte life form groups (isoetids, elodeids, nymphaeids, lemnids and charids) and produce a number between +100 and -100 (+100 when all species are defined as Sensitive and -100 when all species are Tolerant) for an individual lake .

Ecological Quality Ratios (EQR) were calculated as a ratio between the observed biological metric value and the reference value (European Communities 2000). The calculated EQR values are divided into classes: high, good, moderate, poor and bad. EQR values were calculated by dividing the observed metric values by the reference values. Developed multimetric index consists of these three metrics (Proportion of type specific taxa, Percent Model Affinity and Reference index).

The upper quartile of each parameter in the reference lakes was used as the type-specific reference value. The lower quartile of the metric values of the reference lakes was utilized as the high/good class boundary. Because EQR values depend on the type-specific reference conditions, the values need to be re-scaled before a comparison of the EQRs over lake types is possible. Linear re-scaling method of Hämäläinen et al. (2007), where the boundary value of high/good condition of the original EQR value is set to 0.8, original value of zero to zero and original value of 1.0 to 1.0.

Then, re-scaled EQRs were calculated with the following formulas:

1. EQRfinal = b1 * EQRoriginal = (0.8/EQRhg) * EQRoriginal, if EQRoriginal < EQRhg, or
2. EQRfinal = a + b2 * EQRoriginal, if EQRoriginal > EQRhg, where

constant *a* and coefficient *b2* are derived by fitting a line connecting the points ($EQR_{original}$, 0.8) and (1,1), thus *b2* = 0.2/(1- EQR_{hg}) and a = *1- b2*. EQR_{hg} refers to EQR boundary value of high/good.

The re-scaled EQRs correspond to ecological status classes as follows: EQR ≥ 0.8 (high), 0.8> EQR ≥0.6 (good), 0.6 > EQR ≥ 0.4 (moderate), 0.4 > EQR ≥ 0.2 (poor), 0.2 > EQR ≥ 0 (bad). Final metric is median value of all metrics and calculated as EQR.

Detailed values for all Finnish lake types are presented in Table A.5.

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Table A.2 Type specific species in different national lake types. Southern Finland types.

South-Finland	**Lake types**										
Type specific species	**Kh**	**Mh**	**MRh**	**MVh**	**Ph**	**Rh**	**RrRk**	**Sh**	**SVh**	**Vh**	**tot.**
Equisetum fluviatile	100	100	100	100	100	100	Not available	100	100	89	10
Lysimachia thyrsiflora	100	67	100	100	100	100		57	88	89	10
Nuphar lutea	100	100	100	100	100	100		100	100	100	10
Phragmites australis	100	100	100	100	100	100		100	100	100	10
Carex rostrata	80	100	100	100	100	100			75	89	9

South-Finland	Lake types										
Type specific species	Kh	Mh	MRh	MVh	Ph	Rh	RrRk	Sh	SVh	Vh	tot.
Eleocharis palustris	100	83	100	100	100			100	88	78	9
Potamogeton natans	100	100		100	100	100		100	88	67	9
Sparganium gramineum	100	100		100	75	100		100	75	56	9
Isoëtes lacustris	100		100	100	75	100		100	75	100	8
Lobelia dortmanna	100		100	100	75	100		100	88	100	8
Alisma plantago-aquatica	60	83			75			86	100	67	7
Carex lasiocarpa	80	83	100	100	100	100				89	7
Myriophyllum alterniflorum	100	67		100	100			86	88	89	7
Potentilla palustris	80			100	100			57	63	78	7
Ranunculus reptans	100			100	75	100		100	88	100	7
Isoëtes echinospora	80		100			100		71	75	67	6
Nymphaea alba ssp. candida		100	100	100	75					78	6
Potamogeton perfoliatus	60	83			75			100	100		6
Schoenoplectus lacustris	80		100		100	100				56	6
Subularia aquatica	80	67			75			100	75	56	6
Eleocharis acicularis	80							100	75	78	5
Juncus bulbosus	60			100	75					56	4
Lythrum salicaria	60							71	63		4
Ranunculus peltatus	60							86	88	56	4
Carex acuta								100	88		3
Lemna minor								71	63		3
Sparganium emersum		83						71			3
Typha latifolia		67		100							3
Utricularia vulgaris			100					57			3
Warnstorfia procera				100		100				56	3
Calla palustris					100					56	2
Carex elata	60								75		2
Elodea canadensis								71	88		2
Fontinalis antipyretica	60					100					2
Glyceria fluitans								86	63		2
Iris pseudacorus				100				57			2
Persicaria amphibia								100	88		2

South-Finland	Lake types										
Type specific species	**Kh**	**Mh**	**MRh**	**MVh**	**Ph**	**Rh**	**RrRk**	**Sh**	**SVh**	**Vh**	**tot.**
Potamogeton berchtoldii		83									2
Sparganium angustifolium				100						56	2
Warnstorfia trichophylla		83		100							2
Alopecurus aequalis								57			1
Calliergon megalophyllum		67									1
Callitriche palustris	60										1
Caltha palustris								57			1
Carex vesicaria								71			1
Drepanocladus longifolius								57			1
Drepanocladus sordidus		83									1
Elatine hydropiper								57			1
Fontinalis hypnoides						100					1
Menyanthes trifoliata				100							1
Nymphaea tetragona			100								1
Phalaris arundinacea								86			1
Potamogeton alpinus		67									1
Potamogeton gramineus	60										1
Potamogeton obtusifolius											1
Sagittaria natans								71			1
Sparganium natans		67									1
Utricularia intermedia			100								1
Utricularia minor				100							1
Number of typesp. species	27	22	15	23	21	16		34	26	25	
Number of ref. lakes	5	6	2	1	4	2	*	7	8	9	

Table A.3 Type specific species in different national lake types. Northern Finland types.

Northern-Finland	Lake type										
Type specific species	**Kh**	**Mh**	**MRh**	**MVh**	**Ph**	**PoLa**	**Rh**	**RrRk**	**Sh**	**SVh**	**Vh**	**tot.**
Carex rostrata	89	91	100	100	100	74	100	100	100	63	75	11
Equisetum fluviatile	89	97	100	83	92	68	100	93	100	88	100	11
Potentilla palustris	89	97	82	83	75	53	100	93	100	63	92	11

Northern-Finland	Lake type											
Type specific species	Kh	Mh	MRh	MVh	Ph	PoLa	Rh	RrRk	Sh	SVh	Vh	tot.
Ranunculus reptans	89	83	91	67	92	68	100		100	75	92	10
Carex aquatilis	67	91	73		67		100	64	100	75	75	9
Isoëtes lacustris	67	60		83	75	58	67		100		92	8
Potamogeton perfoliatus	100	77	91		58		67	86		88	67	8
Isoëtes echinospora	67	71	91	83			67		100	75		7
Menyanthes trifoliata	78	63	64	67	58			64			67	7
Phragmites australis	78	69	82		67		100	86	100			7
Sparganium angustifolium	56	54		83	83	79				63	92	7
Eleocharis acicularis	67	57					67		100	63	75	6
Lysimachia thyrsiflora		74	91		100		100	64	100			6
Myriophyllum alterniflorum	78	57		67		68				100	92	6
Nuphar lutea x pumila	56	66	82	67	58		67					6
Ranunculus peltatus	78	54		67		63				100	92	6
Potamogeton gramineus	67		55						100	63		4
Subularia aquatica	100	60			58					63		4
Caltha palustris	67						67		100			3
Fontinalis antipyretica	56		73				67					3
Nuphar pumila		60	73				67					3
Callitriche palustris			55				100					2

Northern-Finland	Lake type											
Type specific species	Kh	Mh	MRh	MVh	Ph	PoLa	Rh	RrRk	Sh	SVh	Vh	tot.
Hippuris vulgaris	56							57				2
Nuphar lutea			64					79				2
Nymphaea alba ssp. candida			73				67					2
Potamogeton alpinus			64					64				2
Potamogeton berchtoldii	78							71				2
Potamogeton natans			64				67					2
Scorpidium scorpioides										88	58	2
Utricularia vulgaris		51			58							2
Warnstorfia trichophylla	56		55									2
Calliergon megalophyllum			64									1
Carex lasiocarpa				67								1
Drepanocladus longifolius			55									1
Drepanocladus sordidus			64									1
Myriophyllum sibiricum								57				1
Nitella opaca									100			1
Potamogeton praelongus								71				1
Schoenoplectus lacustris							67					1
Sparganium gramineum			55									1
Warnstorfia procera			55									1

Northern-Finland	Lake type											
Type specific species	**Kh**	**M h**	**MR h**	**MV h**	**Ph**	**PoL a**	**Rh**	**RrR k**	**Sh**	**SV h**	**Vh**	**tot .**
Number of typesp. species	22	19	25	12	14	8	19	14	13	14	13	
Number of ref. lakes	9	35	11	6	12	19	3	14	2	8	12	

Table A.4 Response of macrophyte species for eutrophication.

Sensitive species	Tolerant species	Indifferent species
Callitriche hermaphroditica	*Callitriche cophocarpa*	*Callitriche palustris*
Chara aspera	*Ceratophyllum demersum*	*Chara fragilis*
Chara globularis	*Elatine triandra*	*Crassula aquatica*
Elatine hydropiper	*Hydrocharis morsus-ranae*	*Elodea canadensis*
Eleocharis acicularis	*Lemna minor*	*Nuphar lutea*
Isoëtes echinospora	*Lemna trisulca*	*Nuphar pumila*
Isoëtes lacustris	*Myriophyllum verticillatum*	*Nymphaea alba ssp. candida*
Littorella uniflora	*Potamogeton obtusifolius*	*Nymphaea tetragona*
Lobelia dortmanna	*Potamogeton pusillus*	*Persicaria amphibia*
Myriophyllum alterniflorum	*Sagittaria natans x sagittifolia*	*Potamogeton alpinus*
Myriophyllum sibiricum	*Spirodela polyrhiza*	*Potamogeton natans*
Nitella flexilis	*Stratiotes aloides*	*Sagittaria natans*
Nitella opaca		*Sparganium gramineum*
Nuphar lutea x pumila		*Sparganium natans*
Potamogeton berchtoldii		*Utricularia intermedia*
Potamogeton compressus		*Utricularia minor*
Potamogeton filiformis		*Utricularia vulgaris*
Potamogeton gramineus		
Potamogeton perfoliatus		
Potamogeton praelongus		
Ranunculus confervoides		
Ranunculus peltatus		
Ranunculus reptans		
Sparganium angustifolium		
Sparganium hyperboreum		
Subularia aquatica		
Utricularia australis		

Table A.5 Finnish lake types and boundary values for different ecological quality classes.

Type	Proportion of type specific taxa PTST								Percent Model Affinity PMA						Reference Index RI					
	Unit	Ref.lakes	Imp. lakes	Ref. value	H/G	G/M	M/P	P/B	Unit	Ref. lakes	Imp. lakes	Ref. value	H/G	G/M	Unit	Ref. lakes	Imp. lakes	Ref. value	H/G	G/M
Vh-N	PTST	12	2	0,71	0,47	0,35	0,24	0,12	PMA	53,33	41,92	31,44	20,96	10,48	RI	100,00	75,53	56,65	37,77	18,88
Vh-N	EQR				0,67	0,50	0,33	0,17	EQR		0,79	0,59	0,39	0,20	EQR		0,76	0,57	0,38	0,19
Vh-S	PTST	11	1	0,71	0,58	0,44	0,29	0,15	PMA	53,91	52,32	39,24	26,16	13,08	RI	68,75	58,33	43,75	29,17	14,58
Vh-S	EQR				0,82	0,62	0,41	0,21	EQR		0,97	0,73	0,49	0,24	EQR		0,85	0,64	0,42	0,21
Ph-N	PTST	12	7	0,65	0,46	0,34	0,23	0,11	PMA	54,25	45,29	33,97	22,65	11,32	RI	85,71	57,14	42,86	28,57	14,29
Ph-N	EQR				0,71	0,53	0,35	0,18	EQR		0,83	0,63	0,42	0,21	EQR		0,67	0,50	0,33	0,17
Ph-S	PTST	4	7	0,80	0,57	0,42	0,28	0,14	PMA	58,80	49,87	37,40	24,93	12,47	RI	68,18	38,96	29,22	19,48	9,74
Ph-S	EQR				0,70	0,53	0,35	0,18	EQR		0,85	0,64	0,42	0,21	EQR		0,57	0,43	0,29	0,14
Kh-N	PTST	9	2	0,76	0,62	0,47	0,31	0,16	PMA	60,28	47,99	35,99	23,99	12,00	RI	91,67	66,67	50,00	33,33	16,67
Kh-N	EQR				0,81	0,61	0,41	0,20	EQR		0,80	0,60	0,40	0,20	EQR		0,73	0,55	0,36	0,18
Kh-S	PTST	5	6	0,79	0,70	0,53	0,35	0,18	PMA	60,94	57,66	43,24	28,83	14,41	RI	66,67	58,82	44,12	29,41	14,71
Kh-S	EQR				0,88	0,66	0,44	0,22	EQR		0,95	0,71	0,47	0,24	EQR		0,88	0,66	0,44	0,22
SVh-N	PTST	8	0	0,66	0,47	0,35	0,23	0,12	PMA	46,71	36,66	27,49	18,33	9,16	RI	90,00	81,25	60,94	40,63	20,31
SVh-N	EQR				0,70	0,53	0,35	0,18	EQR		0,78	0,59	0,39	0,20	EQR		0,90	0,68	0,45	0,23
SVh-S	PTST	8	15	0,72	0,60	0,45	0,30	0,15	PMA	56,13	53,04	39,78	26,52	13,26	RI	57,49	43,47	32,60	21,74	10,87
SVh-S	EQR				0,84	0,63	0,42	0,21	EQR		0,94	0,71	0,47	0,24	EQR		0,76	0,57	0,38	0,19
Sh-N	PTST	2	5	0,57	0,52	0,39	0,26	0,13	PMA	61,88	61,88	46,41	30,94	15,47	RI	83,46	50,00	37,50	25,00	12,50
Sh-N	EQR				0,92	0,69	0,46	0,23	EQR		1,00	0,75	0,50	0,25	EQR		0,60	0,45	0,30	0,15
Sh-S	PTST	7	14	0,86	0,71	0,53	0,35	0,18	PMA	65,66	51,40	38,55	25,70	12,85	RI	52,27	37,72	28,29	18,86	9,43
Sh-S	EQR				0,82	0,62	0,41	0,21	EQR		0,78	0,59	0,39	0,20	EQR		0,72	0,54	0,36	0,18
Rh-N	PTST	3	2	0,78	0,74	0,55	0,37	0,18	PMA	63,59	62,30	46,72	31,15	15,57	RI	75,00	56,67	42,50	28,33	14,17
Rh-N	EQR				0,95	0,71	0,47	0,24	EQR		0,98	0,73	0,49	0,24	EQR		0,76	0,57	0,38	0,19
Rh-S	PTST	2	2	0,57	0,56	0,42	0,28	0,14	PMA	72,32	72,32	54,24	36,16	18,08	RI	65,91	38,46	28,85	19,23	9,62
Rh-S	EQR				0,98	0,74	0,49	0,25	EQR		1,00	0,75	0,50	0,25	EQR		0,58	0,44	0,29	0,15
MVh-N	PTST	6	1	0,69	0,53	0,40	0,27	0,13	PMA	63,08	55,70	41,77	27,85	13,92	RI	97,22	84,38	63,28	42,19	21,09
MVh-N	EQR				0,78	0,58	0,39	0,19	EQR		0,88	0,66	0,44	0,22	EQR		1,54	1,16	0,77	0,39
MVh-S	PTST	1	3	1,00	1,00	0,75	0,50	0,25	PMA	100,00	100,00	75,00	50,00	25,00	RI	50,00	50,00	37,50	25,00	12,50
MVh-S	EQR				1,00	0,75	0,50	0,25	EQR		1,00	0,75	0,50	0,25	EQR		1,00	0,75	0,50	0,25
Mh-N	PTST	35	13	0,71	0,46	0,34	0,23	0,11	PMA	47,72	34,94	26,21	17,47	8,74	RI	81,67	58,33	43,75	29,17	14,58
Mh-N	EQR				0,64	0,48	0,32	0,16	EQR		0,73	0,55	0,37	0,18	EQR		0,71	0,54	0,36	0,18
Mh-S	PTST	6	7	0,59	0,49	0,37	0,25	0,12	PMA	50,97	47,27	35,45	23,63	11,82	RI	47,79	6,25	4,69	3,13	1,56
Mh-S	EQR				0,83	0,63	0,42	0,21	EQR		0,93	0,70	0,46	0,23	EQR		0,13	0,10	0,07	0,03
MRh-N	PTST	11	15	0,72	0,64	0,48	0,32	0,16	PMA	50,75	39,31	29,49	19,66	9,83	RI	69,62	41,88	31,41	20,94	10,47
MRh-N	EQR				0,89	0,67	0,44	0,22	EQR		0,77	0,58	0,39	0,19	EQR		0,60	0,45	0,30	0,15
MRh-S	PTST	2	11	0,58	0,55	0,41	0,28	0,14	PMA	62,47	62,47	46,85	31,23	15,62	RI	37,69	33,08	24,81	16,54	8,27
MRh-S	EQR				0,94	0,71	0,47	0,24	EQR		1,00	0,75	0,50	0,25	EQR		0,88	0,66	0,44	0,22

Type	Proportion of type specific taxa PTST								Percent Model Affinity PMA						Reference Index RI					
	Unit	Ref.lakes	Imp. lakes	Ref. value	H/G	G/M	M/P	P/B	Unit	Ref. lakes	Imp. lakes	Ref. value	H/G	G/M	Unit	Ref. lakes	Imp. lakes	Ref. value	H/G	G/M
PoLa	PTST	19	0	0,56	0,31	0,24	0,16	0,08	PMA	45,34	32,60	24,45	16,30	8,15	RI	100,00	84,52	63,39	42,26	21,13
PoLa	EQR				0,56	0,42	0,28	0,14	EQR		0,72	0,54	0,36	0,18	EQR		0,85	0,63	0,42	0,21
RkRr-N	PTST	14	1	0,59	0,33	0,25	0,16	0,08	PMA	46,12	38,54	28,90	19,27	9,63	RI	69,05	40,83	30,63	20,42	10,21
RkRr-N	EQR				0,56	0,42	0,28	0,14	EQR		0,84	0,63	0,42	0,21	EQR		0,59	0,44	0,30	0,15
RkRr-S	PTST	0	17						PMA						RI					
RkRr-S	EQR								EQR						EQR					

N = Northern Finland
S = Southern Finland
Not reliable
No reference lakes, not used

A.2 Ireland

Description of Free Macrophyte Index

The following describes the Free Macrophyte Index. It was developed to cover all types of Irish lakes and therefore is not type specific.

There are 6 components to the Macrophyte Index (Free *et al.*, 2006):

1. %RF (percentage relative frequency) Chara;
2. %RF (percentage relative frequency) Elodeids;
3. Plant trophic score;
4. %RF (percentage relative frequency) Tolerant taxa;
5. Zc – maximum depth of colonisation;
6. Mean depth of presence;

There are conditions for assigning values for some of the metrics (see relevant sections). Only the submerged and floating taxa listed in Table A.6 (Palmer *et al.* 1992) were utilised in devising the index and calculating metrics. This list is not exclusive, any submerged and floating taxa encountered should be included in the analysis (see Table A.6). The Index is dependent on expressing data as percentage relative frequency. Therefore, transect data is required.

Calculating % Relative Frequency (%RF)

1. Use only taxa identified as submerged or floating according to Palmer et al. (1992) in the calculation, see Table A.6. This list is not exclusive.
2. Sum occurrence of each taxon e.g. Chara spp., occurred at 10 transect points, occurrence =10
3. Sum occurrences of all taxa e.g. Chara =10, Sp A= 20, Sp B=5 therefore total =35 (occurrence of all taxa)
4. Relative Frequency for Chara spp. = (10/35)*100=29%

Maximum transect depth

The maximum transect depth must be determined first before scores can be assigned to Zc – Maximum Depth of Colonisation - and mean depth of presence. As it suggests, the maximum transect depth is the maximum depth recorded regardless of whether macrophytes were present or not.

Metric Descriptions

%RF Chara

This is the sum of the % RF of all *Chara* spp. This metric is only included for lakes with an alkalinity of 100mg l^{-1} $CaCO_3$ or greater.

%RF Elodeids

This is the sum of the % RF of all elodeid like plants. This metric will only add to a 100% if elodeids are the only plants present. Elodeids (functional form – resembles *Elodea* spp. as opposed to rosette or isoetid forms which resemble *Isoetes* sp.) were defined according to the list in Jensen (1979) which is not exclusive (Table A.7). If a new taxa is encountered with an elodeid form (extends into the water column) then it is included in the calculation.

Plant trophic score

Plant trophic scores are presented in Table A.8. The Plant trophic score was calculated based on the list of submerged and floating taxa listed in Table A.8 that were present in a lake i.e. on a lake basis not for records on a transect basis. The scores were summed and the average calculated. Note: that other moss and Filamentous algae were scored, these are not listed in Palmer e*t al.* (1992) - see Macrophyte chapter in Free *et al.* (2007) for development of the Plant Trophic Score.

1. Assign relevant scores to taxa
2. average trophic scores

%RF Tolerant taxa

Tolerant taxa are also listed in Table A.8. They are any taxa that had a TP score > 25 µg l^{-1} (see Macrophyte chapter in Free *et al.* (2007) for development). Percentage relative frequency (%RF) is the sum of their relative frequencies and only adds up to a 100% if they are the only taxa present. *(Note: this highlights the importance of identification to species in the field).*

Zc – Maximum Depth of Colonisation

As it suggests, Maximum Depth of Colonisation is the maximum depth at which plants (Floating and submerged taxa only) were recorded. A score is not assigned where Zc is less than 3 m <u>and</u> is between 80 (i.e. >80%) and 100% of the maximum transect depth recorded. This is done to prevent a low score being assigned to shallow lakes. See examples in Table A.9.

Average depth of presence

Average depth of presence is the average of all the depths recorded for which plants (floating and submerged only) were present. A score is not assigned for the average depth of presence if it is less than 1.8 m <u>and</u> it is within 50% of the maximum transect depth. This is done to prevent a low score being assigned to shallow lakes. See examples in Table A.10.

Macrophyte Index

Each of the above metrics were scaled from 0.1 to 1. Scores are assigned based on the metric value according to Table A.11. The average of the assigned metric scores is the Index value.

Table A.6 The list of submerged and floating taxa from Palmer et al. (1992). Taxa not encountered during Irish Lake Surveys are highlighted in bold. Taxa used in the calculation of %RF for Irish surveys are also listed. This list shows additional taxa not listed in Palmer.

Palmer's taxa	Taxa used in calc of %RF for Irish Surveys
Apium inundatum	*Apium inundatum*
Callitriche hamulata	*Callitriche hamulata*
Callitriche hermaphrodita	*Callitriche hermaphrodita*
	Callitriche sp.
Callitriche obtusangula	
Callitriche stagnalis	
Ceratophyllum demersum	*Ceratophyllum demersum*
	Ceratophyllum submersum
Chara sp.	*Chara sp.*
Elatine hexandra	*Elatine hexandra*
Eleocharis acicularis	
Elodea canadensis	*Elodea canadensis*
Elodea nuttallii	
	Eriocaulon septangulare
	filamentous algae
Fontinalis antipyretica	*Fontinalis antipyretica*
Glyceria fluitans	
Hippuris vulgaris	*Hippuris vulgaris*
	Hydrocharis morsus-ranae
	Isoetes echinospora
Isoetes lacustris	*Isoetes lacustris*
Juncus bulbosus	*Juncus bulbosus*
	Lemna gibba
Lemna minor	*Lemna minor*
	Lemna polyrrhiza
Lemna trisulca	*Lemna trisulca*
Litorella uniflora	*Litorella uniflora*
Lobelia dortmanna	*Lobelia dortmanna*
Myriophyllum alterniflorum	*Myriophyllum alterniflorum*
Myriophyllum spicatum	*Myriophyllum spicatum*
	Myriophyllum verticillatum
	Najas flexilis
Nitella sp.	*Nitella sp.*
Nuphar lutea	*Nuphar lutea*

Palmer's taxa	Taxa used in calc of %RF for Irish Surveys
Nuphar pumila	
Nymphea alba	*Nymphea alba*
	Other Moss
Oenanthe aquatica	
Polygonum amphibium	*Polygonum amphibium*
Potamogeton alpinus	*Potamogeton alpinus*
Potamogeton berchtoldii	*Potamogeton berchtoldii*
Potamogeton crispus	*Potamogeton crispus*
Potamogeton filiformis	*Potamogeton filiformis*
Potamogeton friessi	*Potamogeton friessi*
Potamogeton gramineus	*Potamogeton gramineus*
Potamogeton lucens	*Potamogeton lucens*
Potamogeton natans	*Potamogeton natans*
Potamogeton obtusifolius	
Potamogeton pectinatus	*Potamogeton obtusifolius*
Potamogeton perfoliatus	*Potamogeton pectinatus*
Potamogeton polygonifolius	*Potamogeton perfoliatus*
	Potamogeton polygonifolius
Potamogeton praelongus	
Potamogeton pusillus	*Potamogeton pusillus*
	Potamogeton sp
	Potamogeton x nitens
	Potamogeton zizii
Potamogeton trichoides	
Ranunculus aquatilis	
Ranunculus baudotii	
Ranunculus circinatus	*Ranunculus circinatus*
Ranunculus hederaceus	
Ranunculus peltatus	
Ranunculus trichophylus	
Scirpus fluitans	
	Ranunculus penicillatus var penicillatus
	Sagittaria sp
Sparganium angustifolium	*Sparganium angustifolium*
Sparganium emersum	*Sparganium emersum*
Sparganium minimum	*Sparganium minimum*
Sphagnum sp	*Sphagnum sp*
Subularia aquatica	*Subularia aquatica*
	Unidentified submergent

Palmer's taxa	Taxa used in calc of %RF for Irish Surveys
Utricularia intermedia	*Utricularia intermedia*
	Utricularia sp.
Utricularia minor	
Utricularia vulgaris	*Utricularia vulgaris*
Zannichellia	*Zannichellia*

Table A.7 List of elodeid forms after Jensen (1979) with taxa selected from Palmer et al. (1992) with an elodeid form. Bolded taxa were not found in the 2000-2003 Irish Lakes surveys. The list is not exclusive – any elodeid form taxa encountered should be included.

Taxa	Jensen	Palmer *et al.* (2001)
Apium inundatum	1	
Apium nodiflorum		1
Ceratophyllum demersum	1	
Ceratophyllum submersum		1
Elodea canadensis	1	
Juncus fluvitans	1	
Juncus sp.		1
Myriophyllum alterniflorum	1	
Myriophyllum spicatum	1	
Myriophyllum verticillatum		1
Nitella sp.	1	
Potamogeton crispus	1	
Potamogeton fresii	1	
Potamogeton gramineus		1
Potamogeton lucens		1
Potamogeton obtusifolius		1
Potamogeton pectinatus	1	
Potamogeton perfoliatus	1	
Potamogeton polygonifolius		1
Potamogeton sp		1
Potamogeton x nitens		1
Ranunculus circinatus	1	
Ranunculus penicillatus var penicillatus		1
Ranunculus sp.		1
Ranunculus sceleratus		1
Utricularia intermedia	1	
Utricularia sp.		1
Utricularia vulgaris	1	

Table A.8 Weighted spring TP (µg l-1) where taxa present. The score was calculated using data from all lakes (n = 159). The highlighted taxa were not listed in Palmer et al. (1992). After Free et al. (2005).

Taxa	Weighted TP of lakes where taxa present (Scores)	n	Tolerant taxa
Ranunculus penicillatus var penicillatus	7	5	
Utricularia intermedia	7	5	
Lobelia dortmanna	10	47	
Eriocaulon septangulare	11	26	
Isoetes lacustris	12	55	
Juncus bulbosus	15	55	
Elatine hexandra	15	22	
Sphagnum sp	17	8	
Myriophyllum alterniflorum	17	46	
Hippuris vulgaris	20	12	
Nitella sp.	20	62	
Nymphea alba	21	19	
Utricularia vulgaris	21	25	
Sagittaria sp	22	13	
Chara sp.	23	70	
Other Moss	23	31	
Potamogeton gramineus	23	16	
Fontinalis antipyretica	26	78	T
Potamogeton perfoliatus	28	43	T
Potamogeton pectinatus	31	17	T
Lemna trisulca	31	35	T
Myriophyllum spicatum	32	27	T
Litorella uniflora	34	109	T
Potamogeton natans	34	51	T
Callitriche hamulata	34	6	T
Potamogeton lucens	35	32	T
Potamogeton berchtoldii	37	34	T
Filamentous algae	39	96	T
Sparganium emersum	40	46	T
Nuphar lutea	43	66	T
Elodea canadensis	48	62	T
Potamogeton obtusifolius	54	14	T
Potamogeton crispus	59	10	T
Ceratophyllum demersum	62	9	T
Polygonum amphibium	67	12	T

Callitriche sp.	68	13	T
Lemna minor	88	11	T
Lemna polyrrhiza	145	5	T

*Table A.9 Examples of how to apply scoring restrictions to Zc. % is the Zc/max. transect depth*100.*

Z_C	Max. transect depth	%	Score??	Score
1.8	2	90.00	no score	
2.6	3.2	81.25	no score	
2.6	2.8	92.86	no score	
2.9	4	72.50	score	0.7
2.9	3.5	82.86	no score	
2.9	3	96.67	no score	
2.9	3.7	78.38	score	0.7
3	3	100.00	score	0.7
3	6	50.00	score	0.7
3	6	50.00	score	0.7
4	6	66.67	score	0.8
5	6	83.33	score	0.9

*Table A.10 Examples of how to apply scoring restrictions to average depth of presence. % is the average depth of presence/max. transect depth*100.*

average deoth of presence	Max transect depth (m)	%	Z presence score
0.6	1	60.00	no score
0.7	0.9	77.78	no score
0.9	1.8	50.00	0.3
1	1.7	58.82	no score
1.1	1.1	100.00	no score
1.1	1.9	57.89	no score
1.1	1.8	61.11	no score
1.1	2.1	52.38	no score
1.2	2	60.00	no score
1.3	2.2	59.09	no score
1.3	2.5	52.00	no score
1.5	1.6	93.71	no score
1.6	2.9	55.17	no score
1.7	2.8	60.71	no score
1.8	12.8	14.06	0.9
1.8	5.4	33.33	0.9

1.9	2.8	67.86	0.9
1.9	2.3	82.61	0.9
2	3.2	62.5	0.9

Table A.11 Table of scaled deciles for five metrics that had a log-linear response to spring TP. After Free et al. (2007).

Scaled deciles	Plant trophic score	Zc	Mean depth of presence	RF% Elodeids (functional group)	RF% Chara	RF% Tolerant
1.0	<28.2	>5.1	>2.00	<19	>67	<26
0.9	28.2 - 30.4	5.1 - 4.1	2.00 - 1.66	19 - 31	67 - 61	26.0 - 37.9
0.8	30.4 - 31.8	4.1 - 3.5	1.66 - 1.49	31 - 37	61 - 45	37.9 - 51.7
0.7	31.8 - 33.1	3.5 - 2.9	1.49 - 1.35	37 - 48	45 - 29	51.7 - 60.4
0.6	33.1 - 34.0	2.9 - 2.5	1.35 - 1.25	48 - 53	29 - 23	60.4 - 70.1
0.5	34.0 - 35.2	2.5 - 2.1	1.25 - 1.13	53 - 59	23 - 10	70.1 - 77.9
0.4	35.2 - 38.2	2.1 - 1.8	1.13 - 0.94	59 - 65	10 - 7	77.9 - 84.8
0.3	38.2 - 40.2	1.8 - 1.6	0.94 - 0.81	65 - 75	7 - 5	84.8 - 90.0
0.2	40.2 - 43.7	1.6 - 1.0	0.81 - 0.30	75 - 80	5 - 2	90.0 - 98.9
0.1	>43.7	<1.0	<0.30	>80	<2	>98.9

A.3 Description of boundary setting procedure for Irish Macrophyte methods

The purpose of this study was to set boundaries of high/good and good/moderate status that are of ecological relevance. This was attempted by:

1. Examining published relationships to find criteria that match normative definitions and to define these in terms of TP.
2. To see if the selected TP boundaries are supported by an examination of data from the ROI in the Atlantic GIG typology > 50 mg l-1 CaCO3 alkalinity and 3-15 m mean depth. Marl lakes were excluded from the analysis in order to improve compatibility of the lake type across the region being intercalibrated.

Methods

A series of regression models (Table A.12), initially based on Spring TP, were used to successively predict summer chlorophyll *a* (Dillon & Rigler, 1974), Secchi depth (Free, 2002), depth of colonisation of Charophytes (Blindow, 1992) and depth of colonisation of Angiosperms (Chambers and Kalff, 1985). The prediction of Secchi depth used multiple regression based on predicted chlorophyll *a* and a colour of 30 mg l^{-1} PtCo.

Metrics were examined for potential relationships with TP. This was based on a survey carried out in the Republic of Ireland between 2001 and 2003. Nineteen lakes were selected that were in the Atlantic GIG typology > 50 mg l^{-1} $CaCO_3$ alkalinity and of 3-15 m mean depth.

For the composition metrics the ratio of Littorella to other littoral rosette species (Lobelia and Eriocaulon) was developed with the aim of detecting pollution in low alkalinity lakes as Litorella has a competitive advantage on more nutrient rich sediment (Farmer & Spence, 1986).

Results and Discussion

Figure A.1 shows the predicted relationships between TP (as a pressure gradient) and chlorophyll *a*, Secchi depth, depth of colonisation of Charophytes and the depth of colonisation of Angiosperms. The predictions provide a literature-based example of the interactions between a pressure gradient and ecological quality. As chlorophyll *a* increases with TP it leads to a rapid decrease in Secchi depth (transparency) which reduces the depth of colonisation of Charophytes and Angiosperms. As the extinction of light is exponential with depth, the initial change from an oligotrophic state to a mesotrophic state is where the most change takes place.

The high/good boundary was placed at 10 $\mu g\ l^{-1}$ TP as this is where there appears to be a significant change in slope/response of the depth of macrophyte colonisation to TP concentration (Figure A.1).

The good/moderate boundary was placed at 25 $\mu g\ l^{-1}$ TP as this is where the depth of colonisation of the Charophytes is reduced by 24% from reference condition. This appears to fit normative definitions (Table A.14) where phytoplankton biomass is such as to produce a significant undesirable disturbance in the condition of another biological quality element. The depth of colonisation of angiosperms is less useful in this regard as a reduction in transparency may be accompanied by a shift to taller growing species such as *Potamogeton lucens*.

Ecological data from 19 lakes in the ROI that fit the Atlantic GIG typology (> 50 mg l^{-1} $CaCO_3$ alkalinity and 3-15 m mean depth) were examined to see if the boundaries were relevant. Figure A.2 shows the relationship between four macrophyte metrics and TP (measured in Spring or early Summer). The lakes of presumed high status (< 10 $\mu g\ l^{-1}$ TP) appeared distinct in that they had a deeper depth of colonisation of Charophytes and a low to high species richness (species richness typically having a unimodal relationship with TP). In good status (10-25 $\mu g\ l^{-1}$ TP) species richness reaches a maximum, which may conform to normative definitions in that it is a 'slight' change but one that is not 'undesirable'. At TP concentrations > 25 $\mu g\ l^{-1}$ species richness declines, lakes may have fewer littoral rosette species, the depth of colonisation of Charophytes decreases and the relative frequency of canopy forming species increases.

These changes, especially the decline in species richness appear to match normative definitions for moderate status (Table A.14) where phytoplankton biomass issuch as to produce a significant undesirable disturbance in the condition of another biological quality element.

A draft macrophyte multimetric has been developed in the ROI and is applied by averaging the scaled deciles for six metrics (Table A.13). The macrophyte multimetric shows a linear response to TP (Figure A.3), and although linear relationships may not present clear 'break-points' that might suggest an appropriate point to position a boundary, they do demonstrate that ecological change is clearly taking place across the chosen pressure gradient – total phosphorus.

Ideally, selected boundaries of TP would also be supported by information from the other biological elements required to be monitored by the WFD. Table A.15 shows that the proportion of the generally regarded pollution 'tolerant' genus *Chironomus* increases at TP concentrations > 25 μg l^{-1}. This appears to meet the normative definition in annex 5 of moderate status for benthic invertebrate fauna: "Major taxonomic groups of the type specific community are absent. The ratio of disturbance sensitive to insensitive taxa, and the level of diversity, are substantially lower than the type-specific level and significantly lower than for good status."

Table A.15 summarises the selected TP boundaries of high, good and moderate status and associated ecological changes. This is largely a fixed boundary system the older proposals of the OECD (OECD, 1982). The WFD marks a departure in that a state-change system is favoured. A state-change system recognises is natural variability in lakes. For example, there may be a natural range in concentrations of 5-10 μg l^{-1} for a lake type. While it is accepted that there is natural variation in reference condition, the methods for determining what the variation is, especially in the absence of present day examples of reference not well defined. It may be possible that the variation, at least in background may be sufficiently low that a fixed boundary system may be the most useful. usefulness of the morphoedaphic index (MEI) (Vighi & Chiaudani, 1985) in reference TP concentration may be limited, at least in Ireland.

Table A.16 shows that predicted reference annual TP concentrations were about 10 μg l^{-1} <u>higher</u> than concentrations measured in Spring and Summer in <u>reference</u> lakes.

Ideally, the mean TP within a large population of existing reference lakes would be used used to estimate reference TP concentration. Only three lakes were regarded in potential reference condition: Lough Glencar, Talt and Kindrum (

Table A.16). Although three lakes may be insufficient to determine a type specific reference TP concentration the clear relationship between the macrophyte multimetric and TP (Figure A.3) may partly validate the reference lake selection and provide some confidence in determining that the reference TP is more than likely to be below 10 μg l^{-1} for this type. A palaeolimnological project is currently attempting to validate the reference lake selection.

In conclusion, the proposed boundaries of TP appeared to have broad ecological support. Published models indicated that ecological change was likely to be most dramatic between 0 and 25 μg l^{-1} TP (Figure A.1). This was found to be supported by recent biological surveys within the AGIG type lakes in the Republic of Ireland. Biological metrics appeared to support our reference lake selection. Reference TP concentrations for this type are likely to be below 10 μg l^{-1}.

Table A.12 Models used to generate Figure A.1. Sources: 1: Equation 2 Dillon and Rigler (1974), 2: Free 2002, 3 Equation 4 Chambers and Kalff (1985), 4: Blindow 1992. A colour value of 30 mg l^{-1} PtCo was used.

Source	Dependent variable	r^2	Model
1	Log chlorophyll a ・g l-1	0.92	1.449 log TP ・g l-1 - 1.136
2	Log 1+Secchi depth (m)	0.82	1.34495 -0.414109 log (x + 1) colour -0.205299 log (x + 1) chlorophyll a ・g l-1
3	Zc Angiosperms0.5		1.33 log Secchi depth + 1.4

4	Log Zc Charophyta	0.83	1.03 log Secchi depth + 0.18

Table A.13 Table of scaled deciles for six metrics that were averaged to give the macrophyte multimetric index.

Scaled deciles	Plant trophic score	Zc	Mean depth of presence	RF% Elodeids (functional group)	RF% Chara	RF% Tolerant
1.0	<28.2	>5.1	>2.00	<19	>67	<26
0.9	28.2 - 30.4	5.1 - 4.1	2.00 - 1.66	19 - 31	67 - 61	26.0 - 37.9
0.8	30.4 - 31.8	4.1 - 3.5	1.66 - 1.49	31 - 37	61 - 45	37.9 - 51.7
0.7	31.8 - 33.1	3.5 - 2.9	1.49 - 1.35	37 - 48	45 - 29	51.7 - 60.4
0.6	33.1 - 34.0	2.9 - 2.5	1.35 - 1.25	48 - 53	29 - 23	60.4 - 70.1
0.5	34.0 - 35.2	2.5 - 2.1	1.25 - 1.13	53 - 59	23 - 10	70.1 - 77.9
0.4	35.2 - 38.2	2.1 - 1.8	1.13 - 0.94	59 - 65	10 - 7	77.9 - 84.8
0.3	38.2 - 40.2	1.8 - 1.6	0.94 - 0.81	65 - 75	7 - 5	84.8 - 90.0
0.2	40.2 - 43.7	1.6 - 1.0	0.81 - 0.30	75 - 80	5 - 2	90.0 - 98.9
0.1	>43.7	<1.0	<0.30	>80	<2	>98.9

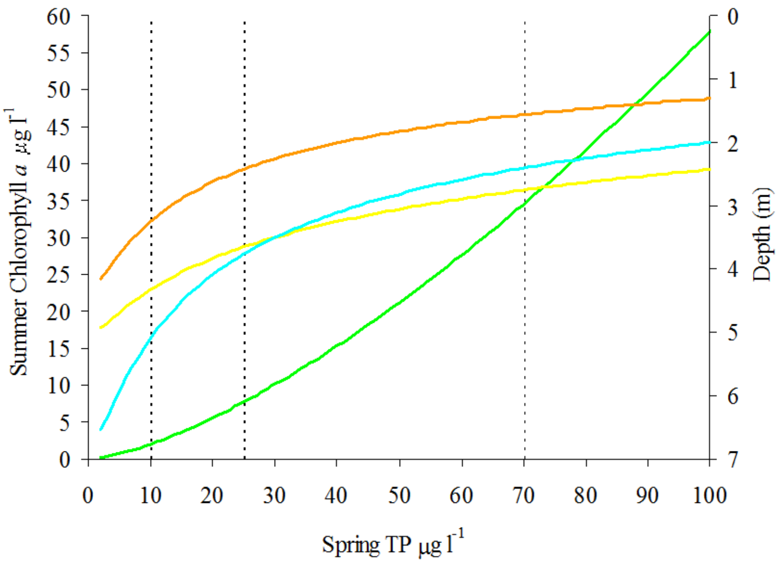

Figure A.1 Relationship between Spring TP and Summer chlorophyll a (—), and predicted Secchi depth (—), predicted depth of colonisation of Charophytes (—) and Angiosperms (—). Sources and models are listed in Table A.12. Dashed lines represent proposed boundaries of 10, 25 and 70 μg l^{-1} TP.

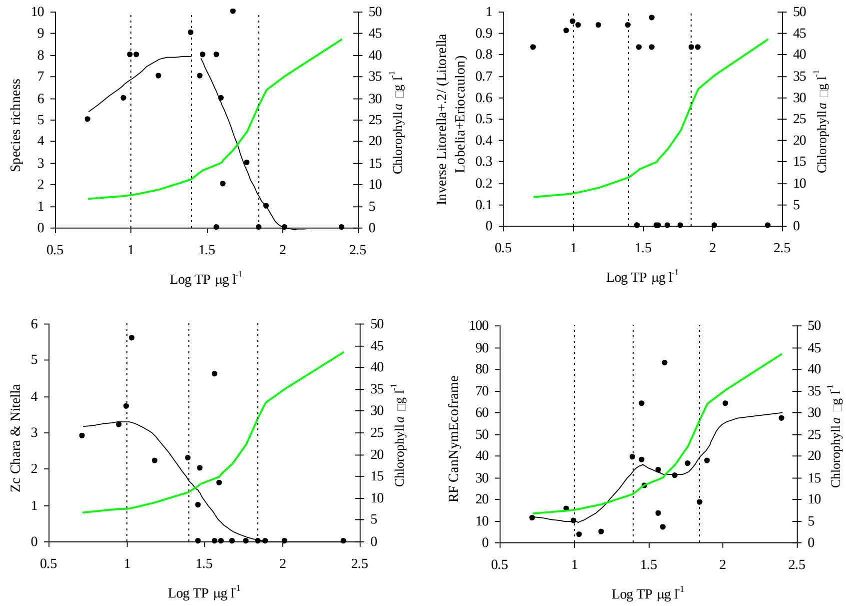

Figure A.2 *Relationship between TP (Spring or early Summer) and species richness, inverse Littorella +0.2/ (Littorella + Lobelia + Eriocaulon), depth of colonisation (Zc) of Chara & Nitella, RF of CanNymEcoframe (canopy forming species – Moss et al. (2003)). The lowess smoothed relationship between TP and chlorophyll a is overlain (▬). n = 19, lakes between 50 and 100 mg l^{-1} $CaCO_3$, 3-15 m mean depth and non-marl precipitating. Dashed lines represent proposed boundaries of 10, 25 and 70 µg l^{-1} TP.*

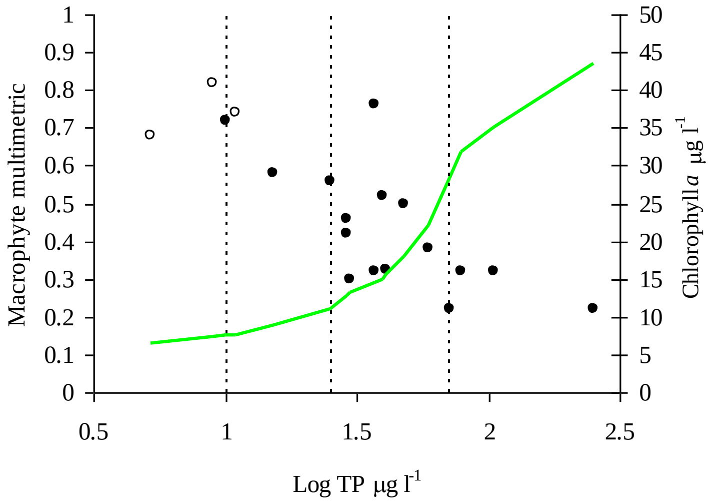

Figure A.3 *The relationship between TP (Spring or early Summer) and the macrophyte multimetric. The lowess smoothed relationship between TP and chlorophyll a is overlain (▬). n = 19, lakes between 50 and 100 mg l^{-1} $CaCO_3$, 3-15 m mean depth and non-marl precipitating. Dashed lines represent proposed boundaries of 10, 25 and 70 µg l^{-1} TP. ○ = lakes of potential reference condition: Lough Glencar, Talt and Kindrum.*

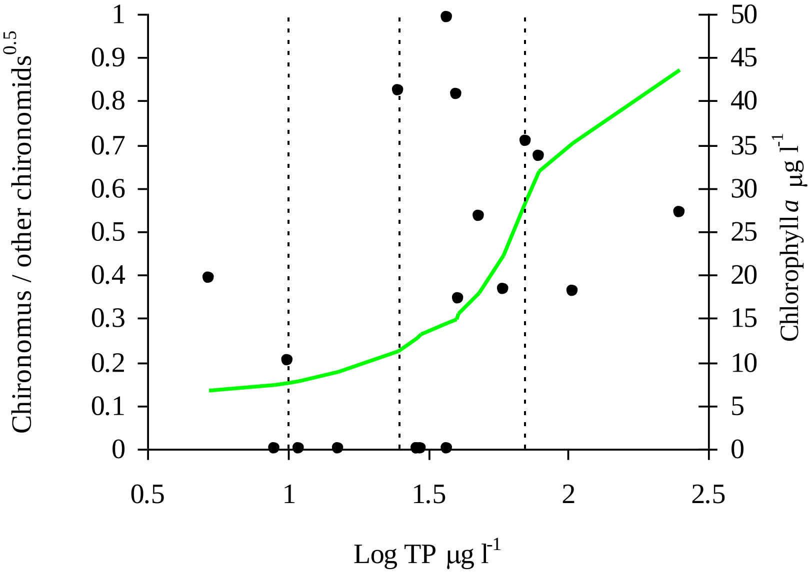

Figure A.4 The relationship between TP (Spring or early Summer) and the transformed (square root) ratio of Chironomus spp. to other chironomids . The lowess smoothed relationship between TP and chlorophyll a is overlain (——). n = 19, lakes between 50 and 100 mg l-1 CaCO3, 3-15 m mean depth and non-marl precipitating. Dashed lines represent proposed boundaries of 10, 25 and 70 • g l-1 TP.

Table A.14 Normative definitions for high, good and moderate ecological status of phytoplankton, macrophytes and phytobenthos in lakes.

Element	High status	Good status	Moderate status
Phytoplankton	The taxonomic composition and abundance of phytoplankton correspond totally or nearly totally to undisturbed conditions. The average phytoplankton biomass is consistent with the type-specific physico-chemical conditions and is not such as to significantly alter the type-specific transparency conditions. Planktonic blooms occur at a frequency and intensity which is consistent with the type specific physico-chemical conditions.	There are slight changes in the composition and abundance of planktonic taxa compared to the type-specific communities. Such changes do not indicate any accelerated growth of algae resulting in undesirable disturbance to the balance of organisms present in the water body or to the physico-chemical quality of the water or sediment. A slight increase in the frequency and intensity of the type specific planktonic blooms may occur.	The composition and abundance of planktonic taxa differ moderately from the type-specific communities. Biomass is moderately disturbed and may be such as to produce a significant undesirable disturbance in the condition of other biological quality elements and the physico-chemical quality of the water or sediment. A moderate increase in the frequency and intensity of planktonic blooms may occur. Persistent blooms may occur during summer months.
Macrophytes and phytobenthos	The taxonomic composition corresponds totally or nearly totally to undisturbed conditions. There are no detectable changes in the average macrophytic and the average phytobenthic abundance.	There are slight changes in the composition and abundance of macrophytic and phytobenthic taxa compared to the type-specific communities. Such changes do not indicate any accelerated growth of phytobenthos or higher forms of plant life resulting in undesirable disturbance to the balance of organisms present in the water body or to the physico-chemical quality of the water. The phytobenthic community is not adversely affected by bacterial tufts and coats present due to anthropogenic activity.	The composition of macrophytic and phytobenthic taxa differ moderately from the type-specific communities and are significantly more distorted than those observed at good quality. Moderate changes in the average macrophytic and the average phytobenthic abundance are evident. The phytobenthic community may be interfered with, and, in some areas, displaced by bacterial tufts and coats present as a result of anthropogenic activities.

Table A.15 Summary of TP bands defining high, good and moderate status and associated ecological changes.

	High status	**Good status**	**Moderate status**
TP	< 10 μg l^{-1}	10-25 μg l^{-1}	25-70 μg l^{-1}
Summer Chlorophyll *a**	< 2 μg l^{-1}	< 2-8 μg l^{-1}	8 - 35 μg l^{-1}
Zc Charophytes From Figure A.1	> 5 m	5 m – 3.8 m	< 3.8 m A 24% reduction in the depth of colonisation found in reference condition.
Zc Angiosperms From Figure A.1	> 4.3 m	4.3 m – 3.7 m	< 3.7 A smaller reduction in the depth of colonisation can be caused by a succession of taller taxa.
Species richness	Tends to be naturally low but variable	Tends to be at a maximum	Declines markedly in moderate status.
Littoral rosette species	Present	Present	May only be present in the shallowest areas of the littoral
Canopy forming taxa	Infrequent	Infrequent	More frequent
Ratio *Chironomus spp.* to other chironomids	Low	Low	High

* Predicted from equation 2: Dillon and Rigler (1972), figures may be higher for a methanol extraction. Zc predictions are for 30 mg l^{-1} PtCo colour.

Table A.16 Summary characteristics and chemistry of the lakes. MEI predicted reference TP calculated using equation 1 from Vighi & Chiaudani (1985).

Lake	Altitude m	Lake area (km2)	predicted mean depth	Alkalinity mg l^{-1} $CaCO_3$	MEI	MEI predicted ref TP	TP ug l^{-1} (Spring)	TP ug l^{-1} (Summer)	Chlorophyll *a* ug l^{-1} (Summer)	Ref lakes
Glencar	28	1.15	8.5	94	0.22	18	5	<10	8	Yes
Talt	130	0.97	8.8	85	0.19	18	9	5	5	Yes
Rowan	73	0.48	5.0	56	0.22	18	10	23	7	
Kindrum	8	0.61	4.7	69	0.29	20	11	11	10	Yes
Melvin	25	22.06	4.7	54	0.23	19	15	10	4	
Drumlaheer	65	0.74	5.9	71	0.24	19	25	56	17	
Corry	41	1.54	3.8	51	0.27	20	29	37	17	
Alewnaghta	31	0.55	3.1	70	0.45	23	29	10	6	
Garadice	49	3.89	3.9	75	0.38	22	30	18	5	
Glasshouse	48	0.54	6.5	52	0.16	16	37	49	27	
Aughrusbeg	8	0.50	4.3	54	0.25	19	37	17	17	
Derryhick	25	0.54	4.9	84	0.34	21	40	21	18	
Derrycassa	45	0.71	3.3	80	0.49	24	41	61	11	
Doon	22	0.49	4.9	82	0.33	21	48	27	13	
Scur	62	1.14	3.4	60	0.35	21	59	71	15	
Muckno L	86	3.57	5.1	62	0.24	19	71	65	21	
Drumlona	77	0.53	5.0	99	0.40	22	79	114	40	
White	75	0.54	4.8	85	0.35	21	105	156	44	
Dromore	79	0.61	5.1	90	0.35	21	250	91	39	

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A.4 Norwegian macrophyte assessment method for lakes: Trophic index (TIc)

Status

The Norwegian national method includes a species composition index (TIc). The method has been designed to detect the impact from eutrophication on aquatic macrophytes, and can be applied to boreal and lowland freshwater lakes in Norway. An abundance index (Cmax) is not developed.

The method is discussed and supported by the Environmental Authorities, but not yet formally agreed.

Sampling and analysis

Sampling method

Each lake is visited once between July and September. The survey extends from the shore to the maximum depth of colonisation of macrophytes, at different localities/habitats spread around the lake.

The plants are recorded using an aqua scope and collected by dredging from a boat. The lakes are surveyed in order to establish the presence of each of the macrophyte taxa listed in Table A.17. The abundance of the species is scored by a semi-quantitative scale, where 1 = rare, 2 = scattered, 3 = common, 4 = locally dominant and 5 = dominant. The macrophytes have to be identified to species level.

All true aquatic macrophytes, belonging to the life form groups isoetids, elodeids, nymphaeids, lemnids and charophytes, are included. Some macrophyte species can occur in both helophyte and true aquatic forms, e.g. *Juncus bulbosus, Sagittaria sagittifolia* and *Hippuris vulgaris*. These are also included in the analyses. Helophytes (emergent aquatic plants) are excluded completely from the analyses, so are water mosses and filamentous algae.

Index description

The index is based on the relationship between number of sensitive, tolerant and indifferent species in the lake (see Table A.17). Presence/absence data only is sufficient:

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where N_S is the number of Sensitive species found in the lake, N_T is the number of Tolerant species, and N is the total number of species, including Indifferent species.

The index produces a number between +100, where all species are defined as sensitive, and -100, where all species are tolerant, and give one value for each lake.

Calculations

Reference lakes and boundaries

The set of reference lakes were derived using a combination of water chemistry data, information about catchment and lake use, and expert judgement.

The average of the index value for reference lakes is chosen as the reference value, while the lower quartile of the index value for the reference lakes represents the high/good boundary. The reference value and the high/good value vary with lake types.

For setting the boundaries for G/M, M/P and P/B, we have used the appearance and abundance of the large isoetids *Isoetes lacustris, I. echinospora, Littorella uniflora* and *Lobelia dortmanna* in low alkaline lakes and *Chara* spp. in high alkaline lakes. The good/moderate boundary is set where the stands of these species disappear (sudden drop), the moderate /poor boundary represents the status where these species disappear totally, while poor/bad boundary is set where all the sensitive species have disappeared and the vegetation consist of only tolerant and/or indifferent species.

The regression lines between total P and TIc-index for very low and low alkalinity types (lake type 001, 002, 101 and 102) have similar slopes, which differ clearly from the slopes for moderate and high alkalinity lakes (lake types 201, 202, 301 and 302).

Boundaries for the different lake types:

Lake type		Index values				
		Ref value	H/G	G/M	M/P	P/B
001	very low alkalinity, clear	95	92	55	40	15
002	very low alkalinity, humic	78	71	55	40	15
101	low alkalinity, clear	79	75	55	40	15
102	low alkalinity, humic	78	71	55	40	15
201	moderate alkalinity, clear	74	65	30	5	-35
202	moderate alkalinity, humic	69	67	30	5	-35
301	high alkalinity, clear	75	63	30	5	-35
302	high alkalinity, humic	73	63	30	5	-35

Calculation of the ecological quality ratio (EQR)

The ecological quality ratio for the TIc index is calculated using the following equation:

$$EQR = \frac{\text{observed index value} + 100}{\text{reference value} + 100}$$

Boundaries for the different lakes types, expressed in EQRs:

Lake type		EQR-values			
		H/G	G/M	M/P	P/B
001	very low alkalinity, clear	0,98	0,79	0,71	0,59
002	very low alkalinity, humic	0,96	0,87	0,79	0,65
101	low alkalinity, clear	0,98	0,87	0,78	0,64
102	low alkalinity, humic	0,96	0,87	0,79	0,65
201	moderate alkalinity, clear	0,95	0,76	0,60	0,37
202	moderate alkalinity, humic	0,99	0,77	0,62	0,38
301	high alkalinity, clear	0,93	0,74	0,60	0,37
302	high alkalinity, humic	0,94	0,75	0,61	0,38

Table A.17 Norwegian aquatic macrophytes in lakes - sensitive, tolerant or indifferent to eutrophication. Species with < 4 loc. in brackets.

Group	Sensitive species	Tolerant species	Indifferent species
ISOETIDS	*Crassula aquatica*	*(Elatine hexandra)*	
	Elatine hydropiper	*Elatine triandra*	
	(Elatine orthosperma)		
	Eleocharis acicularis		
	Isoëtes echinospora		

Group	Sensitive species	Tolerant species	Indifferent species
	Isoëtes lacustris		
	Limosella aquatica		
	Littorella uniflora		
	Lobelia dortmanna		
	Lythrum portula		
	Ranunculus reptans		
	Subularia aquatica		
ELODEIDS	*Callitriche hamulata*	*Callitriche cophocarpa*	*Potamogeton alpinus*
	Callitriche hermaphroditica	*Callitriche stagnalis*	*Potamogeton berchtoldii*
	Callitriche palustris	*Ceratophyllum demersum*	*Potamogeton perfoliatus*
	Hippuris vulgaris	*Elodea canadensis*	*Utricularia vulgaris*
	Juncus bulbosus	*Myriophyllum spicatum*	
	Myriophyllum alterniflorum	*Myriophyllum verticillatum*	
	Myriophyllum sibiricum	*(Najas flexilis)*	
	(Najas marina)	*Potamogeton crispus*	
	(Potamogeton compressus)	*Potamogeton friesii*	
	Potamogeton filiformis	*Potamogeton lucens*	
	(Potamogeton friesii x obtusifolius)	*Potamogeton obtusifolius*	
	Potamogeton gramineus	*Potamogeton pectinatus*	
	Potamogeton x nitens	*Potamogeton pusillus*	
	Potamogeton polygonifolius	*Potamogeton rutilus*	
	Potamogeton praelongus	*(Potamogeton x zizii)*	
	(Potamogeton vaginatus)	*(Potamogeton x suecicus)*	
	(Potamogeton x sparganifolius)	*Ranunculus aquatilis*	
	Ranunculus confervoides	*(Zannichellia palustris)*	
	Ranunculus peltatus		
	Utricularia intermedia		
	Utricularia minor		
	Utricularia ochroleuca		
NYMPHAEIDS	*(Luronium natans)*	*Persicaria amphibia*	*Nuphar lutea*
	Nuphar pumila	*Sparganium emersum*	*Nymphaea alba coll.*
	Sparganium angustifolium		*Potamogeton natans*
	(Sparganium gramineum)		*Sagittaria sagittifolia*
	Sparganium hyperboreum		
	Sparganium natans		
LEMNIDS		*Lemna minor*	
		Lemna trisulca	
		(Ricciocarpus natans)	
		Spirodela polyrrhiza	
CHARIDS	*Chara aspera*	*(Chara intermedia)*	
	(Chara braunii)	*(Chara tomentosa)*	
	Chara contraria		
	Chara delicatula		
	Chara globularis		
	Chara rudis		
	Chara strigosa		
	(Nitella batrachosperma)		
	(Nitella mucronata)		
	Nitella opaca		
	Tolypella canadensis		

Appendix : Example

The surveys in two Norwegian lakes, lake A and lake B, gave these results:

Species	A	B
CHARACEAE		
Chara aculeolata	4	
Chara rudis	4	
ELODEIDS		
Callitriche palustris		1
Hippuris vulgaris	3	
Utricularia vulgaris	3	
NYMPHAEIDS		
Nuphar lutea		3
Nymphaea alba	3	2
Persicaria amphibia		2
Potamogeton natans		2
Sparganium emersum		1-2

Lake A is a high alkalinity, humic lake (type 302), and the aquatic macrophytes is dominated by *Chara* spp.

Index calculation for lake A:

$$TI_c = \frac{3-0}{5} \times 100 \quad \text{which gives} \quad TI_c = 60$$

The reference for the lake type 302 is 73. By using the equation in chapter 3.2 the EQR = 0.92, which shows good status for lake A.

$$\text{EQR} = \frac{60 + 100}{73 + 100} = 0.92$$

Lake B is a low alkalinity, humic lake (lake type 102), dominated by nymphaeids. The submerged vegetatiton where sparse.

Calculation for lake B:

$$TI_c = \frac{1-2}{6} \times 100 \quad \text{which gives} \quad TI_c = 16.67$$

The reference for the lake type 102 is 78. By using the equation in chapter 3.2 the EQR = 0.92, which shows poor status for the lake.

$$\text{EQR} = \frac{16.67 + 100}{78 + 100} = 0.66$$

A.5 Sweden

General information

In order to classify the status of lakes as regards macrophytes, a trophic macrophyte index (TMI) is calculated. It is based on trophic indicator values of all macrophyte species except helophytes. The TMI thus reflect nutrient status, in the first instance the levels of total phosphorus

Sampling

Application of the assessment method for macrophytes in lakes requires that the inventory must cover all macrophytes, including mosses and charophytes, with the exception of helophytes. Sampling must have been carried out during the late summer when aquatic vegetation is fully developed. The inventory is conducted both along the shoreline and from boats using underwater viewing tubes and rake.

The presence of all macrophytes should be noted on a semi-quantitative scale or on a binary scale (present, absent). To define the lakes TMI, however, only binary data are required.

Classification of status

Step 1 Calculate the Trophic Macrophyte Index, TMI.

The TMI of lakes is a weighted mean value of the individual macrophyte indicator values and weight factors:

Trophic Macrophyte Index (TMI) = Sum of (Indicator value for species i * Weight factor for species i)/Sum of weighting factors

Step 2 The Ecological Quality Ration for the respective lakes is calculates as follows:

EQR_{lake} = (Observed TMI_{lake}-1) / (Reference value-1)

Reference values and class boundaries are given in Table A.18.

Table A.18 Reference values and class boundaries for classification of macrophytes in lakes.

Type	Status	TMI EQR
North of Limes Norrlandicus, above the highest coast lime	Reference values	8.54
	High	≥0.97
	Good	≥0.90 and < 0.97
	Moderate	≥0.83 and < 0.90
	Poor, Bad	<0.83
North of Limes Norrlandicus, below the highest coast lime	Reference values	8.16
	High	≥0.97
	Good	≥0.94 and < 0.97
	Moderate	≥0.85 and < 0.94

	Poor, Bad	<0.85
South of Limes Norrlandicus	Reference values	8.27
	High	≥0.98
	Good	≥0.88 and < 0.98
	Moderate	≥0.58 and < 0.88
	Poor, Bad	<0.58

Reference conditions

Key sources to derive reference conditions: Existing near-natural reference sites, Historical data, Least Disturbed Conditions

Reference site characterisation: 49 Sites are scattered all over Sweden.

Criteria: Proportion of clear-cuts within the lakes catchments <10%, that of agricultural land <10% and that of urban areas <0.1%. No lowering of water level. Tot-P <12.5 µg/l, Tot-N <300 microgram µg/l, pH >6.0

Reference community description:

Reference lakes are generally characterized by the occurrence of one or several species of isoetids (Isoetes, Lobelia, Subularia etc).

Boundary setting procedure:

For the high/good boundary, the 5th percentile of the reference lakes was taken.

Class boundaries (between good and poorer status) were determined with classification trees using Tot-P values of species typical for the different classes of ecological status. The species used for classification were those showing sudden drops in their occurrence beyond the 75th percentile.

At good status stands of the sensitive taxa (large isoetids, Littorella, Lobelia, Isoetes in low alkalinity lakes or Chara spp. in high alkalinity lakes) occur, but significantly decrease at good-moderate boundary ("sudden drop") and are replaced by tolerant taxa.

A.6 United Kingdom :UKTAG Lake assessment methods Macrophyte and Phytobenthos

Macrophytes (Lake LEAFPACS)

Which indicators are used?

The method assesses the condition of the quality element by combining information on the parameters listed below. The parameters are calculated using information on macrophyte species and groups of such species. The results for each parameter are then used to produce an ecological quality ratio (EQR) for the combined parameters. The combined parameters are referred to as Lake LEAFPACS.

I. Lake Macrophyte Nutrient Index (LMNI);

II. Number of functional groups of macrophyte taxa (NFG).

III. Number of macrophyte taxa (NTAXA);

IV. Mean percent cover of hydrophytes (COV); and

V. Relative percent cover of filamentous algae (ALG)

How are these indicators monitored?

In order to obtain the data with which to calculate the observed values for each of the parameters, a minimum of four lake sectors should be surveyed. In the largest lakes eight lake sectors will be required. A sector should comprise a 100 metre length of shoreline. It should extend from the shore to the centre of the lake or to the maximum depth of colonisation of macrophytes, whichever is the shorter distance from the shore. The sectors should be arranged to give an approximately equal spread around the perimeter of the lake.

Surveys should normally be conducted from June until September.

The lake should be surveyed in order to establish the presence of each of the macrophyte taxa listed in column 1 of Table 1. Where it is not possible to identify a macrophyte to the taxonomic level listed in Column 1 of Table 1 it should be recorded using the next highest taxonomic level, provided this is listed in Column 1 of Table 1.

Each taxon listed in Column 1 of Table 1 and present in the lake should be assigned a value (0 -100 %) which is an estimate of the percentage cover of the taxon in the area of the lake surveyed.

The surveying method should conform to EN 15460 : 2007 Water quality – Guidance standard for the surveying of macrophytes in lakes.

Methods of calculation

Calculation of the observed value for each parameter

I. Lake Macrophyte Nutrient Index (LMNI)
In order to calculate the observed value of the parameter, LMNI, each macrophyte taxon listed in Column 1 of Table A.19 and identified as being present in the lake should be assigned the corresponding lake macrophyte nutrient index score in Column 2 of that Table. The observed value of the parameter should be calculated by the equation:

$$\text{Observed value of LMNI} = \frac{\sum_{j=1}^{n} \text{LMNI}_j}{N}$$

where:

" $LMNI_j$ " is the Lake Macrophyte Nutrient Index score for taxon "j" given in Column 2 of Table A.19;

"j" represents a taxon listed in Column 1 of Table A.19 and present in the sample. "j" has a value of 1 to "n" used to indicate which of the all the taxa (total number = "n") listed in Column 1 of Table A.19 and present in the sample it represents; and

"N" is the total number of macrophyte taxa listed in Column 1 of Table A.19 and identified as being present in the lake.

II. Number of functional groups of macrophyte taxa (NFG)
In order to calculate the observed value for the parameter, NFG, each taxon listed in Column 1 of Table A.19 and identified as present in the lake should be assigned to the corresponding functional group in Column 3 of Table A.19, if a corresponding functional group is listed for that taxon in that column.

The observed value for the parameter, NFG, is given by the sum of the number of different functional groups of taxa identified as present in the lake.

III. Number of macrophyte taxa (NTAXA)
The observed value for the parameter, NTAXA, is given by the sum of the number of taxa listed in Column 1 of Table A.19 that are present in the lake.

IV. Mean percent cover of hydrophytes (COV)
The observed value for the parameter, COV, should be calculated according to the following equation:

$$\text{Observed value of COV} = \frac{\sum_{j=1}^{n} \%\text{COV}_j}{N}$$

where:

" $\%COV_j$ " is the percentage cover of taxon "j" in the area of the lake surveyed;

"j" represents a taxon listed in Column 1 of Table A.19 and present in the sample. "j" has a value of 1 to "n" used to indicate which of the all the taxa (total number = "n") listed in Column 1 of Table A.19 and present in the sample it represents; and "N" is the total number of macrophyte taxa listed in Column 1 of Table A.19 and identified as being present in the lake.

V. Relative percent cover of filamentous algae (ALG)

The observed value for the parameter, ALG, should be calculated according to the following equation:

$$\text{Observed value of ALG} = \frac{\sum_{k=1}^{n} \%F_k}{\sum_{j=1}^{n} \%COV_j}$$

where:

"$\%F_k$" is the percentage cover of taxon "k" in the area of the lake surveyed;

"k" represents a taxon listed in Column 1 of Table A.19, indicated as being a filamentous algal taxon in Column 4 of that Table and present in the sample. "k" has a value of 1 to "n" used to indicate which of the all the taxa (total number = "n") listed in Column 1 of Table A.19, indicated as being a filamentous algal taxon in Column 4 of that Table and present in the sample it represents.

Calculation of the reference value for each parameter

Reference conditions were derived using a combination of (a) information from a network of lakes identified as being subject to no or very minor alterations likely to affect their macrophyte communities; and (b) modelling using predictive models and hindcasting methods. For the purposes of the latter, data on individual species-pressure relationships indicated by empirical analysis and historical macrophyte records were used.

I. Lake Macrophyte Nutrient Index (LMNI)
The expected LMNI value is related to the Morpho-Edaphic Index (MEI) where

$MEI = Log_{10}$ ([(Alk + 40) ÷ 1000] ÷ D)

The model that is used to calculate expected LMNI depends on the geology of the lake catchment. This is summarised using the weighted Freshwater Sensitivity Class (wFSC) where:

wFSC = F_1/100 + [F_2/100 x 2] + [F_3/100 x 3] + [F_4/100 x 4] + [F_5/100 x 5];

"Freshwater Sensitivity Class" describes the relative capacity of geology and soils to neutralise incoming acidity and hence limit acid loadings to fresh surface waters. There are five classes ranging from F_1 (highly sensitive) to F_5 (low sensitivity).The classes are derived from the Centre for Ecology and Hydrology Freshwater Sensitivity Class map; Hornung *et.al.* (1995). In the above equation the terms F_1 to F_5 describes the % cover of the lake catchment assignable to each of the five possible sensitivity classes.

The value for the parameter, LMNI, in the reference conditions applicable to the lake should be calculated using the following equation:

If wFSC ≥ 4.0 (i.e. well buffered catchments with soft calcareous geology):

Reference LMNI = 4.969 + 1.272 x MEI + 0.193 x MEI^2

If wFSC< 4.0 (i.e. poorly buffered catchments or those with hard calcareous geology):

Reference LMNI = 4.969 + 1.272 x MEI + 0.193 x MEI^2 - 0.55

II. Number of functional groups of macrophyte taxa (NFG)
The value for the parameter, NFG, in the reference conditions applicable to the lake should be calculated using the following equation:

Reference N_FG = Exponent (0.703 – [0.049 x Log_{10} H] + [0.133 x Log_{10} S] + [0.287 x Log_{10} (Alk +40)] + [0.132 (only if lake is in GB)] + [0.356 (only if wFSC<4.0)])

III. Number of macrophyte taxa (NTAXA)
The value for the parameter, NTAXA, in the reference conditions applicable to the lake should be calculated using the following equation:

Reference NTAXA = Exponent (1.488 – [0.098 x Log_{10} H] + [0.185 x Log_{10} S] + [0.194 x Log_{10} (Alk +40)] + [0.149 (only if lake is in GB)] + [0.287 (only if wFSC <4.0)])

where, in the above equations:

“Alk” is the annual mean reference alkalinity in μeq L^{-1};

“D” is the mean depth of the lake in metres;

"H" is the height in metres of the surface of the lake above mean sea level;

"S" is the surface area of the lake in hectares;

GB refers to those lakes not situated on the island of Ireland.

IV. Mean percent cover of hydrophytes (COV)
The value used for the parameter, COV, in the reference conditions applicable to the lake is dependent on the method of data collection. This metric must be excluded if no formal assessment of cover or frequency has been undertaken, or if data has been collected using strand line surveys (e.g. due to the lack of a boat). Provided that data has been collected using the recommended survey method Reference COV = 8.2% should be applied in all lakes.

V. Relative percent cover of filamentous algae (ALG)
The value used for the parameter, ALG, in the reference conditions applicable to the lake should be 0.05

Calculation of the ecological quality ratio (EQR) for each parameter

I. Lake Macrophyte Nutrient Index (LMNI)

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

If the reference value for LMNI is ≥5:

EQR_{LMNI} = (observed value of LMNI - 10) ÷ (reference value for LMNI - 10)

If the reference value for LMNI is <5:

EQR_{LMNI} = (observed value of LMNI – (reference value for LMNI + 5)) ÷ (reference value for LMNI - (reference value for LMNI + 5))

II. Number of functional groups of macrophyte taxa (NFG)
The ecological quality ratio (EQR) for the parameter, NFG, should be calculated using the following equation:

EQR_{NFG} = observed value of NFG ÷ reference value for NFG

unless the observed value of NFG = 0 in which case EQR_{NFG} = 0.

III. Number of macrophyte taxa (NTAXA)
The ecological quality ratio (EQR) for the parameter, NTAXA, should be calculated using the following equation:

EQR_{NTAXA} = observed value of NTAXA ÷ reference value for NTAXA

unless the observed value of NTAXA = 0 in which case EQR_{NTAXA} = 0.

IV. Mean percent cover of hydrophytes (COV)
The ecological quality ratio (EQR) for the parameter, COV, should be calculated using the following equation:

EQR_{COV} = √ observed value of COV ÷ √ reference value for COV

V. Relative percent cover of filamentous algae (ALG)
If the observed value of ALG is > 0.05, the ecological quality ratio for the parameter should be calculated using the following equation:

EQR_{ALG} = [observed value of ALG - 1] ÷ [0.05 – 1]

If the observed value of ALG is ≤ 0.05, the ecological quality ratio for the parameter should be given the value "1".

Combining the ecological quality ratios for the different parameters

The ecological quality ratio for the combined parameters ($EQR_{LEAFPACS}$) should be determined as follows:

If the values of either EQR_{NFG} or EQR_{NTAXA} are less than the value of EQR_{LMNI}, a diversity adjusted EQR (^{A}EQR) for the parameter, LMNI, should be calculated as follows:

$^{A}EQR_{LMNI}$ = [EQR_{LMNI} + ($^{A}EQR_{NFG}$ or $^{A}EQR_{NTAXA}$, whichever is the smaller x 0.5)] ÷ 1.5

If EQR_{LMNI} is less than the values of either EQR_{NFG} or EQR_{NTAXA} the value of EQR_{LMNI} is unchanged (i.e. EQR_{LMNI} = $^{A}EQR_{LMNI}$).

If the value of $^{A}EQR_{LMNI}$ is larger than whichever is the smaller of the values for EQR_{COV} and EQR_{ALG}, $EQR_{LEAFPACS}$ should be calculated using the following equation:

$EQR_{LEAFPACS}$ = [$^{A}EQR_{LMNI}$ + (0.25 × {EQR_{COV} or EQR_{ALG}, whichever is the smaller})] ÷ 1.25

If the value of $^{A}EQR_{LMNI}$ is smaller than or the same as whichever is the smaller of the values for EQR_{COV} and EQR_{ALG}, $EQR_{LEAFPACS}$ should be assigned the same value as $^{A}EQR_{LMNI}$

Application of the method for the purposes of classification

When using the method for the purposes of classifying the ecological status of a water body:

1. a standardised ecological quality ratio (^{S}EQR) should be calculated for $EQR_{LEAFPACS}$ as follows:

   If the value of $EQR_{LEAFPACS}$ is < 0.20, $^{S}EQR_{LEAFPACS}$ should be assigned a value of "0".
   If the value of $EQR_{LEAFPACS}$ is > 1.05, $^{S}EQR_{LEAFPACS}$ should be assigned a value of "1".
   Otherwise, $^{S}EQR_{LEAFPACS}$ should be calculated using the following equations:

   If $EQR_{LEAFPACS}$ ≥ 0.8: $^{S}EQR_{LEAFPACS}$ = ([$EQR_{LEAFPACS}$ - 0.8] ÷ [1.05 - 0.8]) x 0.2 + 0.8,

   If $EQR_{LEAFPACS}$ ≥ 0.66: $^{S}EQR_{LEAFPACS}$ = ([$EQR_{LEAFPACS}$ – 0.66] ÷ [0.8 - 0.66]) x 0.2 + 0.6

   If $EQR_{LEAFPACS}$ ≥ 0.51: $^{S}EQR_{LEAFPACS}$ = ([$EQR_{LEAFPACS}$ – 0.51] ÷ [0.66 - 0.51]) x 0.2 + 0.4

   If $EQ_{LEAFPACS}$ ≥ 0.35: $^{S}EQR_{LEAFPACS}$ = ([$EQR_{LEAFPACS}$ – 0.35] ÷ [0.51 - 0.35]) x 0.2 + 0.2

   If $EQR_{LEAFPACS}$ < 0.35: $^{S}EQR_{LEAFPACS}$ = ([$EQR_{LEAFPACS}$ – 0.20] ÷ [0.35 - 0.20]) x 0.2

   and

2. the value of $^{S}EQR_{LEAFPACS}$ for surveys carried out between July and September should be used. If surveys have been carried out in more than one year the mean value of $^{S}EQR_{LEAFPACS}$ should be used.

The value of $^{S}EQR_{LEAFPACS}$ should then be assigned to an ecological status class according to the Table below.

$^{S}EQR_{LEAFPACS}$	Status
0.80 - 1.0	High
0.60 - 0.79	Good
0.40 - 0.59	Moderate
0.20 - 0.39	Poor
0 - 0.20	Bad

How are reference conditions, H/G and G/M boundaries derived?

Putative reference sites were identified at a type-specific level initially from their biology, using individual species-pressure relationships indicated by empirical analysis, historical macrophyte records and expert opinion. Finally all reference sites remaining were checked against available land cover, total P and chlorophyll data. Within-type regressions between pressures and biological metrics were used to identify sites where deviating biology was related to increased pressure. Any such outliers or sites with known hydromorphological modifications were then removed.

Individual metrics were modelled using environmental variables to determine their expected value at reference sites. These expected values are used to calculate an EQR for each metric. A multimetric EQR is then calculated based on the national combination rules. The H/G boundary corresponds to the lower 5^{th} percentile of the multimetric EQR in reference sites and is interpreted as representing the lower limit of undisturbed status of the quality element. The GM boundary is based on the interval between the median EQR of the national reference site dataset and the HG boundary and is approximately equivalent to the lower 1%tile of the reference site multimetric EQR. This point is interpreted to represent the limit of slight change in the quality element since there is some but minimal overlap with the natural variation in the population of reference sites Below this the EQR range is divided equally to form the MP and PB boundaries.

How well do these indicators correlate with pressure indicators?

The relationship between LMNI and Total P (annual mean) in the UK dataset is summarised in Figure A.5 below. Since this is an internally validated model (LMNI is calibrated from the mean TP values for each species in lakes where they are recorded) the highly significant relationship that is observed (r^2 = 0.62) is to be expected. However, LMNI also performs extremely well if applied to a completely independent dataset composed of the combined N-GIG and CB-GIG lake datasets (r^2 = 0.5) and in this dataset is only fractionally worse than the internally calibrated ICCM that was based on this data.

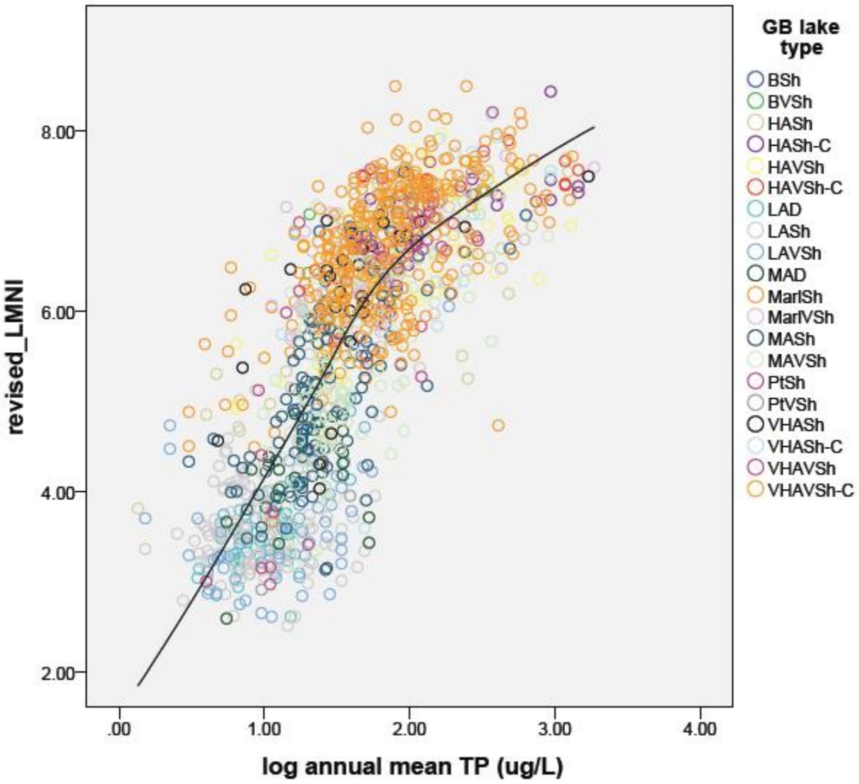


Figure A.5 Global relationship between LMNI and lake Total P (annual mean) in UK lakes dataset (n=1359, relationhip based on lakes containing >4 taxa).

Further reading

Hornung, M et.al. (1995) The sensitivity of surface waters of Great Britain to acidification predicted from catchment characteristics. Environmental Pollution 87, 207-214

Willby, N.J., Pitt, J & Phillips G. L. (2009) The ecological classification of UK lakes using aquatic macrophytes. Environment Agency, Science Report SC010080/SR

Appendix 1 UK Species list

Table A.19 List of lake macrophyte taxa and associated information for the calculation of the values for the parameters

Macrophyte taxa	Lake macrophyte nutrient index score	Number of functional group	Taxa indicated as filamentous algal taxa ("F")	Included in CBGIG
Alisma gramineum	7.65	13		
Apium inundatum	4.32	7		Y
Aponogeton distachyos	8.88	16		
Azolla filiculoides	7.25	1		
Baldellia ranunculoides	3.97	13		Y
Batrachospermum sp.	1.56			

Macrophyte taxa	Lake macrophyte nutrient index score	Number of functional group	Taxa indicated as filamentous algal taxa ("F")	Included in CBGIG
Butomus umbellatus	7.97	13		Y
Callitriche brutia var. brutia	2.26	6		Y
Callitriche brutia var. hamulata	4.08	6		
Callitriche hermaphroditica	8.08	5		Y
Callitriche obtusangula	9.34	6		Y
Callitriche platycarpa	9.50	6		Y
Callitriche sp.	7.11	6		Y
Callitriche stagnalis	6.38	6		Y
Callitriche truncata	8.28	6		Y
Ceratophyllum demersum	7.99	5		Y
Ceratophyllum submersum	6.78	5		Y
Chara aculeolata	3.49	2		Y
Chara aspera	4.19	2		Y
Chara baltica	5.83	2		Y
Chara canescens	4.73	2		Y
Chara connivens	5.60	2		Y
Chara contraria var. contraria	5.06	2		Y
Chara contraria var. hispidula	6.41	2		Y
Chara curta	4.14	2		Y
Chara globularis	6.86	2		Y
Chara hispida	3.95	2		Y
Chara intermedia	5.04	2		Y
Chara rudis	3.93	2		Y
Chara sp.	5.57	2		Y
Chara virgata	4.29	2		Y
Chara virgata var. annulata	4.07	2		Y
Chara vulgaris	5.56	2		Y
Crassula helmsii	5.57	5		Y
Damasonium alisma	6.19	13		
Elatine hexandra	3.81	11		Y
Elatine hydropiper	5.34	11		Y
Eleocharis acicularis	8.68	4		Y
Eleocharis multicaulis	3.03	4		Y

Macrophyte taxa	Lake macrophyte nutrient index score	Number of functional group	Taxa indicated as filamentous algal taxa ("F")	Included in CBGIG
Eleogiton fluitans	2.03	15		Y
Elodea callitrichoides	7.64	5		
Elodea canadensis	7.45	5		Y
Elodea nuttallii	6.19	5		Y
Eriocaulon aquaticum	1.47	4		
Filamentous algae	6.70		F	Y
Fontinalis antipyretica	4.19	3		Y
Fontinalis squamosa	3.09	3		Y
Groenlandia densa	5.35	5		
Hippuris vulgaris	5.23	7		Y
Hottonia palustris	6.29	7		Y
Hydrocharis morsus-ranae	6.51	8		Y
Hydrodictyon reticulatum	8.42		F	
Hypericum elodes	3.56	11		Y
Isoetes echinospora	2.47	4		Y
Isoetes lacustris	2.22	4		Y
Isoetes sp.	2.22	4		Y
Juncus bulbosus	2.42	4		Y
Lagarosiphon major	3.51	5		Y
Lemna gibba	7.66	1		Y
Lemna minor	8.52	1		Y
Lemna minuta	10.00	1		Y
Lemna trisulca	7.96	1		Y
Leptodyction riparium	8.71	3		
Limosella aquatica	3.80	11		Y
Littorella uniflora	3.73	4		Y
Lobelia dortmanna	2.16	4		Y
Ludwigia palustris	3.82	11		Y
Luronium natans	3.52	13		Y
Lythrum portula	4.31	11		Y
Menyanthes trifoliata	5.17	10		Y
Myriophyllum alterniflorum	2.66	7		Y
Myriophyllum aquaticum	6.87	7		
Myriophyllum spicatum	6.23	7		Y
Myriophyllum verticillatum	5.32	7		Y
Najas flexilis	2.89	14		Y

Macrophyte taxa	Lake macrophyte nutrient index score	Number of functional group	Taxa indicated as filamentous algal taxa ("F")	Included in CBGIG
Najas marina	5.24	14		Y
Nitella confervacea	3.28	2		Y
Nitella flexilis agg.	5.19	2		Y
Nitella gracilis	3.56	2		Y
Nitella mucronata	5.67	2		Y
Nitella opaca	2.36	2		Y
Nitella sp.	4.66	2		Y
Nitella translucens	2.73	2		Y
Nitellopsis obtusa	5.23	2		Y
Nuphar lutea	7.47	12		Y
Nuphar pumila	4.82	12		Y
Nuphar x spenneriana	3.65	12		Y
Nymphaea alba	6.84	12		Y
Nymphoides peltata	6.75	10		Y
Persicaria amphibia	8.25	10		Y
Pilularia globulifera	3.59	4		Y
Potamogeton alpinus	4.48	16		Y
Potamogeton berchtoldii	6.58	14		Y
Potamogeton coloratus	3.46	16		Y
Potamogeton compressus	5.18	14		Y
Potamogeton crispus	7.50	17		Y
Potamogeton epihydrus	1.00	16		
Potamogeton filiformis	3.68	15		Y
Potamogeton friesii	4.71	14		Y
Potamogeton gramineus	2.85	16		Y
Potamogeton lucens	4.37	17		Y
Potamogeton natans	4.71	16		Y
Potamogeton obtusifolius	6.97	14		Y
Potamogeton pectinatus	7.19	15		Y
Potamogeton perfoliatus	4.42	17		Y
Potamogeton polygonifolius	2.39	16		Y
Potamogeton praelongus	3.92	17		Y
Potamogeton pusillus	7.54	14		Y
Potamogeton rutilus	5.49	14		Y
Potamogeton trichoides	5.79	14		Y
Potamogeton x cooperi	4.93	17		

Macrophyte taxa	Lake macrophyte nutrient index score	Number of functional group	Taxa indicated as filamentous algal taxa ("F")	Included in CBGIG
Potamogeton x griffithii	2.57	16		
Potamogeton x lintonii	7.21	14		
Potamogeton x nitens	3.48	17		Y
Potamogeton x salicifolius	5.89	17		
Potamogeton x sparganifolius	3.71	16		Y
Potamogeton x suecicus	4.62	15		Y
Potamogeton x zizii	4.04	16		Y
Ranunculus (sub sect. Batrachian) sp.	5.31	18		
Ranunculus aquatilis agg.	6.30	18		Y
Ranunculus aquatilis var diffusus	4.20	18		
Ranunculus aquatilis var. aquatilis.	5.81	18		Y
Ranunculus circinatus	8.70	5		Y
Ranunculus fluitans	5.65	18		
Ranunculus hederaceus	8.33	11		Y
Ranunculus lingua	6.79	10		Y
Ranunculus omiophyllus	5.51	11		Y
Ranunculus peltatus subsp. baudotii	6.48	18		Y
Ranunculus peltatus subsp. peltatus	6.49	18		Y
Ranunculus penicillatus subsp. penicillatus	4.21	18		
Ranunculus penicillatus subsp. pseudofluitans	6.68	18		
Riccia fluitans	6.35	1		Y
Ricciocarpus natans	5.32	1		Y
Ruppia cirrhosa	7.03	15		Y
Ruppia maritima	7.85	15		Y
Ruppia sp.	8.08	15		
Sagittaria sagittifolia	6.01	12		Y
Sparganium angustifolium	2.52	13		Y
Sparganium emersum	6.06	13		Y
Sparganium natans	2.79	13		Y
Sphagnum (aquatic indet.)	2.74	3		Y

Macrophyte taxa	Lake macrophyte nutrient index score	Number of functional group	Taxa indicated as filamentous algal taxa ("F")	Included in CBGIG
Spirodela polyrhiza	9.62	1		Y
Stratiotes aloides	6.20	8		Y
Subularia aquatica	1.80	4		Y
Tolypella glomerata	5.32	2		Y
Ulva (Enteromorpha) flexuosa	9.05		F	
Utricularia australis	2.87	9		Y
Utricularia intermedia sens.lat.	1.61	9		Y
Utricularia minor	2.36	9		Y
Utricularia ochroleuca	1.04	9		Y
Utricularia sp.	3.34	9		Y
Utricularia stygia	1.30	9		Y
Utricularia vulgaris	4.24	9		Y
Zannichellia palustris	8.69	15		Y

Appendix 2 Worked example

The following data were obtained from a GB lake survey.

The values below represent the cover determined from a single survey covering a minimum of four sectors of a lake.

Taxon identified as present in the lake	% cover in sampled area	Lake macrophyte nutrient index score	Number of functional group
Chara aspera	10	4.19	2
Elodea canadensis	1	7.45	5
Hippuris vulgaris	5	5.23	7
Nitellopsis obtusa	2	5.23	2
Nymphaea alba	10	6.84	12
Potamogeton obtusifolius	5	6.97	14

In addition, the following environmental data were obtained:

Variable	Value
Lake altitude (H)	15 metres
Mean depth (D)	2.7 metres
Area (S)	3.1 hectares
Reference alkalinity (Alk)	1700 μeq L^{-1}

weighted Freshwater Sensitivity Class (wFSC)	4.1

LMNI

The observed value of LMNI is calculated as follows:

1. Sum LMNI scores for all taxa = 35.91
2. Divide this value by the number of taxa present (6) = 5.99

The reference value is calculated using the equation in section 3.2. This results in a reference value for LMNI of 4.73.

EQR_{LMNI} = (5.99 – (4.73 + 5)) / (4.73 – (4.73 + 5)) = 0.75.

Functional diversity (NFG)

The observed number of functional groups (NFG) for this lake is 5 (*Chara aspera* and *Nitellopsis obtusa* are in group 2, *Potamogeton obtusifolius* group 14, *Nymphaea alba* group 12, *Hippuris vulgaris* group 7 and *Elodea canadensis* group 5).

The reference value is calculated using the equation in section 3.2. This results in a reference value for NFG of 5.89.

EQR_{NFG} = observed NFG / reference NFG = 0.85

Number of taxa (NTAXA)

The observed number of taxa (NTAXA) is 6.

The reference value is calculated using the equation in section 3.2. This results in a reference value for NTAXA of 9.41.

EQR_{NTAXA} = observed value of NTAXA / reference value for NTAXA = 0.64

Mean percent cover (COV)

The observed value for COV is calculated as follows:

1. Sum % cover values for all taxa = 33
2. Divide this value by the number of taxa present (6) = 5.5

A reference value for COV of 8.2 applies to those lakes where data is collected by the recommended method.

$EQR_{COV} = \sqrt{5.5} \div \sqrt{8.2} = 0.82$

Relative cover of algae (ALG)

As the relative cover of filamentous algae is < 0.05, EQR_{ALG} = 1.00

Combining metrics

The complete results for this lake are, therefore, as follows:

Parameter	Observed value	Reference value	EQR
LMNI	5.99	4.73	0.75
NFG	5.00	5.89	0.85
NTAXA	6.00	9.41	0.64
COV	5.50	8.20	0.82
ALG	0.0	0.05	1.00

EQR_{LMNI} is larger than the lowest of EQR_{NFG} and EQR_{NTAXA} (0.64) so the diversity adjusted ecological quality ratio for LMNI is given by:

$$^{A}EQR_{LMNI} = [(0.75 + (0.64 \times 0.5)] \div 1.5 = 0.71$$

The value of $^{A}EQR_{LMNI}$ (0.71) is less than the values of EQR_{COV} (0.82) and EQR_{ALG} (1.00) and is therefore taken as the value for $EQR_{LEAFPACS}$. This value is then standardised according to the formula in Section 3.5 such that:

$$^{S}EQR_{LEAFPACS} = ([0.71 – 0.66] \div [0.8 – 0.66]) \times 0.2 + 0.6 = 0.67$$

$^{S}EQR_{LEAFPACS}$ values in the range 0.6 to 0.8 are assigned to Good Ecological status (section 3.5). Therefore the status of this water body based on its macrophyte assemblage would be G

B. Maximum Colonization Depth (Cmax) as a Predictor of Macrophyte Trophic State in Norwegian Lakes - Preliminary Analysis

Introduction

Preliminary analyses within the WISER project have pointed to the potential for C_{max} to improve current or proposed macrophyte-based indices, but also to high levels of uncertainty (Kolada et al. 2011).

The purpose of this report is to assess whether C_{max} should be used as an abundance metric in Norwegian lakes; i.e. evaluate: 1) if the collection methods and data for lower depth and secchi depth are sufficiently accurate, 2) if the index has an acceptable correlation with pressure, and 3) the potential contribution of C_{max} as an additional variable to the purely compositional TIc-index.

The report presents preliminary results from the first quantitative analysis.

Methods

The maximum colonization depth (C_{max}) is assessed by different methods in the field. In most lakes C_{max} is assessed from a boat, using an aquascope and rake. In around 25 lakes C_{max} is based on analysis of underwater photos (Rørslett 1978). The differences in the two field methods need to be tested further. The dataset includes 149 lakes, from almost every county in Norway.

After some empirical applications of several types of regression models, including nonlinear and non-parametric, linear regression was found to be the best descriptor (i.e., the one consistently yielding the highest R^2 values) of the Secchi–C_{max} relationship. Also, the analysis of residuals revealed (near) homoscedasticity for all performed regressions. The parametric linear model was then applied to all subsequent analysis, except for the few instances the model was evidently nonlinear (Zar 2009). Most regressions were run against Secchi, with a lower number of regressions run against TP.

Kolada et al. (2011) adopted an approach similar to Prairie (1996), with the lower threshold value of R^2=0.30 assumed to accurately describe their high-replication regression models. Because the methods, rationales, general scopes, and datasets used in this analysis overlap to a high degree with Kolada et al.'s (2011) own analysis, regressions models in the current analysis that yielded values of $R^2 \geq 0.30$ were assumed as significant quantitative descriptors of the relationships under examination. Nonetheless, some *p* values were provided to allow for a full statistical report. Statistical analysis was carried out with Addinsoft© XLSTAT 2011.1.05 software.

In case of multiple-lake data, data were retained when sampling referred to well-separated spatial and/or temporal observations. Data deemed spatially and/or temporally too close were discarded; in such cases, the most reliable or complete data point was retained for any particular subset of closely related points. The original dataset included 149 data points encompassing all Norway. Variables for each data point included C_{max}, Secchi, lake type based on alkalinity and colour, and a short list of the dominant macrophyte species at C_{max}. Because not all data points had a complete set of variables, *n* varied according to the scope of the analysis at hand (e.g., data points missing information on alkalinity were discarded from the analysis targeting the effect of alkalinity on the Secchi–C_{max} relationship, leading to a grand total <149 lakes).

Regression models were run for all Norwegian lakes and for separate subsets of data, targeting different situations. Norway was divided into three main regions — northern, central, and southern (Table B.1). The regional division was based on major climate and ecoregional characteristics, and was carried out by county. Though some southern counties include high-mountain areas, the vast majority of the sampled lakes in such counties were located in coastal and lowland areas. In later analysis we will divide by latitude, as well.

Table B.1 The northern, central, and southern regions of Norway (as determined by main climate and ecoregion characteristics) used for the regional analysis in this report, by county.

Northern		Central		Southern	
Abbrev.	**County**	**Abbrev.**	**County**	**Abbrev.**	**County**
FI	Finnmark	NO	Nordland	SF	Sogn og Fjordane
TR	Tromsø	NT	Nord-Trøndelag	OP	Oppland
		MR	Møre og Romsdal	HE	Hedmark
		ST	Sud-Trøndelag	HO	Hordaland
				BU	Buskerud
				RO	Rogaland
				VA	Vest-Agder
				AA	Aust-Agder
				TE	Telemark
				VE	Vestfold
				OS	Oslo
				AK	Akershus
				ØS	Østfold

Subsets were also obtained by dividing the lakes into two categories: according to type of macrophyte species dominating C_{max} (angiosperms *vs.* charophytes), and according to the degree of alkalinity of lake water (Table B.2).

Table B.2 Lake types, all counties combined. Number of lakes refers to lakes used in the alkalinity regressions only.

Abbrev.	Alkalinity	Water Colour	No. of lakes, by region		
			North	Central	South

001	<1 mg Ca L^{-1}	Clear	-	-	6
002		Humic	-	-	-
101	1–4 mg Ca L^{-1}	Clear	-	3	6
102		Humic	-	1	3
201	4–20 mg Ca L^{-1}	Clear	2	11	13
202		Humic	-	3	4
301	> 20 mg Ca L^{-1}	Clear	4	12	25
302		Humic	-	9	3

In a series of preliminary regression analyses, the slope for lakes with C_{max} dominated by the macroalga *Nitella* spp. was evidently steeper than for other lakes, regardless of region or lake type, supporting earlier findings of biological characteristics for *Nitella* different from other macrophytes (e.g., much higher tolerance for high hydrostatic pressure, and hence much deeper possible C_{max}, for *Nitella*: Golubić 1963). Therefore, lakes whose C_{max} was dominated by *Nitella* were excluded from further general analysis, and treated separately.

The quantitative Secchi–C_{max} relationship obtained by regression analysis was used as the basis to attempt to "add value" to the TI_c index (a trophic state index based on presence/absence of sensitive and tolerant macrophyte species) by Mjelde (unpublished), according to which

$$TI_C = \frac{N_S - N_T}{N} \times 100$$

where N_S is the number of species sensitive to eutrophication, N_T is the number of tolerant species, and N is the total number of species, including indifferent species. All aquatic macrophyte life form groups (isoetids, elodeids, nymphaeids, lemnids, and charophytes) are included. The index can range between +100, when all species found in the lake are defined as sensitive (i.e., the lake is oligotrophic), and -100, when all species are tolerant (i.e., the lake is highly eutrophic) (Mjelde, unpublished).

The search for a relationship between C_{max} — alone or in relation to Secchi — and the TI_c index was attempted with a quantitative approach. Lakes were divided into three broad categories, namely lakes with Secchi to bottom, lakes with Secchi < bottom, and lakes with C_{max} dominated by *Nitella*, regardless of region or alkalinity.

Results

Regressions Secchi depth - C_{max}

As for the broader-reaching panEuropean and North American analyses (Carlson 1977; Kolada et al. 2011), the C_{max}–Secchi relationship was highly significant for Norway

(Figure B.1), suggesting that C_{max} has a high potential as a measure of trophic state in Norway.

The high degree of correlation between C_{max} and Secchi was maintained when data were analyzed by region, with the highest R^2 values observed for northern lakes despite the low sample size (Figure B.2). Regression slopes for the three regions were statistically similar [ANCOVA for multiple comparisons (Zar 2009): pooled F=0.659, df=99, p=0.776]. Further analysis by region was thus limited to specific examples. Similar slopes across Norway may be due to the combination of relatively similar ecological conditions across much of Norway and to a relatively high number of lakes in good and high ecological state. However, earlier analysis (Kolada et al 2011) showed different slopes from south to north. This difference has to be further analyzed. The high R^2 for northern lakes despite the small sample size (n=6) is likely due to the low variability in climate, geological, and lake characteristics in this region, which tend to share high water transparency and healthy ecosystem state.

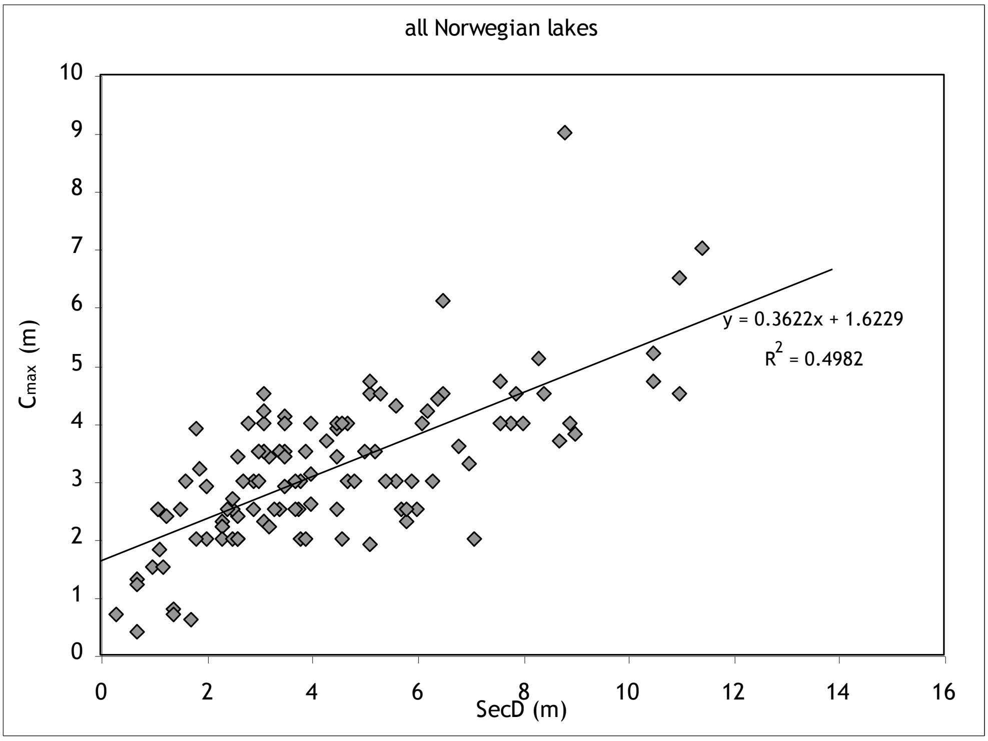


Figure B.1 Linear regression between Secchi depth and C_{max} for all Norwegian lakes, excluding lakes with C_{max} dominated by nymphaeids, mosses, or Nitella (n=117, df=115, p<0.0001).

Table B.3 Linear regressions between Secchi depth and C_{max} for Norwegian lakes, by region and macrophyte type at C_{max} (ang= angiosperms, char=charophytes). Lakes with C_{max} dominated by nymphaeids, mosses, or Nitella were excluded. Regressions *and slope comparisons (t-test-based ANCOVAs) were performed only for datasets with n≥3.*

	Northern		Central		Southern	
	Ang	Char	Ang	Char	Ang	Char
n	5	-	38	2	61	7
df	3	-	36	-	59	5
R2	0.5899	-	0.3789	-	0.4731	0.9272
p	0.129	-	0.0001	-	<0.0001	0.0005
slope	0.2474	-	0.2858	-	0.3865	0.3556
					t=1.185	df=64
					p=0.240	

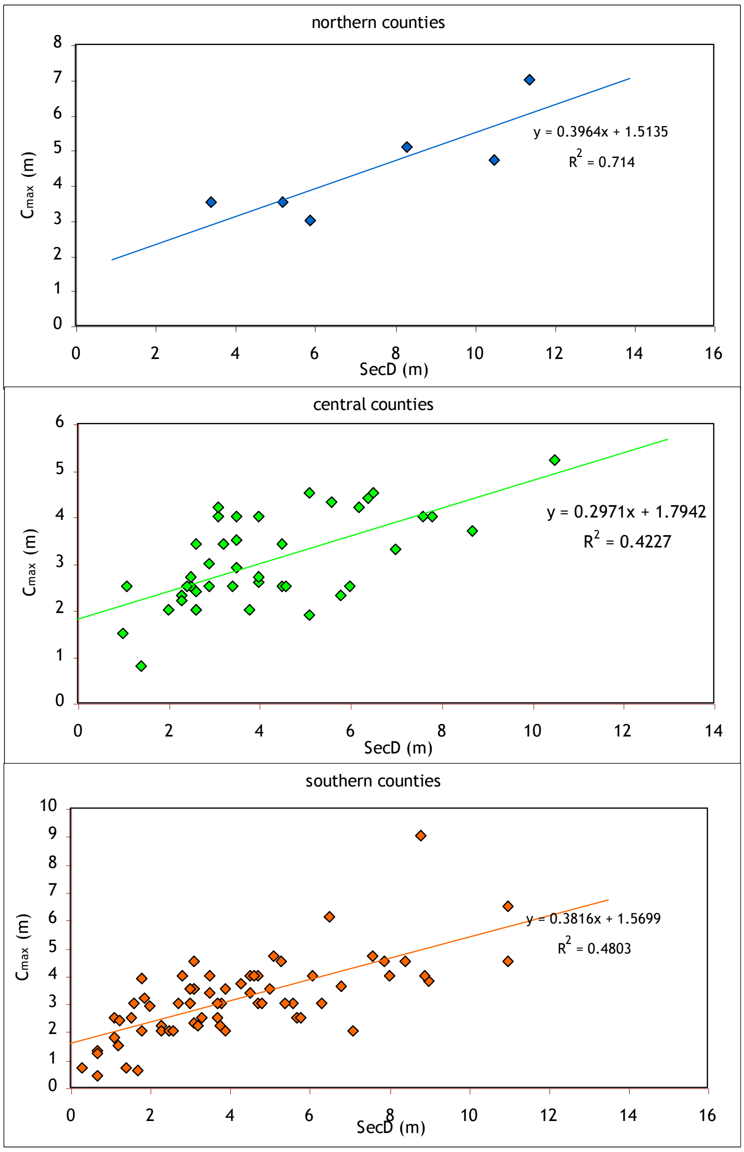


Figure B.2 Linear regressions between Secchi depth and C_{max} for Norwegian lakes, by region. Lakes with C_{max} dominated by nymphaeids, mosses, or Nitella were excluded. Northern counties: n=6, df=4, p=0.034; central counties: n=43, df=41, p<0.0001; southern counties: n=67, df=65, p<0.0001.

Similarly, the C_{max}–Secchi relationship remained significant when lakes with C_{max} dominated by angiosperms (elodeids and/or isoetids) were analyzed separately from lakes with C_{max} dominated by charophytes (excluding *Nitella*) (Figure B.3). The regression model for charophytes was highly significant, more so than for angiosperms, again likely reflecting less variability in lake conditions for lakes with C_{max} dominated by charophytes. We expect the C_{max} for charophytes to be easier to determine in the field. In further analysis the angiosperms will be divided into isoetids and elodeids, while *Nitella* spp and *Chara* spp will be separated . This may explain some of the differences between angiosperms and charophytes. However, the slopes for angiosperm- and charophyte-dominated C_{max} remained statistically similar (ANCOVA: pooled t=0.878, df=113, p=0.382). Such trends were confirmed when Norway's lakes were divided by region and macrophyte type at C_{max} (Table B.3 and Table B.4).

Table B.4 Linear regressions between Secchi depth and C_{max} for Norwegian lakes by region and alkalinity category. Lakes with C_{max} dominated by nymphaeids, mosses, or Nitella were excluded. Regressions and slope comparisons (t-test-based ANCOVAs) were performed only for datasets with n≥ 3.

	Northern		**Central**		**Southern**		**Whole Norway**	
	High	**Low**	**High**	**Low**	**High**	**Low**	**High**	**Low**
n	6	-	35	4	45	15	**86**	**19**
df	4	-	33	2	43	13	**84**	**17**
R^2	0.7140	-	0.4531	0.4383	0.5679	0.3483	**0.5388**	**0.5532**
p	0.034	-	<0.0001	0.338	<0.0001	0.021	**<0.0001**	**0.0003**
slope	0.3964	-	0.3089	0.2358	0.5306	0.2751	**0.4158**	**0.3276**
			t=0.274	df=35	t=5.380	df=56	**t=3.066**	**df=101**
			p=0.785		P<0.001		**p=0.003**	

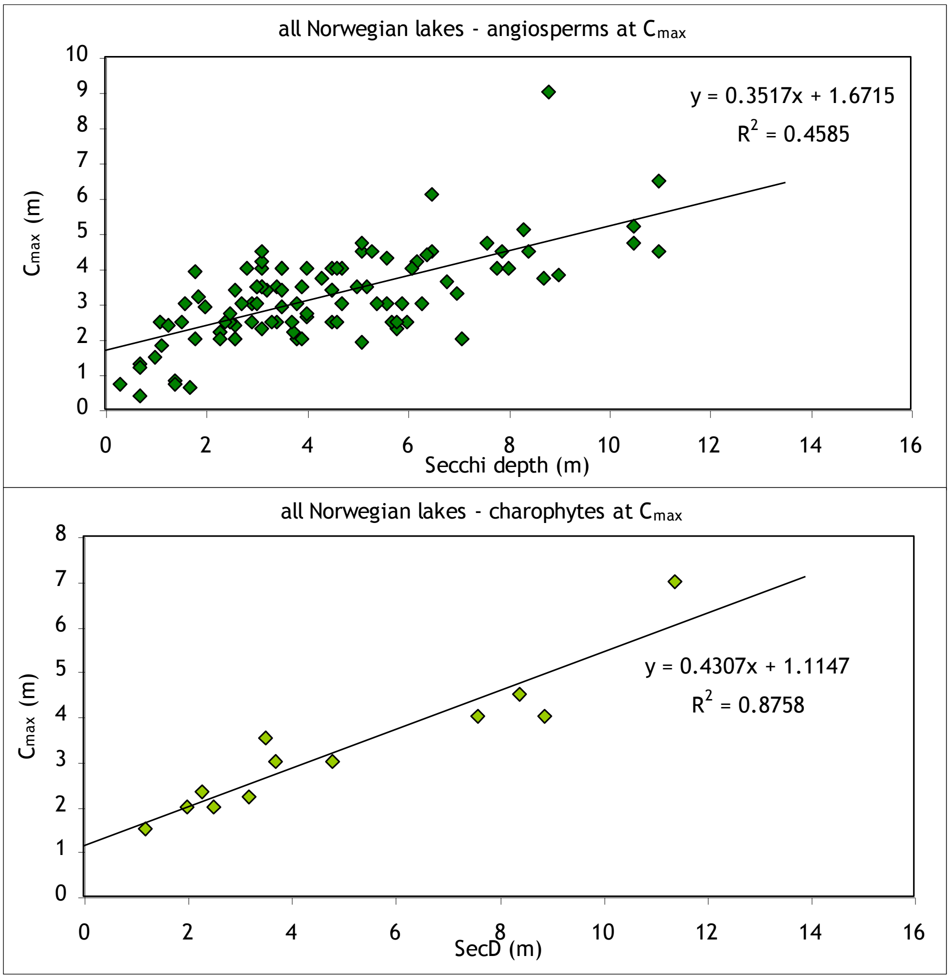
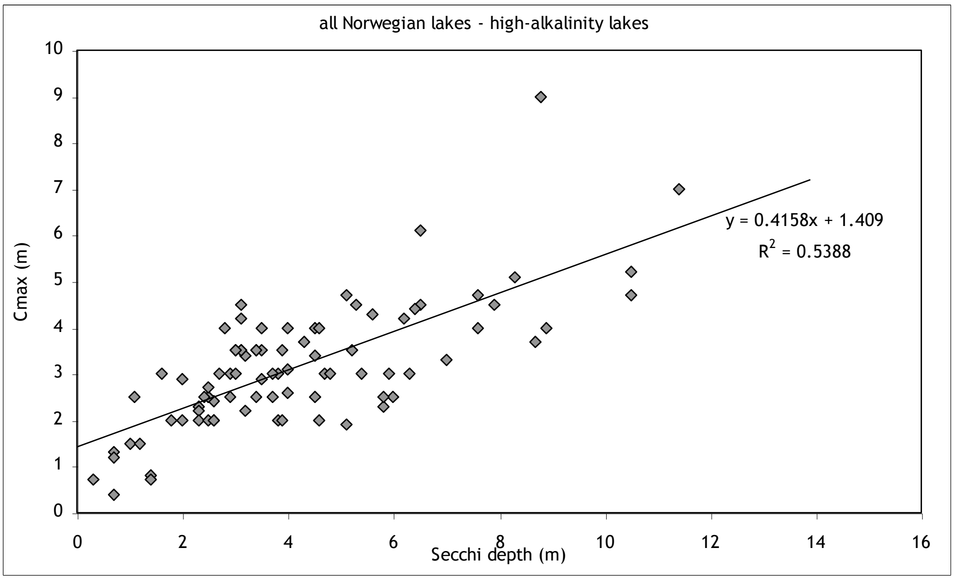


Figure B.3 Linear regressions between Secchi depth and C_{max} for Norwegian lakes, by macrophyte type at C_{max}. Lakes with C_{max} dominated by nymphaeids, mosses, or Nitella were excluded. Angiosperm-dominated C_{max}: n=105, df=103, p<0.0001; charophyte-dominated C_{max}: n=12, df=10, p<0.0001.

The C_{max}–Secchi relationship was highly significant for both low alkalinity lakes (lake types 001,002,101 and 102, cfr. Table B.2) and higher alkalinity lakes (lake type 201, 202, 301 and 302) (Figure B.4 and Table B.4).



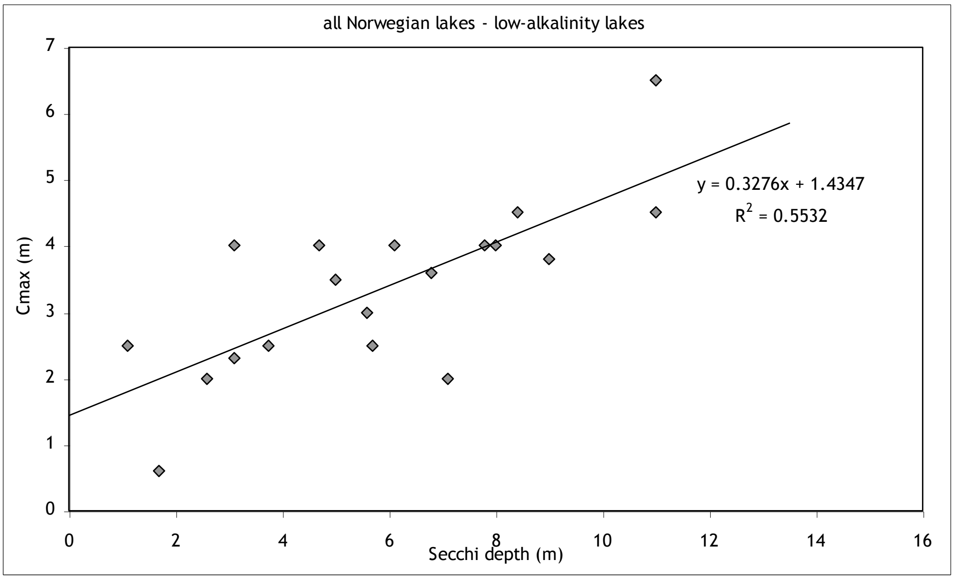


Figure B.4 Linear regressions between Secchi depth and C_{max} for Norwegian lakes, by lake type. Lakes with C_{max} dominated by nymphaeids, mosses, or Nitella were excluded. High-alkalinity lakes: n=86, df=84, p<0.0001; low-alkalinity lakes: n=19, df=17, p=0.0003.

Correlations with pressure: comparing TIc and C_{max}

The regressions with Secchi depth were different for TI_c (which displayed a logarithmic relationship: $R^2_{log} > R^2_{lin}$) and C_{max} (for which the relationship was linear: $R^2_{lin} > R^2_{log}$). When taking such differences into consideration, R^2_{log} for TI_c (0.5299) was > R^2_{lin} for C_{max} (0.4599): the relationship with Secchi depth therefore is stronger for TI_c than for C_{max}. The different type of relationship, however, makes direct quantitative comparisons difficult (or at least not straightforward).

For TP, both the TI_c and the C_{max} relationships were better explained by a (negative) logarithmic than a linear model. The relationship was stronger for TI_c than for C_{max} also for TP (R^2_{TIc}=0.5931; R^2_{Cmax}=0.1872). Removing an "outlier" with TP=798 (Søylandsvatn) did not improve R^2 for C_{max}.

In conclusion, TI_c appears to be a better descriptor/predictor of lake trophic state than C_{max} whether trophic state is based on Secchi depth or TP concentration. These differences between TI_c and C_{max} should be "real", as the inclusion of only no-nymphaeids, no-*Nitella* lakes with Secchi depth < bottom decreases the degree of environmental variability. Lakes with Secchi depth to bottom make, however, a small part of the total dataset (*n*~10, depending on the completeness of the subdataset), so that the analysis can be considered sufficiently representative of Norway.

The relationship between TI_c and TP for the total TI_c dataset (247 lakes, also including lakes without C_{max} data): high alkalinity lakes R^2=0.6133 and low alkalinity lakes: R^2=0.3475 (Mjelde, unpubl). So for the low alkalinity lakes, the C_{max} shows better correlation with TP. However, the comparison will also be done for the same dataset used in Table B.4.

Possible G/M boundary for C_{max}

As for the G/M boundary in the TI_c index, we use the sudden drop for isoetids/charophytes as an argument for the boundary. Based on the regression between TP and Secchi, and between Secchi and C_{max}, we suggest a preliminary G/M boundary for C_{max} at 2.7 m (corresponding to a Secchi depth at 3 m) for all lake types, all regions and all life forms.

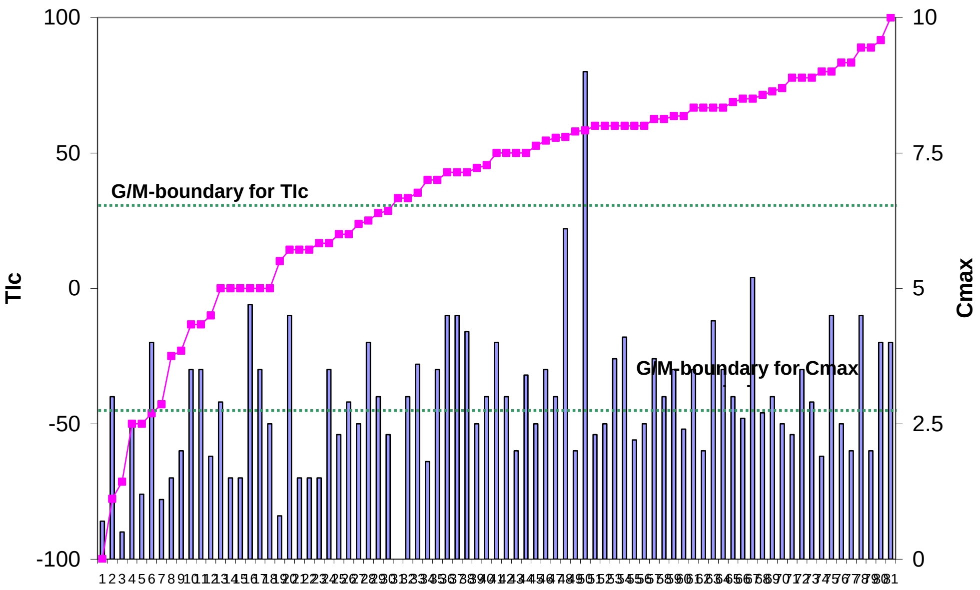


Figure B.5 Comparing G/M boundaries for TIc and C_{max} in high alkalinity lakes (201-302)

However, the status classes based on C_{max} seems very different from the status based on the TI_c index (Figure B.5).

The ability of the TI_c to discern differences in macrophyte-based ecological status regardless of differences in Secchi and independently of C_{max}, along with the unclear influence of C_{max} on TI_c, suggest that the incorporation of a factor measuring C_{max} into the TI_c would not enhance the descriptive or predictive performance of the TI_c, with the latter being a sufficiently robust and accurate measure of macrophyte-based lake trophic state, in its own right.

Conclusions

The maximum colonization depth (C_{max}) has been found to be associated with several lake variables, especially with Secchi depth (Secchi), in a number of independent investigations in temperate-climate Europe and North America. The C_{max}–Secchi relationship for Norwegian lakes has been investigated quantitatively using the extensive database gathered at NIVA. The C_{max} is closely, positively related with Secchi also in Norway. Because Secchi is also a measure of lake trophic state, a quantitative investigation was performed to see if incorporation of C_{max} and/or Secchi in the species composition index (TI_c) would be an additional variable.

However, despite the C_{max}–Secchi relationship, most regressions (R^2) for C_{max} appeared weaker than the same regressions for the TI_c index. Despite some well known systematic differences between the C_{max} of individual macrophyte taxathe C_{max}–TI_c relationship was also weak and uncertain, suggesting that C_{max} may need further, more detailed and/or extensive mathematical analysis before integration with the species composition index TI_c, or being added as a new metric to the macrophyte classification.

We do not know uncertainty in the different field methods. We assume that the uncertainty, especially for the "aquascope-rake"-method, can be large for some lakes, for instance in oligotrophic lakes with *Isoetes lacustris* at 5-7 m depth. The C_{max} for this species is difficult to determine with aquascope and rake. Such an uncertainty may also be specific for some macrophyte taxa and/or lake types. This has to be investigated further.

Based on the weaker correlation with pressure than the TI_c index and the uncertainty in field methods, we do not, at this point, recommend to include C_{max} in the macrophyte classification in Norwegian lakes. However, the TI_c in its current, straightforward formulation appears to be sufficiently robust and accurate to describe lake trophic state from macrophyte data. Its simple formulation and the relatively rapid assessment method needed to calculate it also make it particularly suitable for monitoring lake trophic state in Norway within the WFD context.

C. Examples of relationships between the land use and lake status in Swedish lakes

The unavailability of land use and land cover data in some member states makes it difficult and even impossible to incorporate land use as reference criteria. The N-GIG for macrophytes is of the opinion that the relationship between land use and macrophytes needs to be further studied prior setting any thresholds in land use for identifying reference lakes. The reasoning for this is threefold:

1. Preliminary analyses (here shown for Sweden, only) indicate that relationships between land use and tot-P (Figure C.1 and Figure C.2), the number of isoetid species (Figure C.3 and Figure C.4) and the trophic macrophyte index (Figure C.5 and Figure C.6) are scale dependent. A whole-catchment approach that considers land use in the entire catchment of the lakes is thus probably not appropriate for identifying reference lakes.

2. Most of the relationships between land use and water quality variables in the preliminary analyses were linear (Figure C.1 - Figure C.6) and did not allow for the identification of thresholds. As an exception, the number of isoetid species in relation to the percentage area of arable land (Figure C.3) and the area of arable land (Figure C.4) indicates indeed a threshold value. However, it needs to be evaluated if this result is also valid for the other N-GIG members.

3. Relating water quality variables to the percentage area of land use types does not consider the area of a) the lake catchments and b) the land use type under consideration. This might lead to misleading results. To incorporate this aspect in the analyses, the area of a certain land use might rather be related to the area of the lakes (Figure C.2, Figure C.4, Figure C.6). Also this aspect needs, however, to be studied in more detail.

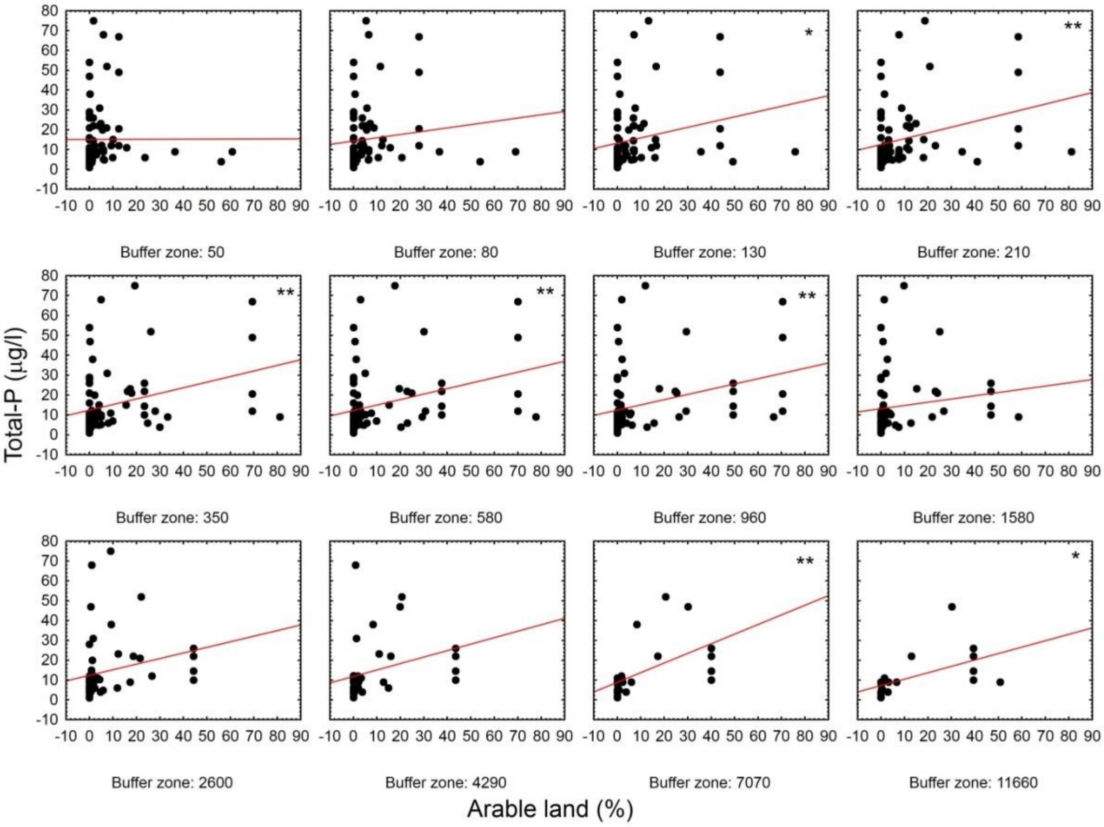


*Figure C.1 Relationship between the percentage of arable land in the lake catchments and total phosphorus divided by buffer zones (0 – 11660 m) from the lake shores. * p<0.05, ** p<0.01.*

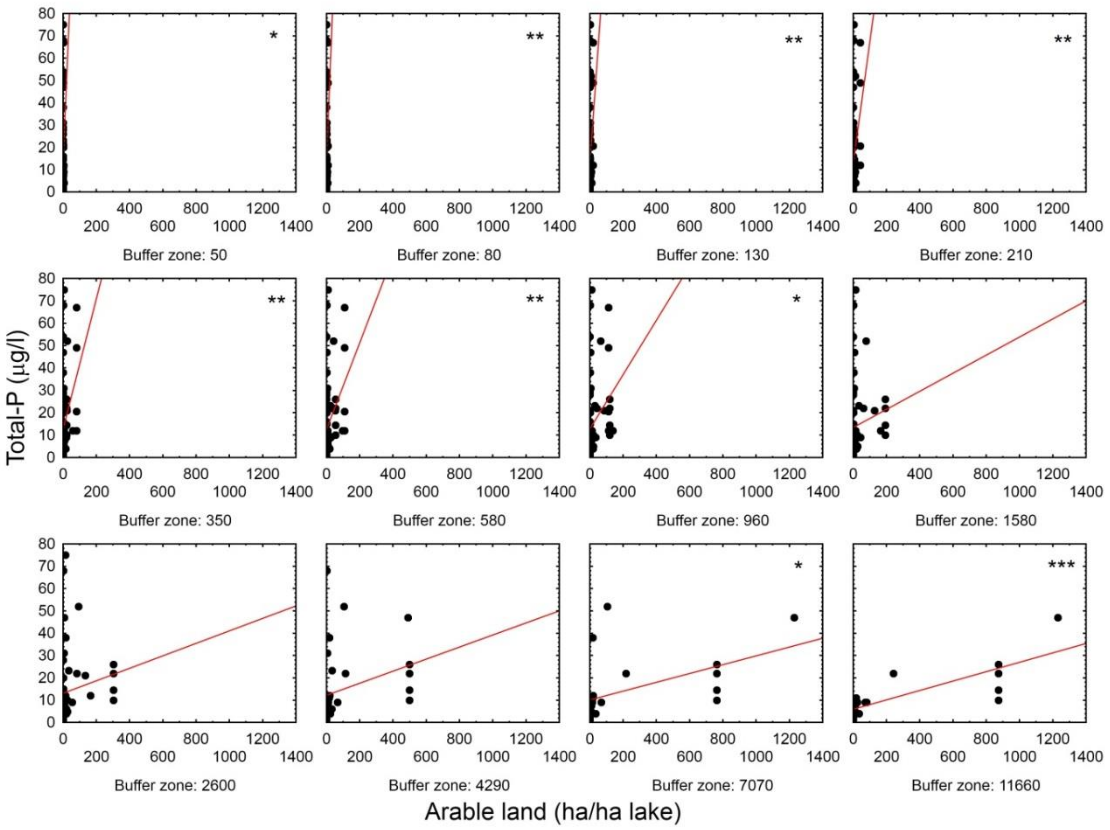


*Figure C.2 Relationship between the total area of arable land (per ha lake) in the lake catchments and total phosphorus divided by buffer zones (0 – 11660 m) from the lake shores. * p<0.05, ** p<0.01, *** p<0.001.*

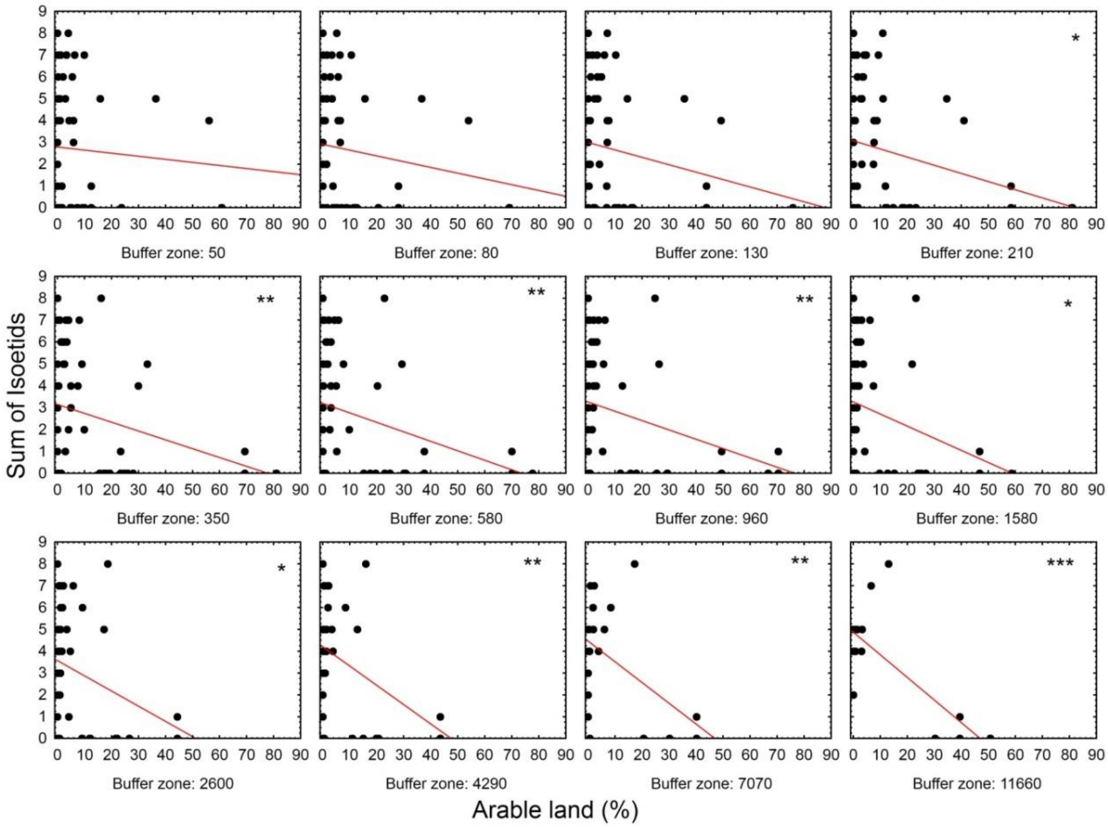


*Figure C.3 Relationship between the percentage of arable land in the lake catchments and the number of isoetids divided by buffer zones (0 – 11660 m) from the lake shores. * p<0.05, ** p<0.01, *** p<0.001.*

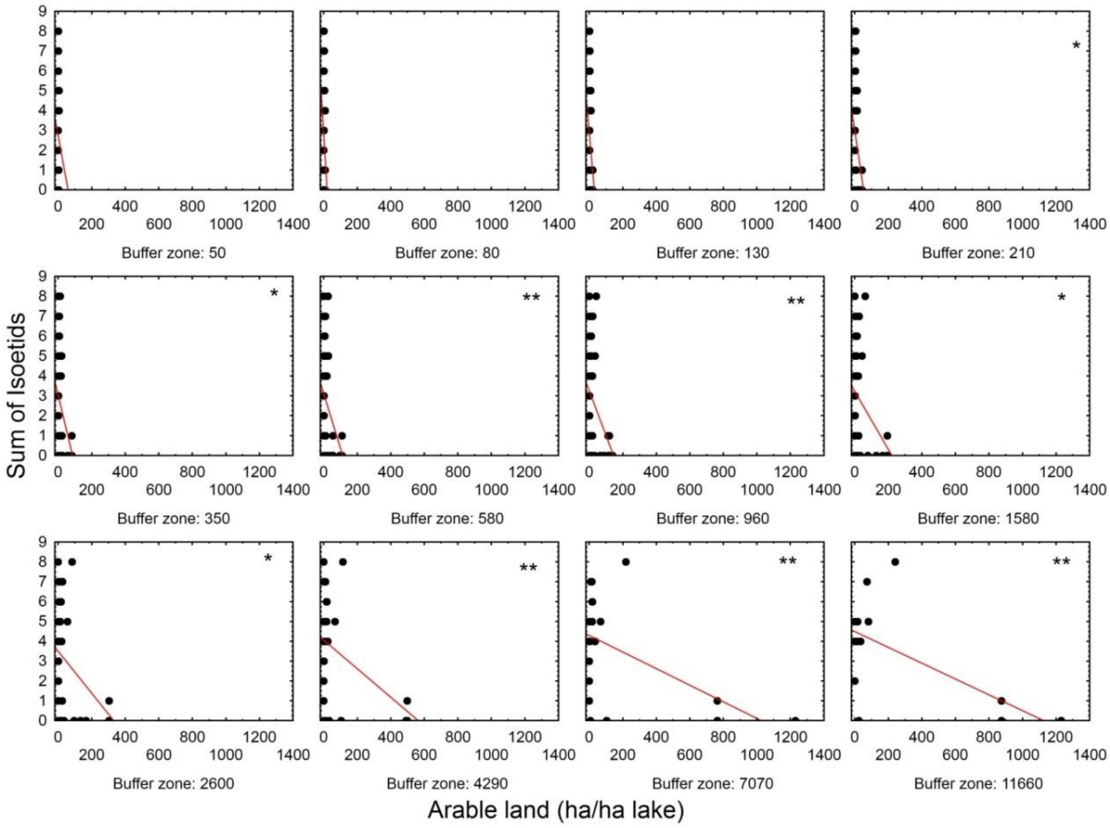


*Figure C.4 Relationship between the total area of arable land (per ha lake) in the lake catchments and the number of isoetids divided by buffer zones (0 – 11660 m) from the lake shores. * p<0.05, ** p<0.01.*

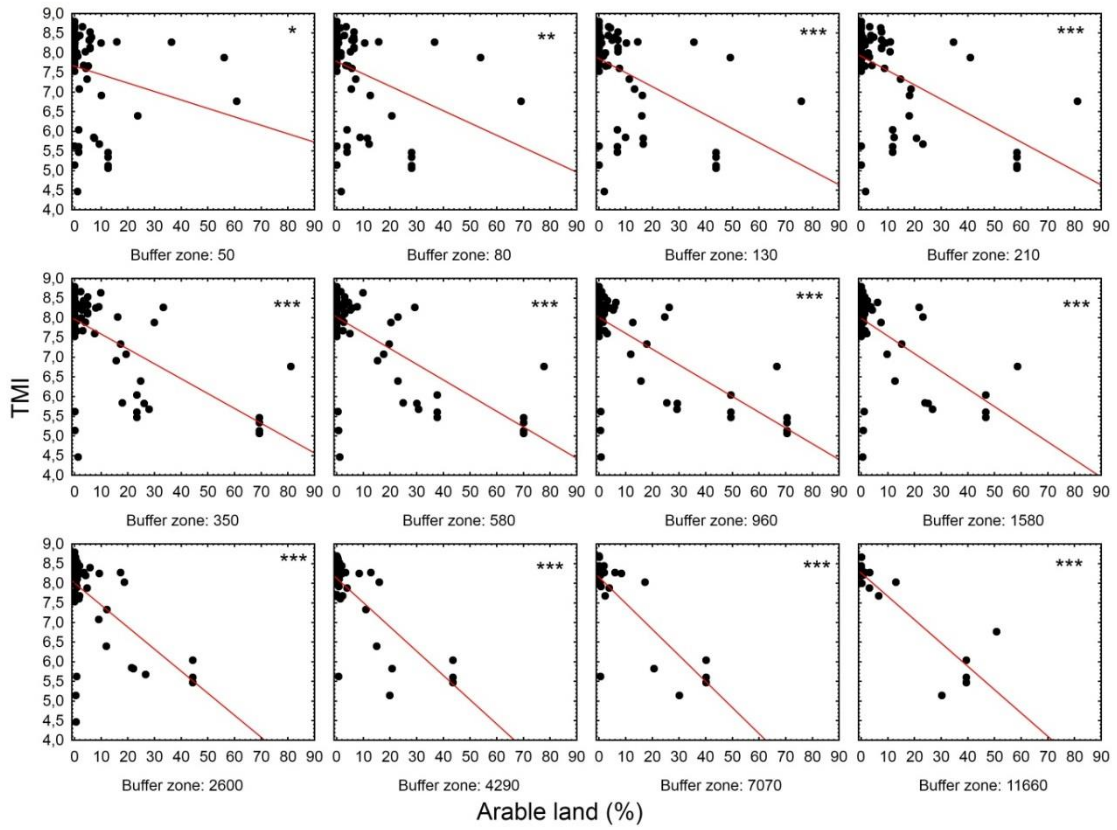


*Figure C.5 Relationship between the percentage of arable land in the lake catchments and the Trophic Macrophyte Index (TMI) divided by buffer zones (0 – 11660 m) from the lake shores. * p<0.05, ** p<0.01, *** p<0.001.*

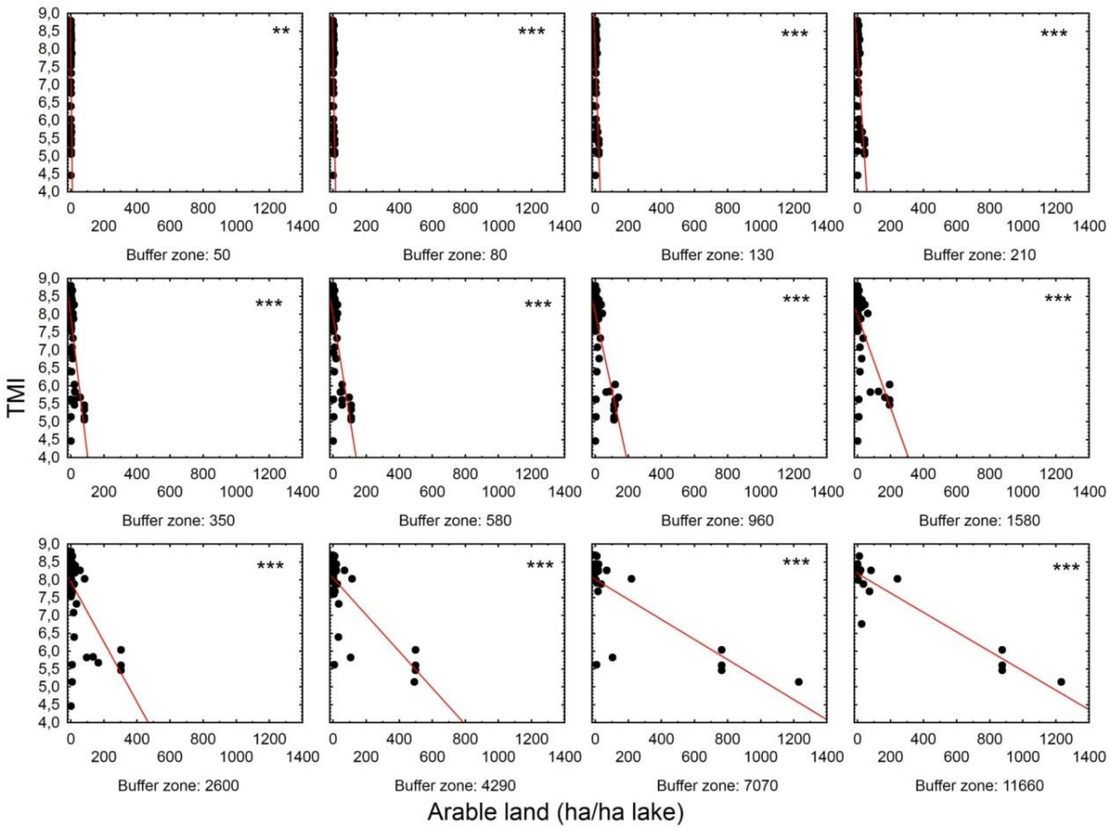


*Figure C.6 Relationship between the total area of arable land (per ha lake) in the lake catchments and the Trophic Macrophyte Index (TMI) divided by buffer zones (0 – 11660 m) from the lake shores. * p<0.05, ** p<0.01, *** p<0.001.*

D. Reference communities of different lake types of N-GIG.

Reference communities were analysed by using N-GIG dataset representing 420 reference lakes (original dataset submitted by member states).

Table D.1 Number of reference lakes per IC common type in the Northern Macrophyte GIG

IC type	101	102	201	202	301	302
N of ref lakes	160	119	50	54	31	6

A small number of lakes exist in the dataset more than once due to investigations in multiple years. Six lakes were without vegetation. These were removed from analysis.

Communities we analysed by using non-metric scaling (PC Ord version 5.0). Different lake type communities were clearly overlapping, but especially clear water low (101) and moderate alkalinity lakes (102) differed clearly from more humic lakes (102, 201) (Figure D.1). Macrophyte communities in high alkalinity lakes were different, but similarity in the species composition was lower within this lake type.

Analysis of country specific data showed that geographically distant countries like Ireland and Finland had most different macrophyte community (Figure D.2). Macrophyte communities in Nordic countries Finland, Sweden and Norway were quite similar.

Typical communities of reference lakes were analysed by using a simple type specific taxa approach where probability of species presence is calculated. In low alkalinity clear water lakes (101) the most common species, according to probability of presence (P) were the following:

- *Isoetes lacustris* 0.86
- *Lobelia dortmanna* 0.69
- *Sparganium angustifolium* 0.65
- *Myriophyllum alterniflorum* 0.6
- *Littorella uniflora* 0.53

All of these species are representing large isoetids, nymphaeids or elodeids favouring oligotrophic waters.

Low alkalinity humic lakes (102) are carrying some similar communities, although also small isoetids and soft bottom elodeids are common:

- *Isoetes lacustris* 0.79
- *Ranunculus reptans* 0.64
- *Isoetes echinospora* 0.63
- *Myriophyllum alterniflorum* 0.56
- *Lobelia dortmanna* 0.55
- *Sparganium angustifolium* 0.53
- *Subularia aquatica* 0.53
- *Potamogeton natans* 0.52

- *Potamogeton perfoliatus* 0.50

Moderate alkalinity with clear waters (201) favour species like *Potamogeton berchtoldii* and *P. alpinus*. High richness of *Potamogeton* species is very typical for such lakes. Only *Isoetes lacustris* has remained frequent from low alkalinity lake isoetid communities:

- *Myriophyllum alterniflorum* 0.8
- *Isoetes lacustris* 0.62
- *Potamogeton natans* 0.58
- *Potamogeton perfoliatus* 0.58
- *Potamogeton berthtoldii* 0.56
- *Potamogeton gramineus* 0.56
- *Rananculus reptans* 0.52
- *Sparganium angustifolium* 0.52
- *Potamogeton alpinus* 0.5

Increasing color in moderate alkalinity lakes (202) removes isoetid communities which cannot tolerate alkalinity or sedimentation. Vegetation consists typically of soft bottom elodeids and nymphaeids which favour soft bottom and can use apical or floating leaves for photosynthesis despite low transparency:

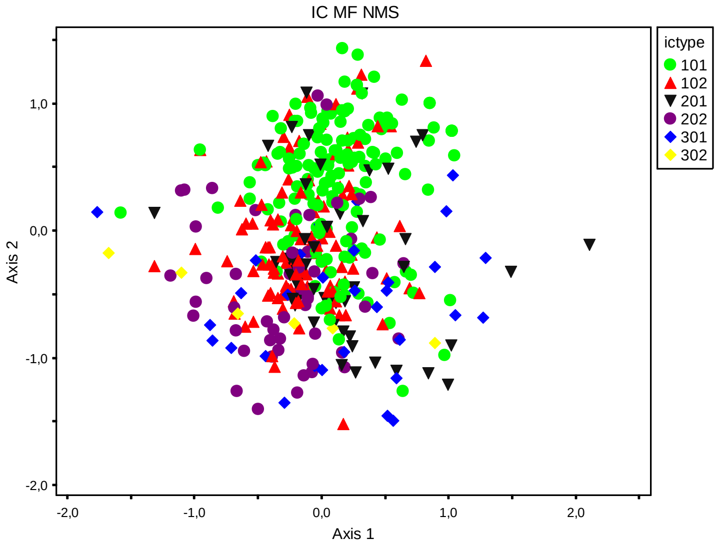
- *Potamogeton natans* 0.70
- *Potamogeton perfoliatus* 0.70
- *Myriophyllum alterniflorum* 0.59
- *Potamogeton alpinus* 0.57
- *Potamogeton gramineus* 0.56
- *Nymphaea alba* 0.56
- *Sparganium angustifolium* 0.5

The most common species in high alkalinity clear water lakes (301) are all elodeids although charids are also commonly present (P = 0.89). Diversity in charid species is large and therefore we cannot show any typical *Chara* species representing all reference lakes:

- *Potamogeton gramineus* 0.65
- *Potamogeton perfoliatus* 0.61
- *Potamogeton natans* 0.48
- *Myriophyllum alterniflorum* 0.45
- *Potamogeton filiformis* 0.45
- *Fontinalis antipyretica* 0.42

A higher amount of humic substances (302) does not bring any bigger changes in composition maybe due to the very low number of sites (6). *Potamogeton* species are dominating in this type:

- *Potamogeton filiformis* 0.83
- *Potamogeton natans* 0.67
- *Myriophyllum alterniflorum* 0.5
- *Potamogeton berchtoldii* 0.5
- *Sparganium angustifolium* 0.5
- *Potamogeton alpinus* 0.5



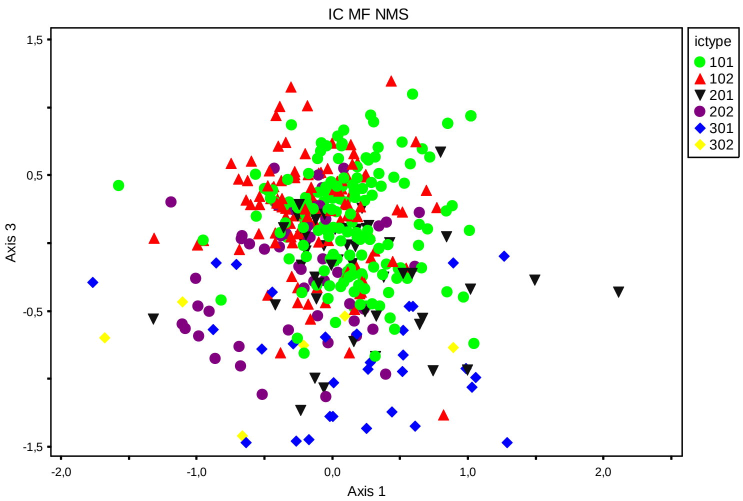
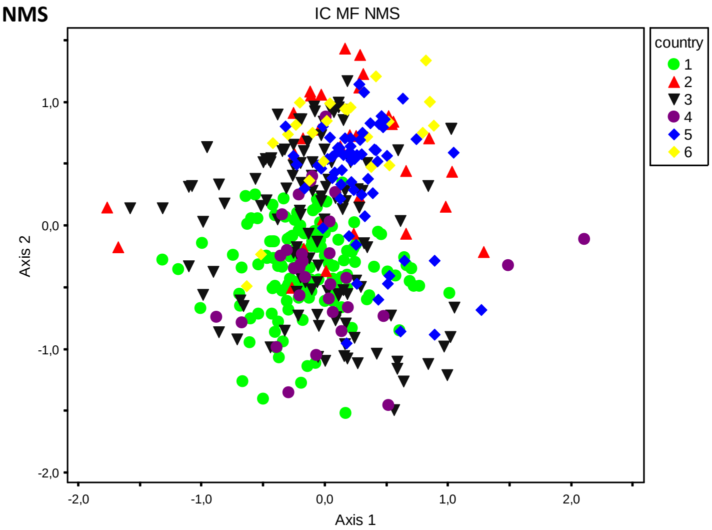


Figure D.1Distribution of the lake type specific macrophyte data along three NMS-dimensions.

NMS



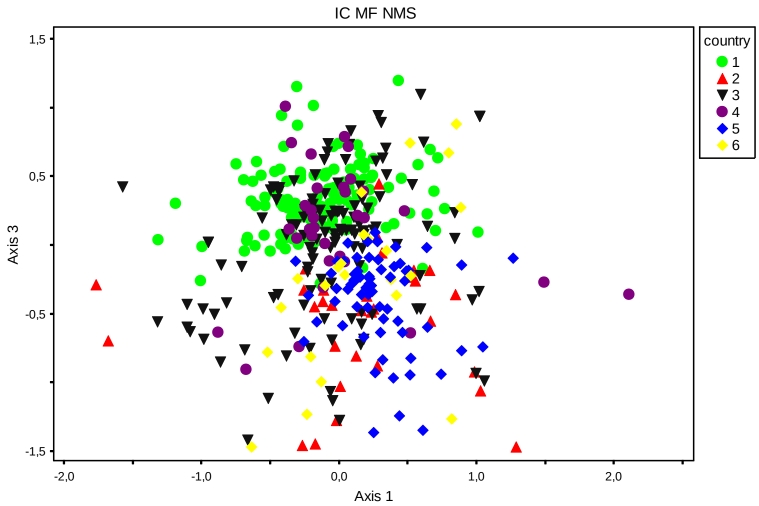


Figure D.2 Distribution of the country specific macrophyte data along three NMS-dimensions. 1 = Finland, 2 = Ireland, 3 = Norway, 4 = Sweden, 5 = UK, 6 = Northern Ireland

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Abstract

One of the key actions identified by the Water Framework Directive (WFD; 2000/60/EC) is to develop ecological assessment tools and carry out a European intercalibration (IC) exercise. The aim of the Intercalibration is to ensure that the values assigned by each Member State to the good ecological class boundaries are consistent with the Directive's generic description of these boundaries and comparable to the boundaries proposed by other MS.
In total, 83 lake assessment methods were submitted for the 2nd phase of the WFD intercalibration (2008-2012) and 62 intercalibrated and included in the EC Decision on Intercalibration (EC 2013). The intercalibration was carried out in the 13 Lake Geographical Intercalibration Groups according to the ecoregion and biological quality element. In this report we describe how the Intercalibration exercise has been carried out in the Northern Lake Macrophyte group.

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