A REFERENCE BASED TYPOLOGY AND ECOLOGICAL ASSESSMENT SYSTEM FOR IRISH LAKES.

PRELIMINARY INVESTIGATIONS

FINAL REPORT

(Project 2000-FS-1-M1 Ecological Assessment of Lakes
Pilot Study to Establish Monitoring Methodologies EU (WFD))

Prepared for the Environmental Protection Agency

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1. Introduction

The concept of water quality is now changing at a more rapid pace than ever before, being radically broadened by the publication of the EU Water Framework Directive (WFD) in December 2000 (CEC, 2000). The WFD has marked a turning point in the perception and management of water quality in Europe, establishing a framework for the protection of rivers, lakes, transitional waters, coastal waters and groundwaters.

The WFD is ambitious, stipulating that water quality must not deteriorate further and that all waters should be restored to good status by the end of 2015. This is to be achieved using a programme of measures which will be detailed in a river basin management plan prepared by river basin districts (the new unit of management of water quality).

The WFD has provided the impetus for this research project through its requirement for lakes to be assessed ecologically. Ecological monitoring programmes must be established before the end of 2006. The biological elements to be monitored are: phytoplankton, macrophytes and phytobenthos, benthic invertebrates and fish. The WFD has therefore radically shifted emphasis from chemical measures of water quality to those based on ecology. Chemical and physical components of water quality are still an integral part of assessment but are regarded as 'supporting elements' for the biology.

The steps that are necessary to produce a working ecological assessment system for lakes formed several of the objectives of this research. The main objectives were:

1) To collect detailed ecological information on up to 200 lakes, focusing on three of the WFD biological groups: phytoplankton, macrophytes and macroinvertebrates (from both littoral and profundal zones).
2) To produce a lake typology that is successful in partitioning biological variation in reference condition.
3) To describe type-specific reference conditions.

4) To develop WFD compliant classification tools that describe deviation from reference status across a pressure gradient. Only preliminary work on developing classification tools was within the scope of the project.

Objective 1: Collection of ecological information

The starting point, and the largest and most formidable task of the project was the collection of information on the ecology of 200 lakes. The biological elements that were investigated were phytoplankton, macrophytes and macroinvertebrates (from both littoral and profundal zones). The remaining biological elements required to be monitored by the WFD - phytobenthos and fish - were outside the scope of this study. The sampling and processing of the biological samples took more than three years of the four-year project and the resulting dataset is the most extensive on lake ecology in the Republic of Ireland.

When selecting the lakes to be sampled, one of the objectives was to ensure that there was sufficient representation of lakes that were close to their natural state or in reference condition. Reference condition may be thought of as a condition where there are "no, or only very minor, anthropogenic alterations…[relative to] undisturbed conditions" (Annex V: 1.2 of the WFD). The distribution of the lakes, frequency of sampling, and summary physical and chemical characteristics are presented in Chapter 2.

Objectives 2 and 3: Development of a lake typology and the description of reference conditions

In developing a system to ecologically assess lakes it must be considered that there are different types of lakes, which will have different flora and fauna in their natural state or reference condition. The WFD requires that ecological quality is measured as deviation away from reference condition for each lake type. The purpose of a typology is to allow ecological change, caused by anthropogenic pressure, to be detected more easily (REFCOND, 2003). A typology should therefore ensure that natural differences between lakes are clearly distinguished from those caused by anthropogenic pressure.

The WFD allows member states to define their lake typology using one of two systems: system A or system B (Table 1.1). System A is a fixed typology in that the parameters to be used and the values of their boundaries are set out in Annex II of the WFD. Most member states have opted to use the alternative System B, which, alongside several obligatory factors, allows more choice in the parameters used and in where the boundaries are set (Table 1.1). The proviso is that use of system B must result in at least the same degree of differentiation as would be achieved if system A had been used. EU discussion on the WFD has indicated that the list of factors provided in system B is not exhaustive and that additional factors may be used to define a typology if appropriate (REFCOND, 2003). Moreover, a simple typology - with a lower number of types than would be produced using System A only - may be used once it achieves the same degree of differentiation as system A.

Table 1.1 Parameters that may be used to define types using system A or B (CEC, 2000). Obligatory factors are in bold.

WFD typology factors	**System**	
Ecoregion	**A**	
Latitude		**B**
Longitude		**B**
Altitude	**A**	**B**
Mean depth	**A**	B
Depth		**B**
Lake area	**A**	**B**
Geology	**A**	**B**
Mean air temperature		B
Air temperature range		B
Acid neutralising capacity		B
Residence time		B
Mixing characteristics/Stratification		B
Background nutrient status		B
Lake shape		B
Mean substratum composition		B
Water level fluctuation		B

The objective of this project was to develop a lake typology that is biologically relevant, i.e. a typology defined by environmental boundaries that is demonstratively successful in partitioning natural variation in the biology. The first step was to select a set of lakes thought to be in reference condition that contain representatives of the types of lakes in Ireland. The second step was to examine each of the biological elements to see if 'biological' types were evident using multivariate grouping techniques. Such 'biological' types were then examined in terms of potential typology factors (e.g. Table 1.1) to see if they were also distinct in terms of environmental variables. This led to a working definition of what a lake type is:

A type is a group of lakes that, in reference condition, have a unique composition or abundance of flora or fauna that is related to a distinct combination of environmental factors for that group.

The next step was to define environmental boundaries for each of the types that are effective in partitioning the biological variation in reference condition. The typology was initially developed at biological element level (e.g. macrophytes or phytoplankton) as this is the scale at which assessment systems will work, acknowledging that many typological factors may be of greater relevance to one group over another. For example, residence time is likely to be of greater relevance to phytoplankton than littoral invertebrates. Following this, the proposed types for each biological element were examined and an overall lake typology was prepared to broadly categorise lake types in Ireland.

A sensible balance needs to be achieved so that a typology has just enough classes to adequately describe natural biological variation (Karr and Chu, 1999). The aim should be to provide enough classes for ecological assessment metrics to work effectively at detecting anthropogenic pressure. In addition, Karr and Chu (1999) also advocate developing a typology that is effective at partitioning biological response to pressure. An alternative approach is to develop a site-specific typology, with reference conditions determined by probabilistic assignment to several types such as in RIVPACS (e.g. Clarke *et al*., 2003).

Following the development of a typology, the third objective was to describe type-specific reference conditions. This description is necessary as it is to be used in ecological assessment, which is to be based on deviation away from reference condition for each type. The description may be achieved in two ways. The first is a general description of the composition and abundance of taxa for each of the biological elements and the second is by using a summary statistic such as the mean for the assessment metric used.

One underlying premise in the above approach is that lakes are available that are in reference condition. The reference lakes used in this study were selected by expert opinion using information on lake catchments including underlying geology and land-use, water chemistry and existing biological information. Effort was made to achieve a selection of lakes that corresponded to the WFD definition of reference condition (see above) rather that select lakes that represented the 'best available'. The danger of including the 'best available' lakes is that management targets, such as achieving good status by the end of 2015, may be confused by setting reference condition too low by using lakes that have been significantly affected by anthropogenic pressure. The approach used here resulted in a somewhat unbalanced representation in the reference lakes, with more soft-water (< 20 mg l^{-1} $CaCO_3$) and hard water lakes (> 100 mg l^{-1} $CaCO_3$) than representatives of the moderate alkalinity band where there are fewer existing examples of lakes in reference condition.

An additional project funded by the EPA, (IN-SIGHT ERTDI Project # 2002-W-LS/7) using palaeolimnological techniques is running concurrently to confirm if the lakes are in reference status. In addition to the objectives outlined, the current project also aimed to develop metrics that measure ecological response across a pressure gradient. Some confidence in assignment of reference status may be provided if the preliminary ecological assessment techniques indicate that the selected reference lakes are in high status.

Objective 4: Development of WFD compliant classification tools

The WFD requires that classification tools or metrics are developed that measure ecological quality for each biological element. Ecological quality is to be expressed as deviation away from reference conditions in the form of an ecological quality ratio

(EQR). One recommended option is to calculate an EQR by dividing an observed metric value by the reference metric value for its lake type (REFCOND, 2003). This yields an EQR that ranges from near 1 (high/reference status) to near 0 (bad status). The overall EQR for a lake is assigned using the lowest EQR recorded for each of the biological elements and the supporting physical and chemical elements. Figure 1.1 shows a summary of the steps necessary for assignment of an EQR.

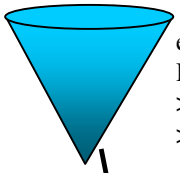
The key step necessary for the WFD EQR system to work is the development of ecological assessment tools or metrics for each of the biological elements that successfully describe deviation from reference condition along a pressure gradient. Currently, there are no widely accepted ecological assessment tools for any of the biological elements. This was the fourth objective of this project, to carry out developmental work on ecological assessment tools for the biological elements: phytoplankton, macrophytes, littoral macroinvertebrates and profundal macroinvertebrates. Metrics may be developed to work along one or more pressure gradients. This project focused on nutrients, i.e.total phosphorus, as the main pressure affecting Irish lakes.

This project examined a number of ecological assessment approaches depending on their suitability to the biological element under consideration. These included multimetric indices, published indices, simple empirically-based indices and multivariate classification. The different methods are described in the relevant chapters.

The aim of this work is to better position Ireland to meet its commitments to assess lakes ecologically as required by the WFD. The successful ecological assessment of lakes was recognised at an early stage by the Irish EPA to be crucially dependent on research. This is the second EPA commissioned research project on the ecological assessment of lakes: the first by Irvine *et al*. (2001) was commissioned in 1995. Effective ecological assessment is an enormous challenge that will necessitate constant refinement, through research, for many years to come.

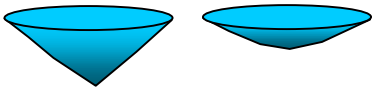
1. Define a typology that partitions biological variation in reference condition

Lake type A



e.g.
High Alkalinity,
> 4 m mean depth,
> 50 ha area

Lake type B……..C etc

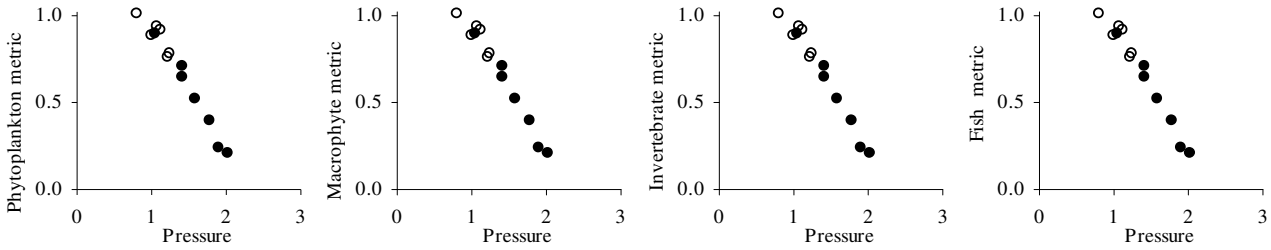


2. Identify and describe <u>type-specific</u> reference conditions

Description of reference condition for lake type A using metric values for the biological elements.

Biological element	Phytoplankton	Macrophytes	Phytobenthos	Littoral invertebrates	Profundal invertebrates	Fish
Metric value	0.95	0.94	24	0.93	1000	0.95

3. Apply assessment metric to measure deviation away from type-specific reference status for each biological element.



4. For each lake, the Ecological Quality Ratio (EQR) is calculated for each element as: observed / reference metric value. Lowest EQR is used for assessment (including a physico-chemical EQR – not shown).

EQR for Lake X. Scale may range from high status: near 1, to bad status: near 0.

Biological element	Phytoplankton	Macrophytes	Phytobenthos	Littoral invertebrates	Profundal invertebrates	Fish
EQR	0.71	**0.64**	0.69	0.74	0.73	0.80

Figure 1.1 Main steps required by WFD for assignment of an Ecological Quality Ratio (simplified).

2. Overview of lakes sampled

2.1 Introduction

This chapter introduces the lakes sampled and provides details on some of their physico-chemical and hydro-morphological characteristics. In total 201 lakes across the country were sampled between 2001 and 2003. These include a large proportion of the lakes in the country over 50 ha surface area (required by the WFD) as well as a selection of smaller lakes principally located in the midlands and along the western seaboard. Figure 2.1 shows the location of the lakes sampled. Grid references for the locations of the lakes are listed in Table 2.1. There are fewer lakes in the east, south-east and south of the country which accounts for the absence or low number of lakes studied in these areas.

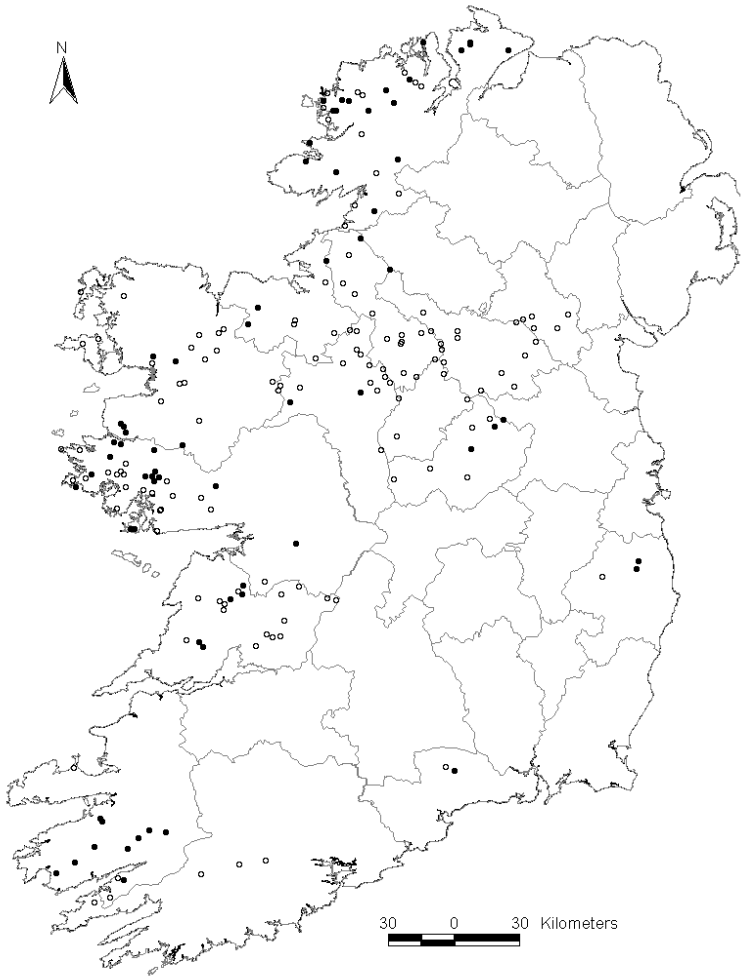


Figure 2.1 Location of the lakes sampled. Candidate reference lakes are indicated by solid circles.

2.2. Frequency of sampling

The lakes sampled and the dates they were sampled on are listed in Table 2.2. Details of sampling methodology and the biological elements collected can be found in Chapter 3. In Spring 2001, 161 lakes were sampled. These lakes were selected primarily on the basis of size and all, apart from four lakes, had a surface area greater than 50 ha. During the Summer of 2001, 98 of these lakes were sampled again along with an additional 13 lakes. Sampling in 2002 concentrated solely on lakes which were potential reference candidates. Lakes were assessed and chosen on the basis of Geographical Information Systems (GIS) mapping and historical chemical and biological results. In Spring 2002, 69 potential reference lakes were sampled, which included 42 lakes that had been sampled the previous year and an additional 27 lakes. Of these 69 lakes 40 were sampled again during the Summer of 2002. In the Summer of 2003, 57 lakes were sampled. All of these lakes had been sampled once in Spring 2001. As time limitations did not permit them to be sampled in Summer 2001, they were sampled in Summer 2003. Thus, the vast majority of the 201 lakes have been sampled in both Spring and Summer.

2.3 Physical characteristics of the lakes

GIS software and a digital elevation model were used to delineate catchments upstream from each lake outflow and to calculate lake and catchment areas and perimeters. Altitude data and the location of the lakes, recorded as Irish Grid Reference, were obtained from the Ordnance Survey "Discovery" Series 1:50 000 scale maps. These data are presented in Table 2.1 for each of the lakes sampled. The National Lake Code is a unique code for each lake in the country and incorporates a Hydrometric Area code, a River Catchment code and a Lake number generated by GIS distance ordering functions based upon a lakes distance from the river mouth.

Table 2.1 Physical characteristics of lakes

Lake Code	Lake name	Co.*	National Grid Ref.	Altitude meters	Lake area km^2	Catchment area km^2
28-00154-0110-000	Acrow	CE	R 193 687	195	0.06	0.30
31-000r4-0260-000	Ahalia North	GY	L 965 390	2	0.39	46.25
38-00016-0400-000	Aleck More	DL	B 762 080	10	0.61	7.66
25-0155b-0350-000	Alewnaghta	CE	R 759 911	31	0.55	26.25
26-0155a-3000-000	Allen	LM	G 960 200	47	33.56	403.35
19-00228-0240-000	Allua	CK	W 190 655	84	1.36	109.28
32-t4_32-0760-000	Anaserd	GY	L 606 445	8	0.87	2.08
26-0155a-1060-000	Annaghmore	RN	M 900 837	46	0.53	3.96
38-00022-0060-000	Anure	DL	B 820 165	36	1.59	37.29
31-000r4-1120-000	Arderry	GY	L 995 457	37	0.81	14.42
35-00116-0360-000	Arrow	SO	G 790 109	53	12.47	65.76
27-00158-0980-000	Atedaun	CE	R 295 885	13	0.38	176.63
25-0155b-0370-000	Atorick	CE	R 630 965	136	0.98	28.92
32-000u4-0240-000	Aughrusbeg	GY	L 555 582	8	0.50	1.69
36-00123-5280-000	Avaghon	MN	H 690 135	127	0.54	3.96
31-00136-0290-000	Ballinahinch	GY	L 765 480	7	1.70	155.23
30-00143-0100-000	Ballycuirke	GY	M 230 315	6	0.74	33.45
27-00158-0770-000	Ballycullinan	CE	R 288 857	20	0.29	10.74
27-00158-1460-000	Ballyeighter	CE	R 357 940	17	0.28	3.17
34-00110-0220-000	Ballymore	MO	G 290 128	15	0.56	27.25
32-00004-0120-000	Ballynakill	GY	L 640 580	8	0.62	7.70
31-000r4-2040-000	Ballynakill (G)	GY	L 865 222	13	0.24	1.41
07-00159-0930-000	Bane	WH	N 550 712	112	0.75	4.70
21-00216-0020-000	Barfinnihy	KY	V 850 768	249	0.14	0.79
38-00048-0490-000	Barra	DL	B 935 120	90	0.63	19.68
35-00117-0170-000	Belhavel	LM	G 880 290	60	1.01	22.24
32-00108-0140-000	Beltra	MO	M 070 980	15	4.10	96.53
26-0155a-1640-000	Boderg	LD	N 015 910	39	5.11	
30-00143-1040-000	Bofin	GY	M 035 440	40	0.92	21.90
26-0155a-1450-000	Bofin	LD	N 040 885	39	2.58	
36-00123-1490-000	Brackley	CN	H 190 205	58	1.67	17.43
27-00250-0460-000	Bridget	CE	R 565 807	30	0.55	4.70
27-00158-1760-000	Bunny	CE	R 375 967	17	1.03	9.32
22-00208-0020-000	Caragh	KY	V 725 905	15	4.91	162.54
30-00143-1690-000	Carra	MO	M 180 710	20	15.64	108.45
19-00228-0070-000	Carraigadrohid	CK	W 360 702	60	5.77	618.97
35-00117-0110-000	Carrigeencor	LM	G 830 336	45	0.44	2.63
34-00110-0230-000	Carrowkeribly	MO	G 266 110	8	0.51	34.28
33-00105-0030-000	Carrowmore	MO	F 836 283	7	9.28	87.95
26-0155a-2100-000	Cavetown	RN	M 830 972	82	0.64	9.06
21-00220-0030-000	Clonee	KY	V 810 640	26	0.71	27.02
27-00250-0280-000	Clonlea	CE	R 510 735	26	0.40	5.91
22-00208-0090-000	Cloon	KY	V 702 777	90	0.77	17.19
31-00138-0130-000	Cloonacleigha	SO	G 610 150	57	0.62	32.63
35-00116-0280-000	Cloonadoon	GY	M 008 315	29	0.49	43.56
26-0155a-2210-000	Cloonagh	RN	M 545 870	81	0.71	47.93
21-00213-0090-000	Cloonaghlin	KY	V 610 707	109	1.28	10.24
32-t4_32-0520-000	Cloongat	GY	L 689 472	14	0.08	6.06
34-00110-0430-200	Conn	MO	G 180 100	9	48.48	416.48
26-0155a-0120-000	Coosan	WH	N 055 450	35	0.56	4.18

Table 2.1 (cont.) Physical characteristics of lakes

Lake Code	Lake name	Co.*	National Grid Ref.	Altitude meters	Lake area km^2	Catchment area km^2
36-00123-2860-000	Corglass	CN	H 346 089	45	0.34	1.17
30-00143-0570-000	Corrib	GY	M 250 415	6	172.91	2954.93
26-0155a-2010-000	Corry	RN	M 945 965	41	1.54	
16-00182-0210-000	Coumduala	WD	S 293 143	468	0.02	0.16
38-00016-0200-000	Craghy	DL	B 795 115	14	0.50	25.22
33-000k5-0160-000	Cross	MO	F 645 296	3	1.11	3.37
16-00182-0150-000	Crottys	WD	S 326 125	419	0.04	0.08
27-00158-1190-000	Cullaun	CE	R 315 905	16	0.50	13.20
27-00158-0750-000	Cullaunyheeda	CE	R 485 745	27	1.55	22.99
34-00110-0670-000	Cullin	MO	G 230 030	9	10.24	802.81
21-00213-0010-000	Currane	KY	V 530 660	6	10.35	103.98
29-00146-0010-000	Cutra	GY	R 475 985	33	3.88	120.95
10-00171-0070-000	Dan	WW	O 150 040	200	1.03	63.18
01-00063-0340-000	Derg	DL	H 080 745	140	8.81	35.14
25-0155b-0450-000	Derg	TN	R 800 900	30	122.20	3958.81
26-00157-0370-000	Derravaragh	WH	N 410 680	61	9.14	559.23
36-00123-2530-000	Derrybrick	CN	H 345 120	45	0.36	1.54
36-00123-1910-000	Derrycassan	CN	H 225 118	45	0.71	309.87
31-00136-0470-000	Derryclare	GY	L 825 485	10	2.24	111.73
34-00110-1110-000	Derryhick	MO	M 205 990	25	0.54	9.06
28-00152-0050-000	Doo	CE	R 120 720	83	1.30	23.11
39-00008-0010-000	Doo	DL	C 359 394	283	0.09	0.43
32-00130-0020-000	Doo	MO	L 833 682	30	1.55	27.52
36-00123-4630-000	Dromore	CN	H 610 161	79	0.61	213.60
26-0155a-2290-000	Drumharlow	RN	G 905 015	40	2.79	
36-00123-1800-000	Drumlaheen	LM	H 090 070	65	0.74	2.34
36-00123-4750-000	Drumlona	MN	H 640 175	77	0.53	20.34
27-00250-0330-000	Dúin	CE	R 545 736	22	0.49	36.67
38-00016-0070-000	Dunglow	DL	B 782 117	13	0.61	37.67
	Dunlewy	DL	B 915 194	61	1.10	35.96
37- a6_37-0010-000	Durnesh	DL	G 878 693	0	0.70	15.10
35-00114-0150-000	Easky	SO	G 442 225	180	1.19	10.80
36-00123-5970-000	Egish	MN	H 795 132	161	1.17	8.72
25-0155b-0950-000	Ennell	WH	N 390 460	81	11.56	117.62
26-0155a-2140-000	Errit	RN	M 540 851	83	0.82	7.15
37-00058-0060-000	Eske	DL	G 972 837	27	3.87	79.65
40-0000c-0020-000	Fad (east)	DL	C 539 393	233	0.12	0.61
40-00004-0010-000	Fad (west)	DL	C 397 427	125	0.40	3.20
32-t4_32-0210-000	Fadda	FY	L 667 455	13	0.47	4.40
32-00132-0020-000	Fee	GY	L 790 613	47	1.74	15.76
32-00107-0070-000	Feeagh	MO	F 965 000	11	3.95	84.68
39-00031-0080-000	Fern	DL	C 180 230	20	1.81	206.17
32-00130-0010-000	Fin	MO	L 841 657	28	0.14	30.96
27-00250-0100-000	Finn	CE	R 435 695	27	0.74	3.40
01-00062-0210-000	Finn	DL	B 910 015	132	1.15	10.61
26-0155a-0930-000	Forbes	LD	N 080 815	37	2.98	
32-00107-0030-000	Furnace	MO	L 968 974	0	1.84	101.86
26-0155a-2260-000	Gara	RN	M 705 995	66	12.57	499.69
36-00123-1830-000	Garadice	LM	H 180 110	49	3.89	183.91
39-00031-0270-000	Gartan	DL	C 050 156	67	2.05	77.44
36-00123-3530-000	Garty	CN	N 280 980	67	0.83	20.76
23-000z3-0010-000	Gill	KY	Q 610 140	3	1.40	16.43
35-00117-0040-000	Gill	SO	G 750 340	0	13.81	365.62
21-00222-0010-000	Glanmore	KY	V 775 550	8	0.57	24.44

Table 2.1 (cont.) Physical characteristics of lakes

Lake Code	Lake name	Co.*	National Grid Ref.	Altitude meters	Lake area km^2	Catchment area km^2
36-00123-2420-000	Glasshouse	CN	H 270 060	48	0.54	117.16
38-00027-0010-000	Glen	DL	C 105 295	24	1.68	124.22
35-00117-0120-000	Glenade	LM	G 825 464	66	0.74	15.87
21-000h3-0040-000	Glenbeg	KY	V 705 530	78	0.66	7.58
35-00119-0010-000	Glencar	LM	G 750 435	28	1.15	41.22
32-00130-0050-000	Glencullin	MO	L 819 696	38	0.34	6.08
31-00136-0490-000	Glendollagh	GY	L 840 475	16	0.83	49.61
31-00138-0060-000	Glenicmurrin	GY	M 000 310	28	1.65	67.05
26-00156-0580-000	Glinn	RN	M 635 865	87	0.55	2.86
26-00157-0650-000	Glore	WH	N 489 719	79	0.24	16.75
36-00123-0330-000	Golagh	DL	G 965 662	101	0.60	4.59
36-00123-4440-000	Gowna	LD	N 284 924	61	4.15	129.40
25-0155b-0320-000	Graney	CE	R 556 930	46	3.72	109.64
22-00207-0530-000	Guitane	KY	W 025 845	77	2.46	19.03
36-00123-3060-000	Gulladoo	CN	N 240 990	51	0.39	82.39
31-000r4-2010-000	Hibbert	GY	L 882 223	15	0.25	3.05
31-00136-0520-000	Inagh	GY	L 845 520	25	3.14	48.08
27-00158-1320-000	Inchiquin	CE	R 270 895	19	1.08	135.58
21-00220-0050-000	Inchiquin	KY	V 835 628	42	0.77	17.94
19-00228-0170-000	Inishcarra	CK	W 480 718	40	4.90	783.79
31-000r4-0050-000	Invernagleragh	GY	L 925 400	4	0.56	11.19
34-00110-2390-000	Islandeady	MO	M 090 880	27	1.39	55.94
38-00030-0020-000	Keel	DL	C 153 250	97	0.61	2.60
33-i5_33-0820-000	Keel (Achill)	MO	F 650 060	6	0.85	12.36
38-00022-0080-000	Keel (Rosses)	DL	B 847 162	136	0.11	3.96
26-0155a-2720-000	Key	RN	G 840 600	42	8.90	
26-0155a-1260-000	Kilglass	RN	M 980 850	38	2.02	230.30
26-0155a-0180-000	Killinure	WH	N 070 460	35	2.56	83.12
38-000j6-0050-000	Kiltooris	DL	G 675 970	7	0.43	5.55
26-00157-0570-000	Kinale	CN	N 390 810	62	1.95	263.37
38-u6_38-0110-000	Kindrum	DL	C 185 430	8	0.61	3.67
32-00125-0020-000	Knappabeg	MO	M 008 803	30	0.22	18.45
32-00133-0050-000	Kylemore	GY	L 770 552	35	1.32	20.91
34-00110-2220-000	Lannagh	MO	M 111 885	28	0.59	62.46
07-00159-1150-000	Lene	WH	N 510 685	93	4.16	12.96
30-00143-0710-000	Lettercraffroe	GY	M 059 375	160	0.82	4.25
34-00110-0930-000	Levally	MO	G 145 045	29	1.23	20.18
28-00149-0080-000	Lickeen	CE	R 175 909	70	0.84	9.03
31-00137-0260-000	Loughanillaun	GY	L 848 415	26	0.57	20.59
30-00143-1360-000	Loughanillaun (M)	GY	L 809 471	40	0.67	9.19
36-00123-1070-000	Macnean	LM	H 032 397	50	9.78	120.37
30-00143-1580-000	Mask	MO	M 100 600	20	83.43	938.40
31-t4_31-0010-000	Maumeen	GY	L 615 412	5	0.56	2.28
30-00143-1460-000	Maumwee	GY	L 977 484	46	0.28	3.97
38-00016-0110-000	Meela	DL	B 740 133	3	0.57	9.52
26-0155a-2950-000	Meelagh	RN	G 890 120	50	1.16	7.04
35-00121-0010-000	Melvin	LM	G 900 540	25	22.06	183.05
01-00063-0050-000	Mourne	DL	H 068 896	168	0.67	8.87
27-00158-1470-000	Muckanagh	CE	R 370 925	17	0.96	22.15
06-00094-0280-000	Muckno	MN	H 845 195	86	3.57	109.08
22-00207-0270-000	Muckross	KY	V 950 852	17	2.67	137.20
38-0016-0780-000	Mullaghderg	DL	B 760 200	3	0.54	5.60
26-0155a-1400-000	Nablahy	RN	M 952 885	40	0.53	104.68
38-00023-0090-000	Nacung Upper	DL	B 894 205	59	2.08	78.62

Table 2.1 (cont.) Physical **c**haracteristics of lakes

Lake Code	Lake name	Co.*	National Grid Ref.	Altitude meters	Lake area km^2	Catchment area km^2
07-00159-1120-000	Nadreegeel	CN	N 545 930	103	0.44	11.44
30-00143-1660-000	Nafooey	GY	L 970 595	25	2.48	33.67
31-000q4-0040-000	Nagravin	GY	L 990 215	14	0.56	5.12
31-000r4-0720-000	Nahasleam	GY	L 971 440	33	0.28	22.78
22-00208-0060-000	Nakirka	KY	V 735 892	173	0.06	0.53
37-00052-0120-000	Nalughraman	DL	G 657 886	180	0.56	2.22
37-00055-0010-000	Namanfin	DL	G 797 839	128	0.23	1.85
32-00132-0040-000	Nambrackkeagh	GY	L 821 604	65	0.07	0.56
40-00004-0020-000	Naminn	DL	C 396 419	150	0.15	1.10
28-00152-0060-000	Naminna	CE	R 176 710	169	0.20	0.91
26-0155a-2460-000	Oakport	RN	G 888 036	42	0.51	
26-00156-0570-000	O'Flynn	RN	M 585 795	77	1.37	18.35
31-00136-0670-000	Oorid	GY	L 930 460	45	0.61	7.40
26-00157-0260-000	Owel	WH	N 400 580	97	10.22	32.15
09-00168-0230-000	Pollaphuca	WW	O 000 010	180	19.53	318.67
07-00159-0600-000	Ramor	CN	N 600 869	82	7.13	234.84
29-00145-0180-000	Rea	GY	M 615 155	81	3.01	234.84
26-0155a-0320-000	Ree	WH	N 000 580	35	106.10	4620.07
26-0155a-1770-000	Rinn	LM	N 100 930	39	1.65	178.06
36-00123-2800-000	Rockfield	CN	H 272 035	47	0.38	82.39
30-00143-0330-000	Ross	GY	M 190 365	6	1.39	53.81
26-0155a-2710-000	Rowan	LM	H 085 060	73	0.48	1.98
26-0155a-1780-000	Sallagh	LM	N160 912	55	0.49	3.68
38-00027-0110-000	Salt	DL	C 124 262	246	0.29	1.01
36-00123-1630-000	Scur	LM	H 027 084	62	1.14	62.87
26-00157-0690-000	Sheelin	CN	N 450 850	61	18.16	242.16
31-000r4-0950-000	Shindilla	GY	L 960 460	38	0.70	9.66
36-00123-5720-000	Sillan	CN	H 700 070	94	1.62	51.26
31-000r4-1600-000	Skannive	GY	L 809 320	16	0.81	14.62
07-00159-0620-000	Skeagh Upper	CN	H 650 010	150	0.61	4.09
29-0155a-2950-000	Skean	RN	G 858 125	48	1.14	78.31
33-i5_33-0660-000	Sruhill	MO	F 724 085	2	0.51	2.71
36-00123-1640-000	St. Johns	LM	H 090 100	60	1.46	22.56
26-00157-0040-000	Sunderlin	WH	N 220 501	77	0.22	2.57
34-00110-0630-000	Talt	SO	G 398 150	130	0.97	5.70
26-0155a-1810-000	Tap	LD	N 006 945	39	0.62	
10-00171-0090-000	Tay	WW	O 160 075	250	0.50	20.03
35-00116-0230-000	Templehouse	SO	G 615 170	54	1.19	268.54
22-00207-0260-000	Upper	KY	V 900 817	18	1.70	113.08
26-0155a-2400-000	Urlaur	MO	M 512 888	81	1.15	13.60
38-00027-0210-000	Veagh	DL	C 017 211	40	2.61	36.88
38-00016-0370-000	Waskel	DL	B 738 161	3	0.31	3.14
36-00123-4940-000	White	MN	H 680 188	75	0.54	132.38

* Co.- County: CE – Clare, CK – Cork, CN – Cavan, DL – Donegal, GY – Galway, KY – Kerry, LD - Longford, LM – Leitrim, MN – Monaghan, MO – Mayo, RN – Roscommon, SO – Sligo, TN – Tipperary, WD – Waterford, WH – Westmeath, WW – Wicklow.

Table 2.2 Names of lakes and dates sampled.

LAKE	Co.*	Spring 2001	Summer 2001	Spring 2002	Summer 2002	Summer 2003
Acrow	CE			18/04/02	04/09/02	
Ahalia	GY	29/05/01				
Aleck More	DL	02/05/01	24/07/01			
Alewnaghta	CE	27/04/01				22/08/03
Allen	LM	05/06/01				
Allua	CK	15/05/01	17/08/01			
Anaserd	GY	09/05/01				30/07/03
Annaghmore	RN	11/06/01		30/04/02	13/09/02	
Anure	DL	02/05/01	25/07/01	17/04/02	14/08/02	
Ardderry	GY	24/04/01	01/08/01	22/04/02	30/07/02	
Arrow	SO	28/06/01				
Atedaun	CE	31/03/01	03/08/01			
Atorick	CE	29/03/01	10/08/01			
Aughrusbeg	GY	09/05/01				31/07/03
Avaghon	MN	07/06/01	31/08/01			
Ballinahinch	GY	08/05/01	05/09/01			
Ballycuirke	GY	29/05/01	03/09/01			
Ballycullinan	CE	02/04/01				30/07/03
Ballyeighter	CE	25/04/01				31/07/03
Ballymore	MO	04/04/01	22/08/01			
Ballynakill	GY	10/05/01				31/07/03
Ballynakill (Gorumna)	GY			23/04/02	06/08/02	
Bane	WH	24/05/01	24/08/01	09/04/02	11/09/02	
Barfinnihy	KY			24/04/02	16/08/02	
Barra	DL	23/05/01	26/07/01	11/04/02	15/08/02	
Belhavel	LM	29/06/01				24/07/03
Beltra	MO	01/06/01	16/08/01	08/05/02	27/08/02	
Boderg	LD		04/07/01			20/08/03
Bofin	GY	24/04/01	30/07/01			
Bofin	LD		05/07/01			20/08/03
Brackley	CN	16/06/01				06/08/03
Bridget	CE	23/04/01	31/07/01			
Bunny	CE	06/04/01	04/08/01	16/04/02	03/09/02	
Caragh	KY	30/04/01		25/04/02	15/08/02	
Carra	MO	17/05/01				
Carraigadrohid	CK	15/05/01				
Carrigeencor	LM	30/06/01				24/07/03
Carrowkeribly	MO	03/04/01	22/08/01			
Carrowmore	MO	09/04/01				
Cavetown	RN	13/06/01	09/08/01			
Clonee	KY	17/05/01	16/08/01			
Clonlea	CE	24/04/01				31/07/03
Cloon	KY	30/04/01	13/08/01	25/04/02	16/08/02	
Cloonacleigha	SO	19/04/01				11/08/03
Cloonadoon	GY		31/07/01			
Cloonagh	RN	11/04/01				12/08/03
Cloonaghlin	KY			26/04/02	21/08/02	
Cloongat	GY			24/04/02	07/08/02	
Conn	MO	14/05/01				
Coosan	WH		03/07/01			21/08/03
Corglass	CN	15/06/01				05/08/03
Corrib	GY	25/05/01		05/06/02	21/08/02	
Corry	RN	14/06/01				13/08/03

Table 2.2 (Cont.) Names of lakes and dates sampled.

LAKE	Co.*	Spring 2001	Summer 2001	Spring 2002	Summer 2002	Summer 2003
Coumduala	WD			22/05/02		
Craghy	DL	01/05/01	24/07/01	16/04/02	13/08/02	
Cross	MO	09/04/01				01/09/03
Crotty	WD			22/05/02		
Cullaun	CE	02/04/01	05/08/01	17/04/02	18/07/02	
Cullaunyheeda	CE	25/04/01	01/08/01			
Cullin	MO	03/04/01	14/05/01			
Currane	KY	01/05/01		26/04/02	20/08/02	
Cutra	GY	28/03/01				29/07/03
Dan	WW	19/04/01	28/08/01	19/04/02	07/08/02	
Derg	DL	05/06/01				05/08/03
Derg	TN		10/07/01			
Derravaragh	WH	23/05/01	22/08/01			
Derrybrick	CN	16/06/01	06/09/01			
Derrycassen	CN	20/06/01	06/09/01			
Derryclare	GY	30/05/01				30/07/03
Derryhick	MO	30/03/01				23/07/03
Doo	CE	04/04/01				
Doo	DL			10/04/02	22/07/02	
Doo	MO	06/04/01		02/05/02	03/09/02	
Dromore	CN	09/06/01				07/08/03
Drumharlow	RN	14/06/01				20/08/03
Drumlaheen	LM	18/06/01	16/09/01			
Drumlona	MN	08/06/01	30/08/01			
Dúin	CE	24/04/01	30/07/01			
Dunglow	DL	01/05/01	24/07/01	16/04/02	13/08/02	
Dunlewy	DL	03/05/01				07/08/03
Durnesh	DL	06/06/01				
Easky	SO	18/04/01	15/08/01	07/05/02	28/08/02	
Egish	MN	08/06/01				10/07/03
Ennell	WH	22/05/01	25/08/01			
Errit	RN	10/04/01	14/08/01			
Eske	DL	05/06/01	29/08/01			
Fad (east)	DL			18/04/02	23/07/02	
Fad (west)	DL			18/04/02	23/07/02	
Fadda	GY		05/09/01			
Fee	GY	30/05/01	02/08/01	25/04/02	08/08/02	
Feeagh	MO	18/05/01		08/05/02	27/08/02	
Fern	DL	21/05/01	27/08/01			
Fin MO	MO			02/05/02	03/09/02	
Finn	CE	30/03/01	02/08/01			
Finn	DL	23/05/01	28/08/01			
Forbes	LD		05/07/01			19/08/03
Furnace	MO	18/05/01				
Gara	RN	29/06/01				
Garadice	LM		03/07/01			13/08/03
Gartan	DL	22/05/01	25/07/01	11/04/02	15/08/02	
Garty	CN	14/06/01	05/09/01			
Gill	KY	28/04/01				
Gill	SO	28/06/01				
Glanmore	KY	18/05/01	15/08/01			
Glasshouse	CN	09/06/01	04/09/01			
Glen	DL	22/05/01	26/07/01			
Glenade	LM	27/06/01	30/08/01			
Glenbeg	KY	18/05/01	15/08/01			
Glencar	LM	27/06/01	30/08/01	12/04/02	30/07/02	
Glencullin	MO			02/05/02	29/08/02	
Glendollagh	GY	08/05/01	06/09/01			
Glenicmurren	GY	28/05/01	31/07/01			
Glinn	RN	11/04/01				

Table 2.2 (Cont.) Names of lakes and dates sampled.

LAKE	Co.*	Spring 2001	Summer 2001	Spring 2002	Summer 2002	Summer 2003
Glore	WH	24/05/01				22/07/03
Golagh	DL	06/06/01		11/04/02	01/08/02	
Gowna	LD	30/05/01				23/07/03
Graney	CE	29/03/01	09/08/01			
Guitane	KY	16/05/01	14/08/01	23/04/02	19/07/02	
Gulladoo	CN	15/06/01	05/09/01			
Hibbert	GY			23/04/02	06/08/02	
Inagh	GY	08/05/01	04/09/01			
Inchiquin	CE	05/04/01	10/08/01			
Inchiquin	KY	17/05/01	14/08/01	27/04/02	06/09/02	
Inishcarra	CK	14/05/01				
Invernagleragh	GY	25/04/01				
Islandeady	MO	29/03/01				22/07/03
Keel	DL	21/05/01	27/08/01			
Keel (Achill)	MO	15/05/01				24/07/03
Keel (Rosses)	DL			16/04/02	14/08/02	
Key	RN	28/06/01				14/08/03
Kilglass	RN	14/06/01				20/08/03
Killinure	WH		03/07/01			03/09/03
Kiltooris	DL	30/04/01		15/04/02	12/08/02	
Kinale	CN	27/06/01				11/08/03
Kindrum	DL	21/05/01	28/08/01	10/04/02	24/07/02	
Knappabeg	MO		03/07/01			23/07/03
Kylemore	GY	10/05/01	02/08/01	24/04/02	07/08/02	
Lannagh	MO	01/06/01				22/07/03
Lene	WH	23/05/01	24/08/01	09/04/02	26/07/02	
Lettercraffoe	GY	28/05/01	03/09/01			
Levally	MO	30/03/01	23/08/01			
Lickeen	CE	31/03/01	11/08/01			
Loughanillaun	GY	25/04/01	31/07/01			
Loughanillaun (Maam)	GY	23/04/01				29/07/03
Mask	MO			01/05/02	20/08/02	
Maumeen	GY			24/04/02	07/08/02	
Maumwee	GY	23/04/01	30/07/01	22/04/02	29/07/02	
Macnean	LM		09/07/01	10/04/02	02/08/02	
Meela	DL	01/05/01				06/08/03
Meelagh	RN	13/06/01	08/08/01			
Melvin	SO	27/06/01	11/09/01	10/04/02	31/07/02	
Mourne	DL			11/04/02	03/08/02	
Muckanagh	CE	02/04/01	04/08/01	17/04/02	03/09/02	
Muckno	MN	13/06/01				11/07/03
Muckross	KY			23/04/02	22/08/02	
Mullaghderg	DL	02/05/01				06/08/03
Nablahy	RN	11/06/01	09/08/01			
Nacung Upper	DL	02/05/01				07/08/03
Nadreegel	CN	29/05/01	31/08/01			
Nafooey	GY	30/05/01	06/09/01	25/04/02	26/08/02	
Nagravin	GY	28/05/01				28/07/03
Nahasleam	GY			23/04/02	30/07/02	
Nakirka	KY			25/04/02	14/08/02	
Nalughraman	DL	30/04/01	23/07/01	15/04/02		
Namanfin	DL			15/04/02	12/08/02	
Nambrackkeagh	GY			25/04/02	26/08/02	
Naminn	DL			17/04/02	23/07/02	
Naminna	CE			17/04/02	04/09/02	
Oakport	RN	28/06/01				21/08/03
O'Flynn	RN	11/06/01	14/08/01	30/04/02	12/09/02	
Oorid	GY	24/04/01	01/08/01	22/04/02	29/07/02	
Owel	WH	23/05/01	23/08/01	08/04/02	25/07/02	

Table 2.2 (Cont.) Names of lakes and dates sampled.

LAKE	Co.*	Spring 2001	Summer 2001	Spring 2002	Summer 2002	Summer 2003
Pollaphuca	WW	11/04/01				15/07/03
Ramor	CN	31/05/01				09/07/03
Rea	GY	27/03/01		16/04/02	02/09/02	
Ree	WH		12/07/01			
Rinn	LM	28/06/01				18/07/03
Rockfield	CN	10/06/01	04/09/01			
Ross	GY	29/05/01				29/07/03
Rowan	LM	18/06/01	16/09/01			
Sallagh	LM	28/06/01				17/07/03
Salt	DL			10/04/02	23/07/02	
Scur	LM	29/06/01	15/09/01			
Sheelin	CN	27/06/01				12/08/03
Shindilla	GY	24/04/01	04/09/01	22/04/02	31/07/02	
Sillan	CN	06/06/01	29/08/01			
Skannive	GY	25/04/01				28/07/03
Skeagh	CN	29/05/01	29/08/01			
Skean	RN	13/06/01	08/08/01			
Sruhill	MO	15/05/01				
St. Johns	LM	20/06/01				14/08/03
Sunderlin	WH	25/05/01				18/08/03
Talt	SO	18/04/01	15/08/01	07/05/02	28/08/02	
Tap	LD		04/07/01			02/09/03
Tay	WW			19/04/02	07/08/02	
Templehouse	SO	19/04/01				11/08/03
Upper	KY			24/04/02	05/09/02	
Urlar	MO	10/04/01	07/08/01			12/08/03
Veagh	DL	22/05/01		17/04/02	16/07/02	
Waskel	DL	01/05/01		16/04/02	13/08/02	
White	MN	07/06/01	30/08/01			

* Co.- County: CE – Clare, CK – Cork, CN – Cavan, DL – Donegal, GY – Galway, KY – Kerry, LD - Longford, LM – Leitrim, MN – Monaghan, MO – Mayo, RN – Roscommon, SO – Sligo, TN – Tipperary, WD – Waterford, WH – Westmeath, WW – Wicklow.

2.4 Chemical characteristics of the lakes

The 201 lakes sampled represented a wide selection across the physico-chemical and trophic gradients typically found in Ireland. Table 2.3 presents some basic chemistry data for each lake from a single date. Details of the sampling methodology can be found in Chapter 3.

Table 2.3. Chemical characteristics of the lakes as measured from a single mid-lake sample collected in 2001 or 2002.

Lake	Co.†	Date sampled	pH	Conductivity µS cm^{-1}	Alkalinity mg l^{-1} $CaCO_3$	Colour PtCo/Hazen	Secchi depth meters	Chlorophyll a µg l^{-1}	Total Phosphorus µg l^{-1}	Total Nitrogen mg l^{-1}
Acrow	CE	18/04/02	5.14	73	-0.4	61	0.9	6.2	18	0.30
Ahalia (North)	GY	29/05/01	6.80	197	6.4	41	n/a	3.4	26	<1
Aleck More	DL	02/05/01	4.77	90	0.9	186	0.5	17.4	20	<1
Alewnaghta	CE	27/04/01	8.00	202	69.7	57	1.2	14.5	29	0.70
Allen	LM	05/06/01	n/a	n/a	n/a	n/a	n/a	4.0	102	n/a
Allua	CK	15/05/01	7.40	78	15.0	27	2.4	14.1	11	0.57
Anaserd	GY	09/05/01	6.74	183	11.2	16	2.5**	0.7	12	<1
Annaghmore	RN	30/04/02	8.46	351	159.4	19	n/a	0.4	6	0.48
Anure	DL	02/05/01	6.57	70	12.5	56	1.8	3.4	20	3.7
Ardderry	GY	24/04/01	6.33	84	6.1	40	1.3	2.0	20	<1
Arrow	SO	28/06/01	8.58	283	120.3	10	3.2	6.1	19	1
Atedaun	CE	31/03/01	8.28	379	169.4	26	1.6**	2.4	30	0.96
Atorick	CE	29/03/01	6.45	61	4.2	148	0.8	4.8	27	0.59
Aughrusbeg	GY	09/05/01	8.25	380	54.1	32	1.8	13.4	37	<1
Avaghon	MN	07/06/01	7.92	160	38.6	13	3.9	3.2	75	0.54
Ballinahinch	GY	08/05/01	6.27	75	5.0	32	3.3	2.6	10	7.6
Ballycuirke	GY	29/05/01	7.79	206	69.1	44	1.4	13.9	30	<1
Ballycullinan	CE	02/04/01	8.38	481	227.6	15	2.4	29.8	25	0.33
Ballyeighter	CE	25/04/01	8.42	462	211.0	27	3.4	3.2	5	0.78
Ballymore	MO	04/04/01	8.17	281	104.7	44	2.5	3.8	40	<1
Ballynakill	GY	10/05/01	7.18	145	22.4	22	3.2	2.6	17	<1
Ballynakill (Gor)	GY	23/04/02	7.1	244	20.0	20	3.2	9.4	12	<1
Bane	WH	09/04/02	8.43	297	132.5	1	7.8	1.4	5	0.46
Barfinnihy	KY	24/04/02	6.84	56	4.2	3	4.8	3.2	4	0.15
Barra	DL	23/05/01	6.31	54	3.8	45	3.2	1.1	7	<1
Belhavel	LM	29/06/01	7.25	79	12.4	156	0.3	9.3	74	0.68
Beltra	MO	01/06/01	7.39	99	19.4	n/a	2.5	2.6	14	<1
Boderg	LD	04/07/01	8.01	212	77.7	54	1.3	10.1	31	0.53
Bofin	GY	24/04/01	6.62	73	6.9	18	1.9	1.6	10	<1
Bofin	LD	05/07/01	8.15	216	80.3	48	1.7	2.8	21	0.48
Brackley	CN	16/06/01	8.03	133	46.6	55	0.9	14.9	33	0.61
Bridget	CE	23/04/01	8.21	444	200.0	55	1.3	26.2	27	0.89
Bunny	CE	16/04/02	8.47	361	156.2	9	5.4	1.4	5	0.37
Caragh	KY	30/04/01	6.73	70	3.6	23	2.8	4.0	9	0.25
Carra	MO	17/05/01	8.34	396	172.6	14	4.7	1.0	19	1.2
Carraigadrohid	CK	15/05/01	7.46	130	36.0	22	n/a	5.2	17	0.88

Table 2.3 (Cont.) Chemical characteristics of the lakes as measured from a single mid-lake sample collected in 2001 or 2002.

Lake	Co.†	Date sampled	pH	Conductivity µS cm^{-1}	Alkalinity mg l^{-1} $CaCO_3$	Colour PtCo/Hazen	Secchi depth meters	Chlorophyll a µg l^{-1}	Total Phosphorus µg l^{-1}	Total Nitrogen mg l^{-1}
Carrigeencor	LM	30/06/01	7.95	134	40.0	28	2.3	5.6	10	0.29
Carrowkeribly	MO	03/04/01	7.98	297	96.5	48	2.5	1.6	30	<1
Carrowmore	MO	09/04/01	7.17	126	15.2	86	0.5	6.4	10	n/a
Cavetown	RN	13/06/01	8.49	342	149.2	48	3.7	3.8	19	<1
Clonee middle	KY	17/05/01	6.85	63	2.7	13	2.5**	2.8	6	0.23
Clonlea	CE	24/04/01	8.49	494	230.6	42	2.5	2.4	12	0.74
Cloon	KY	30/04/01	6.57	60	2.1	14	4.6	5.2	9	0.20
Cloonacleigha	SO	19/04/01	8.34	309	133.1	46	1.7	4.5	30	<1
Cloonadoon	GY	31/07/01	n/a	n/a	n/a	16	n/a	4.6	16	<1
Cloonagh	RN	11/04/01	8.20	328	140.3	n/a	0.8	5.0	<10	<1
Cloonaghlin	KY	26/04/02	6.82	62	2.0	15	n/a	3.6	5	0.21
Cloongat	GY	24/04/02	6.36	138	2.5	29	3.1**	1.1	2	<1
Conn	MO	14/05/01	8.11	283	103.3	41	3.7	0.8	10	1.0
Coosan*	WH	03/07/01	8.18	399	180.0	25	3.15	16.1	34	n/a
Corglass	CN	15/06/01	8.30	342	153.2	24	1.1	18.5	38	0.70
Corrib	GY	25/05/01	8.54	310	92.7	14	5.3	1.5	10	1.1
Corry	RN	14/06/01	7.93	159	51.5	59	1.7	5.6	29	<1
Coumduala	WD	22/05/02	6.33	33	-0.8	13	n/a	82.6	13	0.54
Craghy	DL	01/05/01	5.78	105	3.0	73	2	2.2	n/a	<1
Cross	MO	09/04/01	8.40	536	120.5	26	n/a	46.4	70	n/a
Crotty	WD	22/05/02	6.38	40	1.9	12	n/a	0.8	6	0.30
Cullaun	CE	17/04/02	8.40	393	172.0	16	4.8	0.8	6	0.53
Cullaunyheeda	CE	25/04/01	8.44	473	215.4	55	2.4	3.6	34	1.00
Cullin	MO	03/04/01	8.34	340	128.1	44	0.4	20.4	60	<1
Currane	KY	01/05/01	6.72	84	3.8	17	3.5	5.6	8	0.18
Cutra	GY	28/03/01	7.57	124	33.6	131	1.1	4.0	31	0.69
Dan	WW	19/04/01	4.96	40	-0.4	103	0.9	2.4	13	0.40
Derg	DL	05/06/01	6.45	55	2.5	50	2.9	2.7	11	<1
Derg*	TN	10/07/01	8.47	407	176.6	50	n/a	4.8	43	n/a
Derravaragh	WH	23/05/01	8.43	467	213.2	29	2.9	5.0	12	1.32
Derrybrick	CN	16/06/01	8.34	342	153.4	14	2.1	12.1	26	0.61
Derrycassen	CN	20/06/01	8.09	196	80.5	57	0.4	14.9	41	0.54
Derryclare	GY	30/05/01	6.83	70	6.1	27	5.5	1.49	6	<1
Derryhick	MO	30/03/01	8.29	218	84.3	52	1.3	13.7	40	<1
Doo CE	CE	04/04/01	6.63	85	3.4	97	1.1	10.9	58	0.59
Doo DL	DL	10/04/02	5.88	78	2.1	85	1.8	6.4	12	<1
Doo MO	MO	02/05/02	6.69	73	2.3	8	5.6	1.7	3	<1
Dromore	CN	09/06/01	8.29	244	90.5	40	1.1	31.8	250	1.15
Drumharlow	RN	14/06/01	8.37	358	155.6	39	2.9	3.1	17	<1
Drumlaheen	LM	18/06/01	8.44	182	71.1	21	2.4	4.8	25	0.57
Drumlona	MN	08/06/01	8.18	248	98.9	39	1.2	17.7	79	0.97
Dúin	CE	24/04/01	7.98	225	81.7	54	1.0	n/a	48	1.13
Dunglow	DL	01/05/01	5.73	100	3.8	93	1.4	2.0	20	<1
Dunlewy	DL	03/05/01	6.05	62	5.8	41	2.8	0.8	20	<1
Durnesh	DL	06/06/01	8.30	1104	195.8	20	n/a	16.3	47	<1
Easky	SO	18/04/01	6.53	48	4.0	44	1.8	2.5	<10	<1
Egish	MN	08/06/01	8.21	222	73.7	27	1.2	15.3	211	0.99
Ennell	WH	22/05/01	8.29	448	188.8	18	4.2	1.8	14	1.09
Errit	RN	10/04/01	8.33	348	136.7	46	2.7	6.3	<10	<1

Table 2.3 (Cont.) Chemical characteristics of the lakes as measured from a single mid-lake sample collected in 2001 or 2002.

Lake	Co.†	Date sampled	pH	Conductivity µS cm^{-1}	Alkalinity mg l^{-1} $CaCO_3$	Colour PtCo/Hazen	Secchi depth meters	Chlorophyll a µg l^{-1}	Total Phosphorus µg l^{-1}	Total Nitrogen mg l^{-1}
Eske	DL	05/06/01	7.46	71	11.4	27	3.7	2.3	6	<1
Fad (east)	DL	18/04/02	6.35	81	5.0	53	2.5	3.3	6	<1
Fad (west)	DL	18/04/02	6.4	100	6.4	49	2.8	4.1	3	<1
Fadda	FY	05/09/01	n/a	n/a	n/a	30	4.6	4.4	6	n/a
Fee	GY	30/05/01	6.55	62	3.1	25	4.5	2.7	8	<1
Feeagh	MO	18/05/01	7.39	86	9.6	80	1.6	2.3	9	<1
Fern	DL	21/05/01	7.57	140	32.2	46	1.6	11.3	29	1.2
Fin	MO	02/05/02	6.74	79	0.5	10	5.8	2.3	3	<1
Finn	CE	30/03/01	8.29	376	162.0	27	3.6	4.0	14	0.55
Finn	DL	23/05/01	7.36	91	14.4	39	2.8	1.9	5	<1
Forbes	LD	05/07/01	8.05	226	83.9	42	n/a	5.6	25	0.66
Furnace	MO	18/05/01	7.95	15930	44.8	52	1.3	8.6	<10	<1
Gara	RN	29/06/01	8.43	378	156.0	36	0.9	6.5	43	1
Garadice	LM	03/07/01	7.91	185	74.7	43	1.7	3.6	30	0.51
Gartan	DL	22/05/01	7.06	76	12.0	59	1.5	2.6	24	<1
Garty	CN	14/06/01	8.30	139	40.8	33	1.7	14.1	36	0.77
Gill	KY	28/04/01	8.79	4020	108.9	23	n/a	10.9	44	0.37
Gill	SO	28/06/01	8.09	220	78.08	44	2.2	4.6	20	1
Glanmore	KY	18/05/01	6.63	58	1.9	20	2.3	13.3	9	0.23
Glasshouse	CN	09/06/01	8.04	159	51.7	45	1	13.3	37	0.97
Glen	DL	22/05/01	6.77	75	6.5	60	1.5	2.7	9	<1
Glenade	LM	27/06/01	8.16	193	74.3	28	3.1	12.8	14	1
Glenbeg	KY	18/05/01	6.98	82	4.6	13	2.8	16.9	7	0.24
Glencar	LM	12/04/02	8.44	248	94.3	14	3.6	4.4	5	0.26
Glencullin	MO	02/05/02	6.37	79	2.5	17	2.9	4.4	4	<1
Glendollagh	GY	08/05/01	6.29	75	6.3	38	2.4	1.3	9	<1
Glenicmurren	GY	28/05/01	6.01	72	2.0	51	1.8	1.6	6	<1
Glinn	RN	11/04/01	8.28	302	127.9	14	1.6	6.7	10	<1
Glore	WH	24/05/01	8.28	495	235.0	6	2.7**	7.7	24	1.39
Golagh	DL	11/04/02	7.20	85	6.1	96	1.2	3.6	19	0.43
Gowna North	LD	30/05/01	7.81	129	34.6	27	0.9	10.9	58	0.83
Gowna South	LD	30/05/01	8.17	251	95.7	31	1.8	3.6	35	1.10
Graney	CE	29/03/01	7.49	111	28.6	115	0.9	8.1	23	0.67
Guitane	KY	23/04/02	6.96	56	4.4	13	4.5	2.0	5	0.25
Gulladoo	CN	15/06/01	7.80	145	44.6	59	1	29.4	58	0.99
Hibbert	GY	23/04/02	6.69	201	5.1	25	1.4**	7.5	9	<1
Inagh	GY	08/05/01	6.35	67	6.3	21	3.7	1.7	11	<1
Inchiquin	CE	05/04/01	8.19	373	171.6	23	2.4	4.4	32	1.08
Inchiquin	KY	27/04/02	6.51	60	2.2	12	4.2	2.8	7	0.20
Inishcarra	CK	14/05/01	7.89	140	35.6	23	1.9	14.5	19	1.70
Invernagleragh	GY	25/04/01	5.94	71	3.8	17	3.4	2.1	<10	<1
Islandeady	MO	29/03/01	8.15	297	118.9	59	1.2	14.1	n/a	<1
Keel	DL	21/05/01	7.63	117	16.8	34	2.8	0.4	18	<1
Keel (Achill)	MO	15/05/01	7.56	183	26.2	66	1.6	6.1	24	<1
Keel (Rosses)	DL	16/04/02	5.3	135	2.4	47	1.8	5.8	8	<1
Key	RN	28/06/01	8.40	364	168.2	34	3.2	10.9	30	1.0
Kilglass	RN	14/06/01	8.31	516	250.5	26	2.8	5.8	17	<1
Killinure*	WH	03/09/03	8.25	465	220.0	22	4.1	2.0	13	0.66
Kiltooris	DL	30/04/01	7.18	205	27.4	48	3.3	1.1	20	<1
Kinale	CN	27/06/01	8.28	330	138.9	22	1.4	10.9	23	0.54

Table 2.3 (Cont.) Chemical characteristics of the lakes as measured from a single mid-lake sample collected in 2001 or 2002.

Lake	Co.†	Date sampled	pH	Conductivity µS cm^{-1}	Alkalinity mg l^{-1} $CaCO_3$	Colour PtCo/Hazen	Secchi depth meters	Chlorophyll a µg l^{-1}	Total Phosphorus µg l^{-1}	Total Nitrogen mg l^{-1}
Kindrum	DL	21/05/01	8.27	318	69.5	22	3.9	5.6	13	1.0
Knappabeg	MO	03/07/01	7.45	145	34.8	46	1.3	23.1	37	<1
Kylemore	GY	10/05/01	6.59	72	7.0	23	4.1	1.1	6	<1
Lannagh	MO	01/06/01	8.22	332	142.7	n/a	2.2	5.2	19	<1
Lene	WH	09/04/02	8.46	250	104.9	4	4.8	3.4	6	0.34
Lettercraffroe	GY	28/05/01	5.69	67	2.2	46	1.6	5.3	17	<1
Levally	MO	30/03/01	7.88	136	36.0	38	1.5	5.3	40	<1
Lickeen	CE	31/03/01	7.50	141	24.6	81	1.8	5.2	25	0.56
Loughanillaun	GY	25/04/01	5.68	75	1.9	n/a	1.8	1.1	<10	<1
Loughanillaun (M)	GY	23/04/01	6.23	73	3.6	20	1.8**	1.9	<10	<1
Macnean	LM	10/04/02	7.60	116	23.6	80	1.1	6.9	17	0.45
Mask*	MO	01/05/02	8.19	273	107.0	22	4.3	6.5	7	<1
Maumeen	GY	24/04/02	6.81	215	12.0	27	2.5	14.5	15	<1
Maumwee	GY	23/04/01	6.07	72	6.3	26	3.7	1.7	10	<1
Meela	DL	01/05/01	6.53	886	11.2	127	0.8	3.7	20	<1
Meelagh	RN	13/06/01	8.18	284	64.5	6	5.8	2.7	12	<1
Melvin	LM	10/04/02	8.15	170	54.1	73	1.5	4.4	15	0.52
Mourne	DL	11/04/02	6.77	82	0.7	25	2.2	2.2	6	0.14
Muckanagh	CE	17/04/02	8.53	462	208.6	26	4	0.8	5	0.71
Muckno	MN	13/06/01	9.08	202	61.9	36	0.6	56.8	71	1.62
Muckross	KY	23/04/02	6.82	62	5.3	15	7.5	0.8	3	0.19
Mullaghderg	DL	02/05/01	7.34	192	32.6	74	1.3	2.5	20	<1
Nablahy	RN	11/06/01	8.41	521	254.9	27	3.8	7.4	15	<1
Nacung Upper	DL	02/05/01	5.97	64	4.6	55	2.3	1.6	20	<1
Nadreegeel	CN	29/05/01	7.90	160	47.6	26	2.3	4.2	25	0.84
Nafooey	GY	30/05/01	6.79	53	5.5	18	5.6	1.9	5	<1
Nagravin	GY	28/05/01	7.26	145	15.2	26	1.5**	3.0	26	<1
Nahasleam	GY	23/04/02	6.5	101	9.6	37	1.4**	3.2	7	<1
Nakirka	KY	25/04/02	6.12	70	0.4	43	1.4	3.2	4	0.19
Nalughraman	DL	30/04/01	6.26	85	6.0	28	3.4	0.6	10	<1
Namanfin	DL	15/04/02	6.55	100	8.6	37	n/a	3.9	7	<1
Nambrackkeagh	GY	25/04/02	5.98	101	2.3	43	2.7	2.5	10	<1
Naminn	DL	17/04/02	6.55	112	7.0	40	3	1.4	9	<1
Naminna	CE	17/04/02	6.02	77	0.7	50	1.6	3.8	8	0.31
Oakport	RN	28/06/01	8.41	357	154.4	34	2.9	8.8	22	1
O'Flynn	RN	30/04/02	8.51	333	138.9	63	2.1	0.8	10	0.88
Oorid	GY	24/04/01	6.40	65	8.1	16	3	2.3	20	<1
Owel	WH	08/04/02	8.49	273	109.5	1	5.1	2.4	9	0.49
Pollaphuca	WW	11/04/01	7.78	106	32.4	80	1.1	11.3	22	0.91
Ramor	CN	31/05/01	7.86	184	56.1	41	1.2	6.4	61	1.12
Rea	GY	16/04/02	8.54	308	128.5	3	4.5**	2.4	6	0.50
Ree *	WH	12/07/01	8.43	344	160.0	60	n/a	7.7	36	n/a
Rinn	LM	28/06/01	8.19	312	133.5	75	0.5	18.5	81	0.96
Rockfield	CN	10/06/01	7.86	148	47.0	55	1.2	17.7	86	1.02
Ross	GY	29/05/01	8.19	309	128.5	20	2.3	5.8	12	<1
Rowan	LM	18/06/01	7.98	153	55.9	13	2.5	4.8	10	0.31
Sallagh	LM	28/06/01	7.96	151	47.9	50	0.8	8.5	42	0.86
Salt	DL	10/04/02	7.18	133	26.4	23	4.8	2.0	1	<1
Scur	LM	29/06/01	7.74	153	60.3	91	0.3	18.1	59	0.67

Table 2.3 (Cont.). Chemical characteristics of the lakes as measured from a single mid-lake sample collected in 2001 or 2002.

Lake	Co.†	Date sampled	pH	Conductivity µS cm^{-1}	Alkalinity mg l^{-1} $CaCO_3$	Colour PtCo/Hazen	Secchi depth meters	Chlorophyll a µg l^{-1}	Total Phosphorus µg l^{-1}	Total Nitrogen mg l^{-1}
Sheelin	CN	27/06/01	8.57	383	178.8	15	n/a	9.7	16	0.79
Shindilla	GY	24/04/01	6.45	73	6.2	27	2.7	2.4	10	<1
Sillan	CN	06/06/01	8.07	168	48.6	24	1.7	15.3	91	0.96
Skannive	GY	25/04/01	6.29	116	5.5	14	3.7	1.2	<10	<1
Skeagh	CN	29/05/01	7.66	121	31.4	40	0.9	9.7	43	1.01
Skean	RN	13/06/01	8.37	297	129.9	25	2.5	4.9	23	<1
Sruhill	MO	15/05/01	8.02	50400	172.2	23	0.8	10.8	55	1.1
St. Johns	LM	20/06/01	7.66	114	35.4	114	0.5	10.1	35	0.42
Sunderlin	WH	25/05/01	8.12	473	191.8	25	2.6	4.4	28	0.81
Talt	SO	18/04/01	8.01	190	85.1	20	4.1	0.8	20	<1
Tap	LD	04/07/01	7.96	212	76.9	61	1.2	14.5	33	0.54
Tay	WW	19/04/02	5.12	40	-0.3	134	1.7	0.6	8	0.39
Templehouse	SO	19/04/01	8.66	469	213.8	75	0.9	3.9	80	<1
Upper	KY	24/04/02	6.41	58	2.8	22	4.6	1.8	5	0.19
Urlaur	MO	10/04/01	8.41	373	148.0	33	1.5	7.7	<10	<1
Veagh	DL	22/05/01	6.30	33	2.2	42	2.6	1.4	10	<1
Waskel	DL	01/05/01	5.70	149	3.9	100	1.6	1.8	20	<1
White	MN	07/06/01	8.20	241	84.9	48	1.8	25.4	105	1.50

* some values taken from Bowman (2000)
** clear to lake bed.
†Co. - County: CE – Clare, CK – Cork, CN – Cavan, DL – Donegal, GY – Galway, KY – Kerry, LD - Longford, LM – Leitrim, MN – Monaghan, MO – Mayo, RN – Roscommon, SO – Sligo, TN – Tipperary, WD – Waterford, WH – Westmeath, WW – Wicklow.

2.5 CORINE land use data

CORINE landuse data for each of the lake catchments was generated using GIS mapping and software. The CORINE land cover maps were developed as part of an EC funded project by visual analysis of LANDSAT Thematic Mapper satellite images collected over Ireland in 1989 and 1990. The 44 classes of CORINE land cover, standardised for all of Europe, have been aggregated into six major landuse categories (Appendix 1) and provide an insight into land use within each of the lake catchments. Table 2.4 shows the CORINE landuse data, aggregated into these five categories, expressed as percentage of catchment area for each lake sampled.

Table 2.4 CORINE (1990) landuse data expressed as a percentage of catchment area.

Lake	Co.[†]	Urban	Forestry	Pasture	Other Agriculture	Peat	Other
Acrow	CE	0	0	0	0	100	0
Ahalia North	GY	0	3	0	1	93	3
Aleck More	DL	0	0	0	8	92	0
Alewnaghta	CE	0	27	31	2	34	6
Allen	LM	0	9	15	54	21	0
Allua	CK	0	17	36	3	44	0
Anaserd	GY	0	0	0	7	0	93
Annaghmore	RN	0	0	91	0	0	9
Anure	DL	0	0	7	12	81	0
Ardderry	GY	0	10	0	0	90	0
Arrow	SO	0	4	59	29	8	0
Atedaun	CE	0	1	40	33	4	22
Atorick	CE	0	36	4	0	58	2
Aughrusbeg	GY	0	0	0	100	0	0
Avaghon	MN	0	0	93	7	0	0
Ballinahinch	GY	0	13	0	5	81	0
Ballycuirke	GY	0	4	16	9	68	2
Ballycullinan	CE	0	0	55	27	5	13
Ballyeighter	CE	0	0	51	6	14	29
Ballymore	MO	0	4	47	2	48	0
Ballynakill	GY	0	0	27	15	60	0
Ballynakill (Gorumna)	GY						
Bane	WH	0	0	95	5	0	0
Barfinnihy	KY	0	0	0	5	95	0
Barra	DL	0	0	2	24	62	12
Belhavel	LM	0	18	10	57	16	0
Beltra	MO	0	15	0	19	63	2
Boderg	LD						
Bofin	GY	0	9	0	3	88	0
Bofin	LD						
Brackley	CN	0	11	27	54	8	0
Bridget	CE	0	76	0	24	0	0
Bunny	CE	0	0	37	6	1	56
Caragh	KY	0	5	5	14	67	9
Carra	MO	0	4	51	21	17	6
Carraigadrohid	CK	0	9	60	12	18	0
Carrigeencor	LM	0	1	57	42	0	0
Carrowkeribly	MO	0	6	47	2	45	0
Carrowmore	MO	0	2	16	5	77	0
Cavetown	RN	0	1	60	32	1	5
Clonee	KY	0	7	4	12	74	2
Clonlea	CE	0	0	96	0	0	4
Cloon	KY	0	0	0	17	54	28
Cloonacleigha	SO	0	0	73	6	13	8
Cloonadoon	GY	0	3	0	1	89	6
Cloonagh	RN	0	0	46	2	45	6
Cloonaghlin	KY	0	0	0	29	70	0
Cloongat	GY	0	0	0	0	100	0
Conn	MO	0	3	33	15	47	2
Coosan	WH	0	0	63	33	0	4
Corglass	CN	0	0	100	0	0	0
Corrib	GY	0	3	47	48	26	6
Corry	RN						
Coumduala	WD	0	0	0	0	100	0
Craghy	DL	0	0	2	4	95	0

Table 2.4 (Cont.) CORINE data expressed as a percentage of catchment area.

Lake	Co.[†]	Urban	Forestry	Pasture	Other Agriculture	Peat	Other
Cross Lake	MO	0	0	65	0	0	35
Crottys	WD	0	0	0	0	100	0
Cullaun	CE	0	0	72	5	0	22
Cullaunyheeda	CE	0	6	75	11	6	3
Cullin	MO	0	3	25	23	39	9
Currane	KY	0	4	12	15	64	5
Cutra	GY	0	28	20	5	43	4
Dan	WW	0	9	1	38	52	0
Derg	DL	0	36	0	3	58	3
Derg	TN						
Derravaragh	WH	0	4	81	5	7	2
Derrybrick	CN	0	0	100	0	0	0
Derrycassan	CN	0	7	43	40	10	0
Derryclare	GY	0	14	0	6	79	0
Derryhick	MO	0	0	15	51	35	0
Doo	CE	0	5	25	6	64	0
Doo	MO	0	6	0	61	51	2
Doo	DL	0	0	0	0	100	0
Dromore	CN	0	1	93	1	1	3
Drumharlow	RN						
Drumlaheen	LM	0	13	87	0	0	0
Drumlona	MN	2	1	94	0	0	3
Dúin	CE	0	10	67	2	19	2
Dunglow	DL	0	0	1	5	94	0
Dunlewy	DL	0	2	2	12	75	9
Durnesh	DL	0	0	76	13	0	13
Easky	SO	0	0	0	0	100	0
Egish	MN	0	0	89	11	0	0
Ennell	WH	4	3	80	10	0	2
Errit	RN	0	3	58	0	4	0
Eske	DL	0	4	13	31	47	6
Fad (east)	DL	0	0	0	0	100	0
Fad (west)	DL	0	0	0	0	100	0
Fadda	GY	0	0	0	0	100	0
Fee	GY	0	14	0	14	72	0
Feeagh	MO	0	23	0	10	64	3
Fern	DL	0	6	27	23	40	4
Fin	MO	0	5	0	36	56	2
Finn	CE	0	0	50	50	0	0
Finn	DL	0	0	5	0	95	0
Forbes	LD						
Furnace	MO	0	19	0	13	64	4
Gara	RN	0	1	69	8	22	1
Garadice	LM	0	6	48	38	7	0
Gartan	DL	0	5	5	31	53	6
Garty	CN	0	0	100	0	0	0
Gill	KY	0	10	39	4	39	5
Gill	SO	0	10	27	40	20	3
Glanmore	KY	0	0	4	27	67	0
Glasshouse	CN	0	1	97	1	1	0
Glen	DL	0	6	3	13	74	4
Glenade	LM	0	2	6	29	63	0
Glenbeg	KY	0	0	0	33	67	0
Glencar	LM	0	7	9	43	40	1
Glencullin	MO	0	0	0	0	100	0

Table 2.4 (Cont.) CORINE data expressed as a percentage of catchment area.

Lake	Co.[†]	Urban	Forestry	Pasture	Other Agriculture	Peat	Other
Glendollagh	GY	0	8	1	8	82	0
Glenicmurrin L	GY	0	2	1	10	82	5
Glinn	RN	0	10	83	0	7	0
Glore	WH	0	2	83	3	0	12
Golagh	DL	0	18	0	28	54	0
Gowna	LD	0	2	94	0	4	0
Graney	CE	0	25	32	5	36	2
Guitane	KY	0	0	10	7	83	0
Gulladoo	CN	0	0	99	0	1	0
Hibbert	GY						
Inagh	GY	0	15	0	5	80	0
Inchiquin	CE	0	1	34	38	3	24
Inchiquin	KY	0	7	5	10	77	1
Inishcarra	CK	0	9	63	13	15	0
Invernagleragh	GY	0	0	0	0	100	0
Islandeady	MO	0	6	1	31	35	28
Keel	DL	0	0	25	0	65	0
Keel (Achill)	MO	0	0	10	11	73	6
Keel (Rosses)	DL	0	0	0	0	100	0
Key	RN						
Kilglass	RN	0	0	87	6	6	4
Killinure	WH	0	3	81	6	7	3
Kiltooris	DL	0	0	48	26	17	9
Kinale	CN	0	3	90	4	2	0
Kindrum	DL	0	0	18	22	60	0
Knappabeg	MO	0	0	3	13	52	32
Kylemore	GY	0	12	0	6	67	15
Lannagh	MO	0	6	1	36	32	25
Lene	WH	0	0	79	12	0	9
Lettercraffroe	GY	0	41	0	0	59	0
Levally	MO	0	2	8	25	65	0
Lickeen	CE	0	0	50	19	21	0
Loughanillaun	GY	0	0	0	2	98	0
Loughanillaun (Maam)	GY	0	0	0	0	97	3
Macnean	LM	0	13	26	38	23	0
Mask	MO	0	2	33	22	32	10
Maumeen	GY	0	0	15	0	64	22
Maumwee	GY	0	0	0	0	100	0
Meela	DL	0	0	3	31	66	0
Meelagh	RN	0	23	28	26	23	0
Melvin	LM	0	7	28	34	27	4
Mourne	DL	0	12	3	25	57	3
Muckanagh	CE	0	0	55	10	22	13
Muckno	MN	2	2	90	5	1	0
Muckross	KY	0	9	1	8	81	1
Mullaghderg	DL	0	0	6	32	54	8
Nablahy	RN	0	1	93	4	0	2
Nacung	DL	0	5	2	10	79	4
Nadreegeel	CN	0	0	96	0	0	4
Nafooey	GY	0	0	44	1	55	0
Nagravin	GY	0	0	21	59	20	0
Nahasleam	GY	0	6	0	0	93	1
Nakirka	KY	0	0	0	0	100	0
Nalughraman	DL	0	0	0	28	72	0
Namanfin	DL	0	0	0	0	100	0

Table 2.4 (Cont.) CORINE data expressed as a percentage of catchment area.

Lake	Co.†	Urban	Forestry	Pasture	Other Agriculture	Peat	Other
Nambrackkeagh	GY	0	44	0	2	53	0
Naminn	DL	0	0	0	0	100	0
Naminna	CE	0	37	0	0	63	0
Oakport	RN						
O'Flynn	RN	1	0	54	3	42	0
Oorid	GY	0	3	0	5	92	0
Owel	RN	0	4	80	14	0	1
Pollaphuca	WW	1	15	21	23	40	0
Ramor	CN	0	2	94	2	0	2
Rea	GY	3	0	86	10	0	0
Ree	WH	0	3	75	10	11	1
Rinn	LM	0	2	91	3	3	0
Rockfield	CN	0	0	99	0	1	0
Ross	GY	0	7	12	39	38	5
Rowan	LM	0	0	100	0	0	0
Sallagh	LM	0	0	89	7	4	0
Salt	DL	0	0	0	0	100	0
Scur	LM	0	5	42	40	13	0
Sheelin	CN	1	3	91	3	2	0
Shindilla	GY	0	6	0	0	94	0
Sillan	CN	0	0	100	0	0	0
Skannive	GY	0	0	0	1	99	0
Skeagh Upper	CN	0	0	93	0	0	7
Skean	RN	0	6	42	42	9	0
Sruhill	MO	0	0	25	0	75	0
St. Johns	LM	0	2	55	43	0	0
Sunderlin	WH	0	0	82	18	0	0
Talt	SO	0	1	26	0	73	0
Tap	LD						
Tay	WW	0	1	0	40	59	0
Templehouse	SO	0	1	78	6	13	2
Upper	KY	0	6	0	8	84	2
Urlaur	RN	0	0	41	9	30	20
Veagh	DL	0	3	0	27	65	4
Waskel	DL	0	0	0	39	61	0
White	MN	0	0	97	1	1	0

†Co. - County: CE – Clare, CK – Cork, CN – Cavan, DL – Donegal, GY – Galway, KY – Kerry, LD - Longford, LM – Leitrim, MN – Monaghan, MO – Mayo, RN – Roscommon, SO – Sligo, TN – Tipperary, WD – Waterford, WH – Westmeath, WW – Wicklow.

3. General Methodology

3.1 Introduction

This chapter describes the methodologies used to collect the chemical, biological and environmental data as well as the statistical methods used for data analysis. Chemical analysis of lake water was performed in order to assist in the development of a typology and classification scheme compliant with the requirements of the Water Framework Directive. The three biological elements focused on were phytoplankton, macrophytes and benthic invertebrates (both littoral and profundal) all of which are listed in Annex V of the directive and must be used in the ecological assessment of lakes. Of the other Annex V elements, fish were not monitored and phytobenthos was only assessed by abundance (chlorophyll *a*) rather than composition. Standard operating procedures containing the methodology followed in detail were submitted to the EPA.

3.2 Water chemistry

All lakes were surveyed by boat. Hand-held echo-sounders were used to locate a relatively deep spot close to the lake centre, the location of which was recorded using a Global Positioning System (GPS). Secchi depth was measured using a plain white Secchi disc 30 cm in diameter. A Ruttner sampling bottle was used to collect a 5 metre composite water sample. This entailed the collection of 1 litre of water at 6 discrete depths (subsurface, 1, 2, 3, 4 and 5 m), which were mixed to give a composite sample. Where station depth was less than 5 m, a subsurface sample was obtained. Subsamples of the integrated water sample were obtained for the determination of total phosphorus and total nitrogen. Additional subsamples were filtered on the lakeshore for the determination of dissolved nutrients and colour. Depending on sample turbidity, between 0.1 and 2 litres of water, were filtered using a Whatman GF/C 47 mm diameter filter for chlorophyll *a* analysis. Filters were immediately placed in 14 ml of methanol and kept in the dark. Chlorophyll *a* was determined using a spectrophotometer following hot methanol extraction. Water samples were processed in the chemistry laboratories at the EPA regional Inspectorates in Castlebar and Dublin for total phosphorus (TP $\mu g\ l^{-1}$), total nitrogen (TN $mg\ l^{-1}$) and colour ($mg\ l^{-1}$ PtCo or Hazen).

3.2.1 Conductivity, pH and alkalinity measurements

Conductivity (µS cm^{-1}) and pH were measured on subsamples in the field using a WTW Multiline P3 pH/LF meter with a conductivity/temperature probe and pH electrode. Alkalinity greater than 10 mg l^{-1} $CaCO_3$ was determined on a 50 ml sample by titration to pH 4.5 using 0.01 molarity H_2SO_4. Alkalinity less than 10 mg l^{-1} $CaCO_3$ was determined by Gran titration using a 100 ml sample titrated to four end points between pH 4.4 and 3.7 (Mackereth *et al*., 1978).

3.2.2 Temperature and oxygen profiles

Temperature (^oC) and oxygen (mg l^{-1}) profiles were collected from each lake at 1 m intervals using a WTW OXI 197 meter with a 25 m cable.

3.3 Biological elements

3.3.1 Phytoplankton

Two phytoplankton samples, a quantitative and a qualitative sample, were collected from each site on each date sampled (see Table 2.1 for dates of sampling). The quantitative sample was a 100 ml subsample obtained from the composite water sample (section 3.2). The qualitative sample was a vertical haul from 6 m, or 1 m above the bottom (if depth was less than 6 m), using a 53 µm mesh net (30 cm aperture diameter, 84 cm length). Where phytoplankton densities appeared low, additional hauls were collected. All samples were preserved in Lugol's iodine and stored in darkness until identification. Phytoplankton samples were counted in settling chambers using an inverted microscope following the Utermöl technique (Utermöl, 1958). Taxa were identified using the available taxonomic keys (Appendix 2).

3.3.2 Macrophytes

Macrophyte sampling was undertaken during the Summer sampling season. Dates of sampling are listed in Table 2.2. At each lake, between four and six transects relatively evenly spaced around the lakes were surveyed. A transect was 100 m long, perpendicular to the shore and consisted of a series of sampling sites at intervals of 0, 2.5, 5, 7.5, 10, 25, 50, 75 and 100 m. In addition, a 20 m shoreline investigation was conducted. At each site along the transect, the boat was anchored and the depth and GPS position recorded. Samples were obtained using four throws of a double-headed rake. The macrophytes collected from the first throw of the rake were weighed

collectively on a digital balance, identified and the percentage species composition was recorded. If no macrophytes were collected on the first rake thrown at a sampling site then the weight was recorded as zero. A further three samples were collected at this site and the abundance was recorded using a 5 point scale (5=dominant to1=rare) on the basis of the occurrence of the collected species on all four rakes. If the shoreline was fringed by reeds, the species abundance and their distance inshore was also recorded. Macrophytes were primarily identified in the field. Difficult specimens were returned to the laboratory for closer examination. All specimens were identified using the available taxonomic keys (Appendix 3).

Phytobenthos - epilithic algae

Epilithic algae samples were collected from the littoral zone. Two stones covered by at least 20 cm of water and approximately 12 x 7 cm in size with a flat surface facing uppermost were collected. A plastic mask with a 2 cm^2 opening was held over the flat surface of each stone. Algae were brushed from the opening in the plastic using a small toothbrush until the exposed area was clean. Subsequently, the plastic and toothbrush and the exposed area of the stone were rinsed with filtered lake-water and collected into a 100ml glass bottle. The sample was preserved in Lugol's iodine and stored in darkness.

To estimate epilithic chlorophyll *a*, three small stones from the lake littoral zone were collected and placed into a 15 ml centrifuge tube filled with 10 ml methanol and kept in the dark. Following hot methanol extraction, chlorophyll *a* was measured using a spectrophotometer and expressed as μg cm^2, using an equation modified from the UK Standing Committee of Analysts (1980).

3.3.3 Benthic invertebrates

Profundal invertebrates

Profundal invertebrates were predominantly collected during the Spring sampling period in either 2001 or 2002 (See Table 2.1 for dates of sampling). At each site, five replicate samples were collected using a 225 cm^2 Ekman grab. Samples were transferred to a 500 μm mesh net and filtered *in situ* to remove excess sediment. The remaining sample was preserved in 70% alcohol and subsequently sorted and

identified in the laboratory to lowest taxonomic resolution practicable (typically genus) using available taxonomic keys (Appendix 4).

Littoral macroinvertebrates

Lakes were sampled for littoral macroinvertebrates in Spring 2001 and both Spring and Summer 2002 (see Table 2.1 for dates of sampling). Two minute kick/sweep samples were collected, preferentially from exposed stony shorelines, using a rectangular framed hand net (260mm wide, 200mm high, 670 μm mesh). Multi-habitat samples were avoided and if more than one habitat was available for sampling (e.g. macrophytes) then separate samples were collected from each habitat and treated separately. All samples were preserved on site in 70% alcohol and subsequently sorted and identified to the lowest taxonomic resolution practicable (typically species) using the available taxonomic keys (Appendix 5).

3.4 Statistical analysis

A number of univariate and multivariate statistical techniques were used to assist in the development of a biologically validated typology on an element by element basis.

- Data were classified using Two-Way-Indicator-Species-Analysis (TWINSPAN) and Cluster Analysis to identify lake groups on the basis of their biological communities.
- Indirect ordination methods (Multidimensional Scaling (MDS), non-metric multidimensional scaling (NMS) and detrended correspondence analysis (DCA)) were used to map the community data in two or three dimensional space and provide visual support to the groups obtained by the classification.
- Multiple random permutation procedures (MRPP) or Analysis of Similarities (ANOSIM) were used to test for differences between lake groups based upon their biological communities.
- Indicator Species Analysis identified statistically significant indicator species for each group.
- Direct ordination methods (Canonical Correspondence Analysis (CCA) and Canonical Variates Analysis (CVA)) identified and described underlying environmental gradients.

TWINSPAN, MRPP, NMS and Indicator species analysis were performed in PC-ORD v4.25 (McCune and Mefford, 1999). Cluster analysis, ANOSIM and MDS ordinations were performed using PRIMER v5.2.9 (Clarke and Warwick, 1994). Additional Cluster Analysis was done using STATISTICA™ 5.1 (Statsoft Inc, 1998). CCA and CVA ordinations were performed in CANOCO v4.52 (ter Braak and Smilauer, 2002).

4. Phytoplankton

4.1 Introduction

The Water Framework Directive (WFD) requires that the composition, abundance and biomass of phytoplankton are used to determine the ecological status of lakes (CEC, 2000). Phytoplankton are microscopic algae suspended in a water column. They are of fundamental importance to the functioning of lake ecosystems, forming the basis of the aquatic food chain (Reynolds, 1984). The phytoplankton also represent a major component of aquatic diversity: at least 5000 species have been recorded from the UK and Ireland (John *et al*., 2002).

4.1.1 The influence of nutrient enrichment on phytoplankton

The well-documented relationship between total phosphorus (TP) and chlorophyll *a* (Sakamoto, 1966; Dillon and Rigler, 1974) illustrates the close relationship between nutrient enrichment and overall phytoplankton abundance and biomass. Increased phytoplankton growth severely reduces water clarity and is one of the most visible signs of deteriorating ecological quality. Accompanying the notable increase in abundance are changes in phytoplankton composition. Increased concentrations of nutrients, particularly phosphorus, can lead to an increase in abundance of potentially toxic groups such as blue-green algae (Cyanophyta) (Petersen *et al*., 1999). Other phytoplankton whose abundance can indicate eutrophic conditions include the diatoms (Bacillariophyta): *Asterionella* spp., *Fragilaria crotonensis*, *Aulacoseira granulata* and the green algae (Chlorophyta): *Pediastrum* spp. and *Scenedesmus* spp. (Hutchinson, 1967).

4.1.2 Development of a typology for phytoplankton

One of the problems in trying to measure how phytoplankton populations change as a result of eutrophication is that there are different types of lakes, which may respond to nutrient input differently. Therefore, natural variation in relatively undisturbed or ‘reference’ lakes must first be taken into account before the effects of eutrophication can be ascertained. The WFD recognises this and requires member states to separate lakes into types based on their natural characteristics to allow ecological change to be detected more easily (CEC, 2000; REFCOND, 2003).

The WFD (Annex II) describes two systems, A and B, either of which may be used to derive a typology. Ireland has adopted a system B approach owing to the greater flexibility in both the parameters that can be used and in the setting of their boundaries. The parameters proposed by the WFD and their potential influence on phytoplankton are listed in Table 4.1. A complicating factor is that phytoplankton are also likely to be influenced by other factors that are not considered in a typology, such as grazing by zooplankton. One of the objectives of this chapter is to define a typology for Irish lakes using phytoplankton. The approach taken is to examine phytoplankton in 62 reference lakes and to define the boundaries of distinct biological types in terms of the most important typological parameters.

Table 4.1 System A and B factors that can be used to form a lake typology (obligatory factors are in bold). Examples of other factors and their potential influence on phytoplankton are also listed.

WFD typology factors	**Potential influence on phytoplankton**	**System**
Ecoregion	Varying biogeography of species	**A**
Latitude	"	**B**
Longitude	"	**B**
Mean air temperature	Ice cover? light, influence on metabolic rates	B
Air temperature range	"	B
Altitude	"	**A B**
Mean depth	Light, mixing depth of water column, internal loading	**A** B
Depth	"	**B**
Lake area	Exposure, stratification and mixing	**A B**
Geology	CO_2 system - community composition	**A B**
Acid neutralising capacity	"	B
Residence time	Hydraulic washout, nutrient dynamics	B
Mixing characteristics/Stratification	Light, mixing depth of water column	B
Background nutrient status	Abundance and composition	B
Lake shape	Minor influence	B
Mean substratum composition	"	B
Water level fluctuation	"	B
e.g. of other factors		
Alkalinity	CO_2 system - community composition	
Reference light environment - colour, turbidity	Quantity and quality of light	

4.1.3 Assessment of ecological quality using phytoplankton

Most assessment systems currently in use are based solely on chlorophyll *a* as an indicator of overall phytoplankton abundance and biomass. This does not meet the requirements of the WFD, which stipulates that an assessment system must be based on composition as well as abundance and biomass.

The Swedish EPA (SEPA) uses a system based on chlorophyll *a*, seasonally calculated biovolume of diatoms, water-blooming cyanophytes and total phytoplankton biovolume. The system also includes a count of the number of potentially toxin-producing cyanophytes and the biovolume of the nuisance species *Gonyostomum semen*. Assessment is based on a selection of the aforementioned parameters that are compared with reference values for the relevant lake type (SEPA, 2000).

Another approach, followed by Solheim *et al*. (2004), is to examine the proportion of the total biomass of phytoplankton comprised by Chrysophytes and selected Cyanophytes. The proportion of Cyanophytes was found to increase with total phytoplankton biomass whereas the proportion of Chrysophytes decreased. Such an approach may help identify levels of biomass or chlorophyll *a* where an undesirable proportion of Cyanophytes may occur.

Reynolds *et al*. (2002) have developed an approach where taxa can be assigned to a functional group. Classification could then be carried out based on trophic preferences of observed assemblages relative to those of the type specific reference assemblages (Carvalho, 2005).

4.2 Methods

The methods used for phytoplankton sampling and enumeration were described in Chapter 3. Briefly: quantitative counts were based on a composite sample taken at 1 m intervals to a depth of 5 m. For lakes shallower thatn 5m a subsurface sample was used. Cells and colonies were enumerated following a transect count of a settling chamber using an inverted microscope. Net (53 μm mesh) samples were examined and a 4 - point scale of abundance applied to taxa. The analysis focused on

quantitative summer samples with the exception of Desmid abundance, which was from spring net samples.

4.3 Results

4.3.1 Development of a typology using phytoplankton composition and abundance

The approach taken to develop the typology had the following objectives:

1) To determine if distinct types were evident in phytoplankton community composition and abundance using 62 of the 69 candidate reference lakes.
2) To see if such 'biological types' were also distinct in terms of measured environmental variables.
3) To assign environmental boundaries that define distinct biological types.
4) To test and describe the resulting lake types in reference condition.

In order to perform an initial search for biological types, a cluster analysis was carried out on transformed ($x^{0.5}$) phytoplankton abundance (Figure 4.1). An indicator species analysis was used to prune the dendrogram to nine clusters. Figure 4.2 shows that nine clusters had a high number of significant indicator species (14) and that over all taxa, the indicator values were most significant (minimum p) between the nine clusters. In order to support the findings of the cluster analysis a non-metric multidimensional scaling (NMS) ordination was carried out using the Sorensen (Bray-Curtis) distance measure. Figure 4.3 shows that most of the groups were distinct in the NMS ordination with the exception of cluster 4 (△) and cluster 8 (◊) (same symbols as Figure 4.1 encircled).

The next step was to see if the nine clusters were distinct in terms of phytoplankton taxa and measured environmental variables. To do this, an indicator species analysis was carried out on the nine clusters to identify taxa that were significant indicators of a cluster in terms of phytoplankton abundance and composition (Table 4.2). In order to visualise differences in environmental variables between clusters, box-plots were drawn (Figure 4.4). An estimate of the lakes tendency to stratify was also included (see section 7.2.2 Statistical analysis for calculation) as an environmental variables because stratification can influence phytoplankton distribution. Table 4.3 provides a brief description of the clusters with reference to Table 4.2 and Figure 4.4.

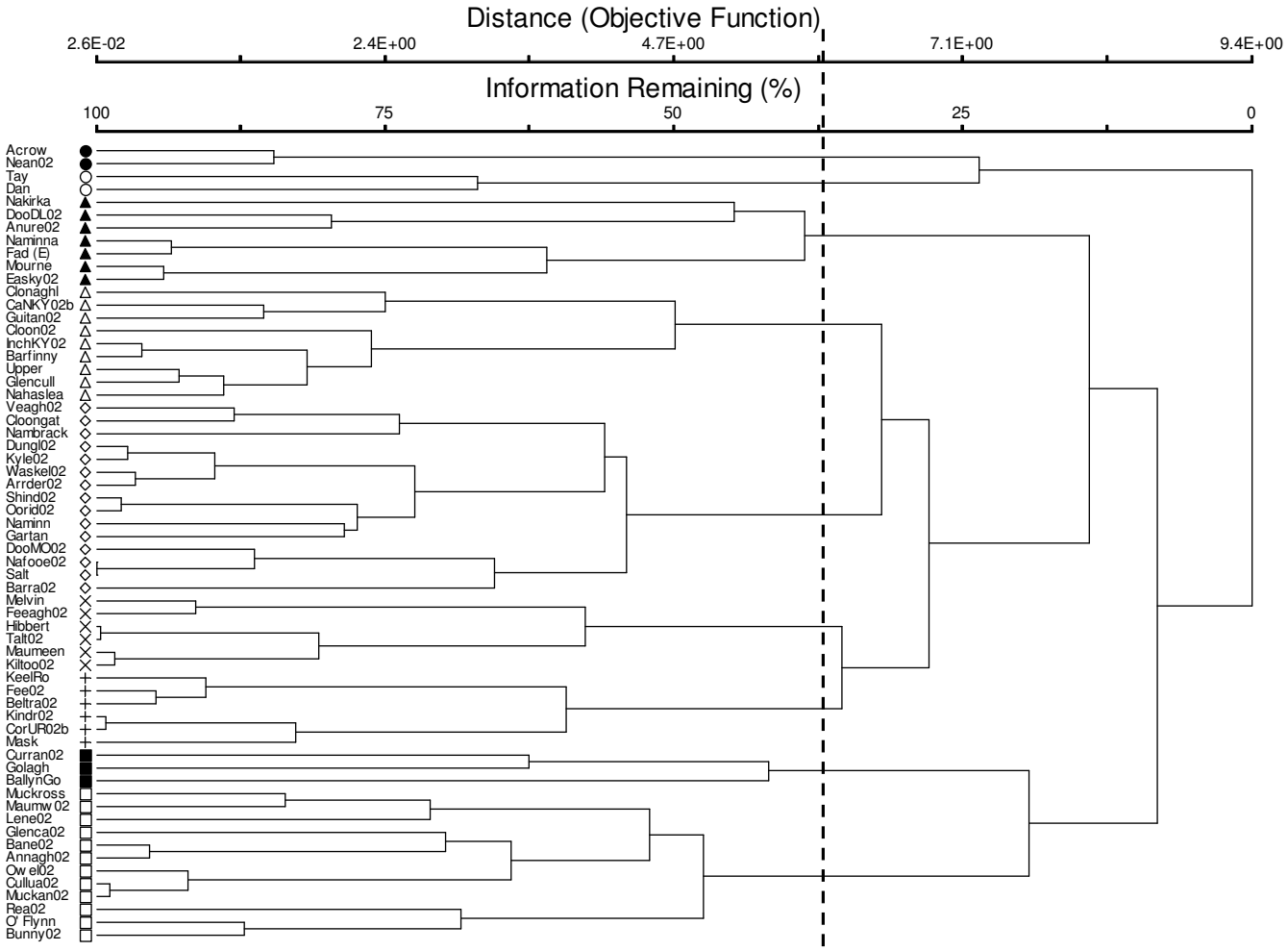


Figure 4.1 Dendrogram from cluster analysis of transformed ($x^{0.5}$) phytoplankton abundance (cells or colonies ml^{-1}) in reference lakes (n = 62). Sorensen (Bray-Curtis) distance measure was used with flexible beta (-0.25) linkage. Dashed line represents cut-off point for nine clusters.

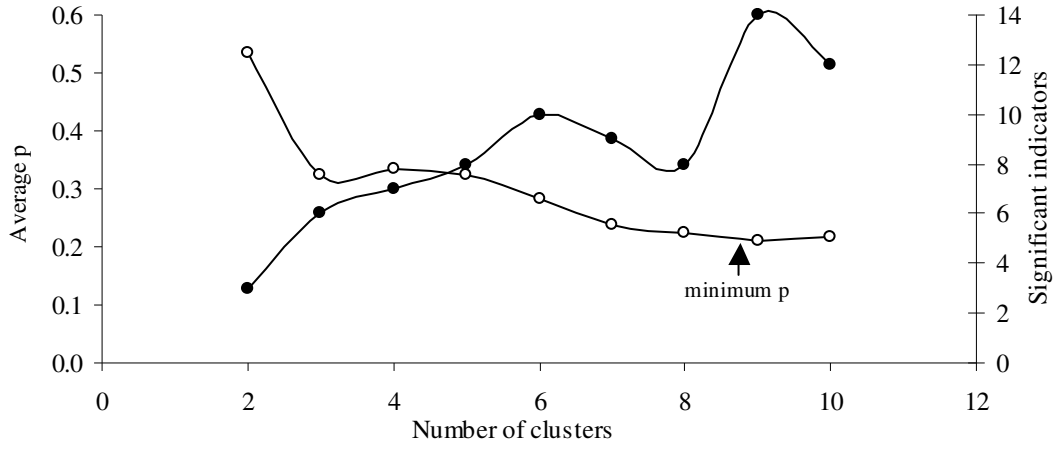


Figure 4.2 Average p for all taxa (○) and number of significant ($p \leq 0.05$) indicator species identified (●) from an indicator species analysis of clusters 2 to 10. Minimum p (0.21) was reached after nine clusters.

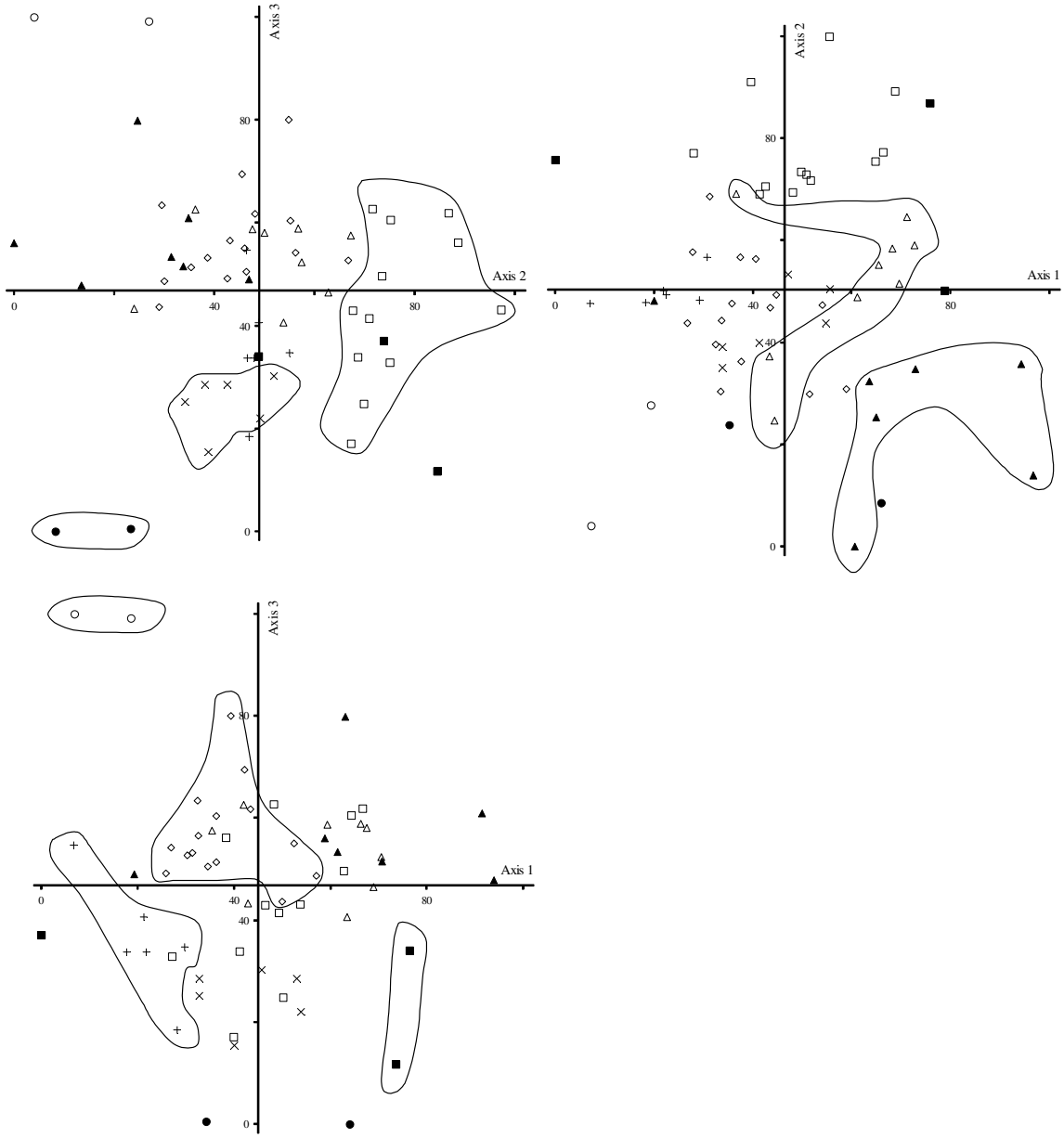


Figure 4.3 NMS ordination of reference lakes using transformed ($x^{0.5}$) phytoplankton abundance (cells or colonies ml^{-1}) (n = 62). Sorensen (Bray-Curtis) distance measure used; stress: 17.2. The proportion of variance explained by axis 2 was 33%, axis 3: 25% and axis 1: 22%. Symbols and overlays identify the nine clusters found in Figure 4.1.

Table 4.2 Taxa that were found to be significant indicators of clusters. %F = % frequency of occurrence in cluster, %RA = % relative abundance in cluster. p was determined by Monte Carlo test - proportion of 1000 randomised trials where the observed indicator value was equalled or exceeded (McCune *et al*., 2002).

Taxa	Cluster taxa indicative of	Indicator value (%)	%F	%RA	p
Ankistrodesmus spp.	1	64	100	64	0.003
Monoraphidium spp.	1	80	100	80	0.002
Closterium spp.	2	72	100	71	0.027
Merismopedia spp.	3	76	86	89	0.008
Staurastrum spp.	3	60	86	70	0.023
Cryptomonas spp.	5	31	100	31	0.025
Peridinium spp.	5	50	100	50	0.036
Ophiocytium spp.	5	61	67	91	0.004
Dictyosphaerium spp	7	38	67	57	0.050
Asterionella formosa	9	63	100	62	0.016
Synedra spp.	9	43	100	43	0.050
Diatoma spp.	9	62	67	92	0.013
Fragilaria crotonensis	9	73	83	88	0.024
Sphaerocystis spp.	9	50	67	74	0.023

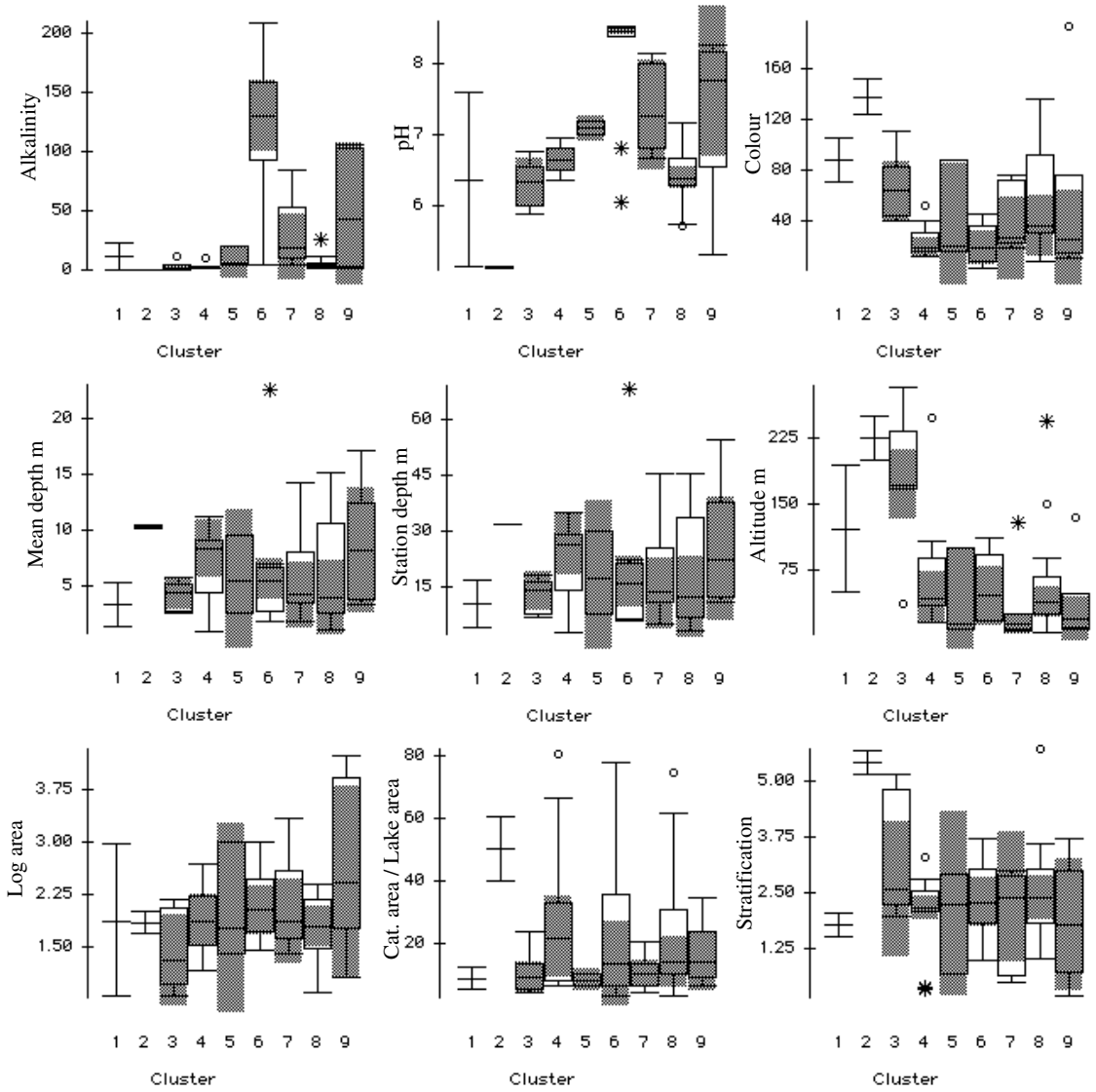


Figure 4.4 Box plots of clusters by alkalinity (mg l^{-1} $CaCO_3$), pH, colour (mg l^{-1} PtCo), predicted mean depth (m), station depth (m), altitude (m), log area (ha), catchment area / lake area and estimated tendency to stratify. Shaded areas represent 95% confidence limits.

Table 4.3 Observations on clusters using Table 4.2 and Figure 4.4.

Cluster	Description
Cluster 1 (●) n = 2	*Ankistrodesmus* and *Monoraphidium* were significant indicator taxa for this cluster. This cluster consisted of 2 lakes which were not distinct environmentally from the other clusters.
Cluster 2 (○) n = 2	Consisted of an upstream and downstream lake - Lough Tay and Dan and had a notably low pH (5.11 and 5.12). *Closterium* was found to be a significant indicator of this cluster, most likely owing to the high abundances when sampled (60-680 cells ml^{-1}). *Closterium* also occurred frequently in other clusters.
Cluster 3 (▲) n = 7	Colour was > 40 mg l^{-1} PtCo in the 7 lakes in this group, 6 of the lakes were above 160 m altitude. *Merismopedia* and *Staurastrum* spp. were significant indicator taxa. *Merismopedia* occurred frequently in other softwater lakes but had a notable concentration of abundance in this cluster (89% of abundance in 62 reference lakes) (Table 4.2).
Cluster 4 (△) n = 9	Cluster 4 was similar to the preceding clusters in having a low alkalinity. No significant indicator taxa were identified.
Cluster 5 (■) n = 3	*Cryptomonas*, *Peridinium* and *Ophiocytium* were significant indicator taxa for this group. The indicator value for *Cryptomonas* was low (31) which is not surprising given that it was ubiquitous, occurring in all 62 of the reference lakes sampled. Cluster 5 was not distinct environmentally from the other clusters.
Cluster 6 (□) n = 12	Cluster 6 had a much higher alkalinity and pH than the other clusters. Ten lakes had an alkalinity > 94 mg l^{-1} $CaCO_3$, 2 lakes appeared to be misclassified – Maumwee and Muckross having an alkalinity of 6 and 5 mg l^{-1} $CaCO_3$. No significant indicator taxa were identified although the centric diatoms *Cyclotella* and *Stephanodiscus* were often present at > 20 cells ml^{-1}.
Cluster 7 (×) n = 6	*Dictyosphaerium* was a weak indicator for this cluster (IV = 38%). The cluster was not distinct environmentally, although 3 of the 6 lakes had an alkalinity in the moderate-range 20-100 mg l^{-1} $CaCO_3$ (Figure 4.4).
Cluster 8 (◊) n = 15	Had no significant indicator taxa and did not appear to be significantly different in terms of measured environmental variables.
Cluster 9 (+) n = 6	Cluster 9 comprised a mix of 3 high alkalinity lakes: Mask, Corrib and Kindrum (all > 69 mg l^{-1} $CaCO_3$) and 3 lakes with alkalinities ranging from 2-19 mg l^{-1} $CaCO_3$. *Sphaerocystis* and 4 diatom taxa were found to be significant indicators (Table 4.2).

Based on a univariate examination of clusters by environmental variables, cluster 6 appears the most distinctive in having a high alkalinity and pH (Figure 4.4). Interestingly, no significant indicator taxa were found for this cluster, although it has been reported that phytoplankton of calcareous lakes tend to be made up of cosmopolitan species (Pearsall and Lind, 1942). Still, the fact that the majority of the high alkalinity lakes clustered out separately does indicate that the phytoplankton of these waters has a distinctive composition and abundance, if not characteristic indicator taxa. Cluster 2 was also distinctive, having a low pH and high altitude but this cluster only comprised two lakes in the same catchment (Lough Tay and the downstream Lough Dan). The phytoplankton composition of downstream lakes may

be similar to that of upstream lakes because of the physical transfer of large volumes of water, upstream lakes acting as a source of inoculate or by simply being subject to similar influences in a shared catchment. Cluster 3 was the only other group that appeared to be characterised by a distinct environmental difference in that most of the lakes were above 160 m altitude (Figure 4.4). Cluster 3 was also highly coloured, being somewhat distinct from the other clusters in having no lakes with a colour below 40 mg l^{-1} PtCo. Most of the other 6 clusters could be described as soft-water lakes with variable environmental characteristics.

Examining the differences in environmental factors between clusters is useful in that factors that exert a strong influence on the biological groups may be readily identified (e.g. alkalinity, Figure 4.4). However, it must also be considered that environmental factors may have compounding effects resulting in a distinct environmental type that is reflected in phytoplankton abundance and composition. For example, it may be expected that a combination of high colour, deep mean depth and long residence time (indicated by a small catchment to lake area ratio) may lead to a lower abundance of phytoplankton due to low light levels and naturally lower nutrient levels (with increased residence time). One way to examine clusters in terms of a combination of environmental variables is to perform a discriminant analysis – also known as canonical variates analysis (CVA). CVA determines which linear combination of environmental factors discriminates best between clusters and can indicate if clusters are different in terms of environmental factors (ter Braak and Smilauer, 2002).

Axes 1 and 2 of the CVA represented 66% of the variation in the relationship between clusters (referred to as species in CVA) and the environment (Table 4.4). Figure 4.5 shows the group centroids for each cluster. In support of the univariate examination of environmental variables, the group centroids and lakes (encircled) for clusters 2, 3 and 6 are located separately, indicating that they are distinct environmentally. Cluster 4 also appears to be somewhat distinct. In contrast, other clusters appeared to be less environmentally distinct, their group centroids being less closely surrounded by their clusters lakes. The arrows (Figure 4.5, Figure 4.6) and standardised canonical coefficients (Table 4.5) give an indication of the relative importance of each environmental factor in cluster separation. Alkalinity, altitude, catchment to lake area ratio and colour appeared to be the most important discriminant factors. In contrast,

mid-lake station depth and lake area were not found to be significant ($p > 0.05$) in separating clusters in the model (Table 4.6).

Selection of environmental factors to define the lake typology

One approach to selecting environmental factors to define a lake typology is to simply use those factors found to be significant in discriminating between the clusters in the CVA analysis: alkalinity, altitude, catchment to lake area ratio and colour (Table 4.6). However, the usefulness of each variable in forming a typology will be interpreted in turn.

Alkalinity was found to be the most important variable distinguishing between clusters and will be used to form a typology. Altitude was also found to be significant in the CVA, but it was not selected for use in defining a typology as its importance may be overemphasised by cluster 2 which comprised an upstream and downstream lake in a high altitude catchment. Given the low range of altitude in the reference lakes sampled (Figure 4.4), the influence of altitude is likely to be related to a surrogate variable such as alkalinity, rather than having a direct influence.

The catchment to lake area ratio was also significant in discriminating between clusters in CVA. The influence of this variable is likely to be related to residence time and its influence on background (reference) nutrient concentrations. It was not included in the typology at this time as it is only an approximate indicator of residence time. When information on residence time and low level nutrient fluctuation becomes available, residence time should be evaluated for inclusion in a typology. Colour was found to be significant in the CVA analysis, although there was considerable overlap in colour between clusters (Figure 4.4). Colour was selected as a factor to form a typology, as it is likely to affect phytoplankton through its influence on light quantity and quality.

Interestingly, depth was not found to have a significant influence in discriminating between clusters ($p = 0.252$, Table 4.6). However, this exercise only considered lakes of potential reference condition. It is likely that depth would influence how phytoplankton in a lake would respond to excessive nutrient loading. It is desirable that a typology should also include natural environmental factors that control the

response to eutrophication, even though differences may not be apparent in reference phytoplankton communities. With increasing nutrient input, deeper lakes are likely to show a smaller increase in phytoplankton abundance than shallow lakes owing to less light availability when mixed to deeper depths. Mean depth was therefore considered essential to form a typology. In summary: alkalinity, colour and depth were considered to be the most important factors necessary to form a typology. The next step is to define values of the environmental variables that will form the boundaries of the typology.

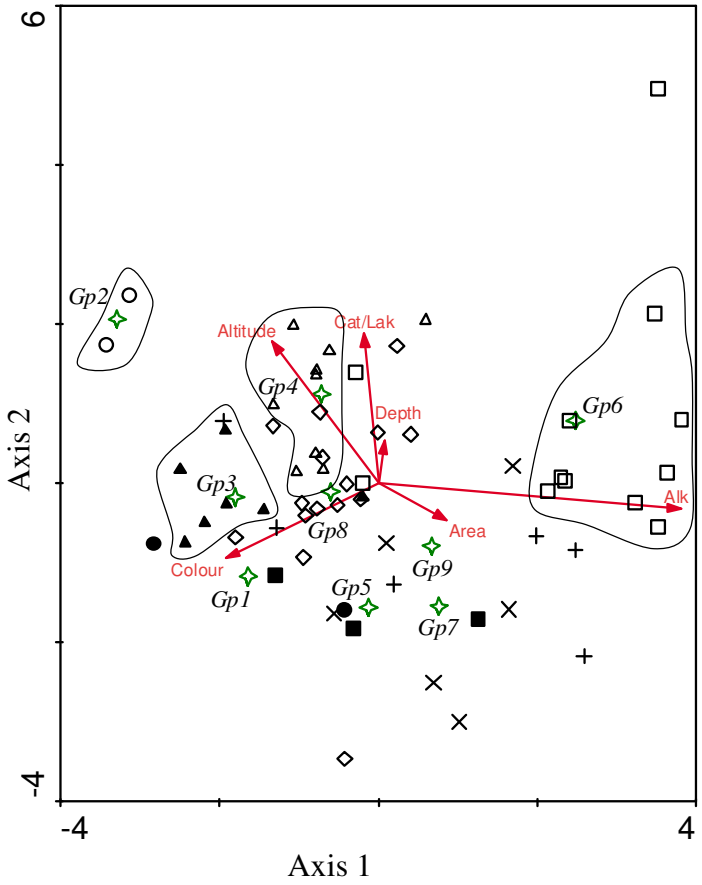


Figure 4.5 Axis 1 and 2 of CVA plot of nine clusters identified in Figure 4.1. Group centroids (✧) are labelled. Groups 2, 3, 4 and 6 are encircled.

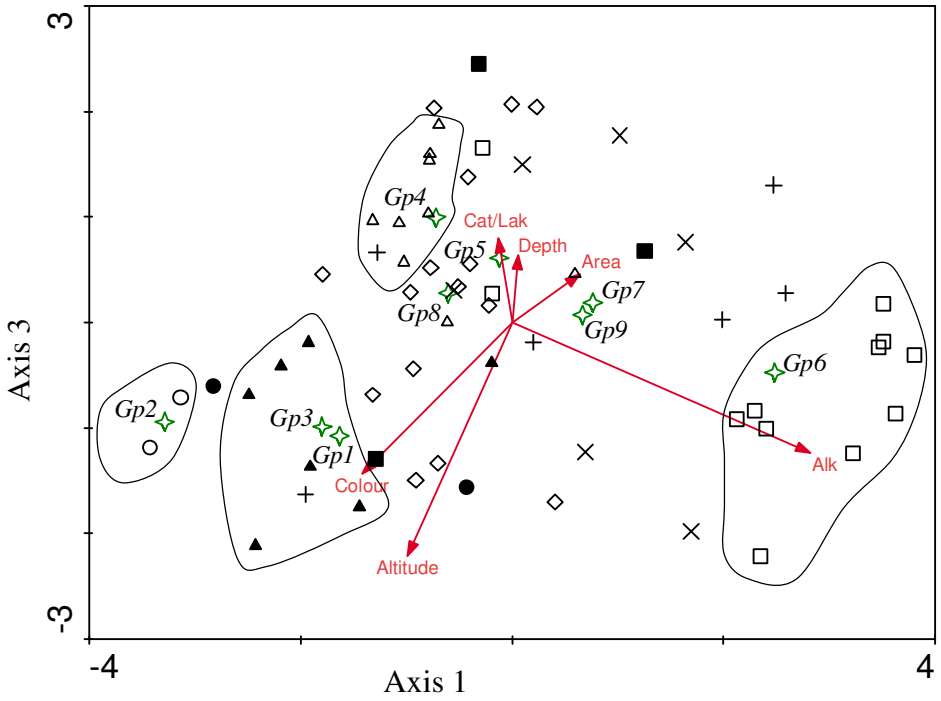


Figure 4.6 Axis 1 and 3 of CVA plot of nine clusters identified in Figure 4.1. Group centroids (✧) are labelled. Groups 2, 3, 4 and 6 are encircled.

Table 4.4 Summary statistics of CVA axes.

	Axis 1	Axis 2	Axis 3	Axis 4
Eigenvalues	0.693	0.473	0.288	0.202
Species-environment correlations	0.832	0.688	0.537	0.450
Cumulative percentage variance				
of species data	8.7	14.6	18.2	20.7
of species-environment relation	39.3	66.2	82.5	94.0

Table 4.5 Standardised canonical coefficients for transformed (Log x + 2) variables.

	Axis 1	Axis 2	Axis 3	Axis 4
Alkalinity	1.526	0.037	-0.961	0.392
Colour	-0.510	-0.904	-0.867	0.783
Station depth	-0.112	0.042	-0.084	0.882
Altitude	-0.335	1.047	-0.782	-0.219
Area	-0.280	-0.181	0.108	0.067
Catchment area / Lake area	0.201	1.260	0.218	0.333

Table 4.6 Summary of automatic forward selection in CVA. Marginal effects lists variables in order of variance explained by each variable alone. Conditional effects lists variables in order of inclusion in model along with additional variation explained and whether it was significant ($p \leq 0.05$). Variables were transformed (Log x + 2). Cat/Lak = catchment to lake area ratio.

Marginal Effects		Conditional Effects			
Variable	Lambda-1	Variable	Lambda-A	p	F
Alkalinity	0.65	Alkalinity	0.65	0.002	5.27
Colour	0.38	Altitude	0.30	0.010	2.56
Altitude	0.37	Cat/Lak	0.28	0.018	2.42
Cat/Lak	0.22	Colour	0.30	0.012	2.65
Area	0.19	Station depth	0.15	0.252	1.26
Station depth	0.12	Area	0.08	0.630	0.77

Selection of lake type boundaries

Boundaries must be defined in terms of environmental variables in order to separate types of lakes that are biologically distinct. In order to define a lower alkalinity band for the typology, the relative abundance of Desmids in net samples was examined as the majority of Desmids are well known to have a preference for soft-water (Hutchinson, 1967). Figure 4.7 shows that the relativised abundance of Desmids increased rapidly in reference lakes at alkalinities slightly below 10 mg l^{-1} $CaCO_3$. The lower alkalinity boundary to the typology was therefore set at 10 mg l^{-1} $CaCO_3$. There was a notable decline in the relativised abundance of Desmids at lower alkalinity and pH (Figure 4.7, Figure 4.8). An upper alkalinity band was set at 100 mg l^{-1} $CaCO_3$ as this was close to the lower value of 94 mg l^{-1} $CaCO_3$ for cluster 6 (excluding 2 misclassified sites) (Figure 4.4, Table 4.3).

The boundary for colour was set at 40 mg l^{-1} PtCo, as this was the value that marked the lower end of the distribution of cluster 3, the cluster that appeared most influenced by colour (Table 4.3, Figure 4.5). The colour value of 40 mg l^{-1} PtCo was also reasonably typical of Irish lakes: Free *et al*. (2000) recorded a median colour of 38 mg l^{-1} PtCo in a survey of 199 lakes in summer.

In the absence of a clear influence of depth on reference lakes in the preceding analysis, consideration was given to setting the mean depth categories at the compensation depth (depth where 1% of surface light remains) for a reference lake of

typical colour. Using a colour of 40 mg l^{-1} PtCo the reference compensation depth was predicted using equations in Free *et al*. (2005) to be 5 m. However, this was lowered to 4 m as this coincides with the mean depth figure chosen for the macrophyte typology in Chapter 5. This represents a depth where 2.6% of surface light would remain, and is therefore not markedly different from the compensation depth.

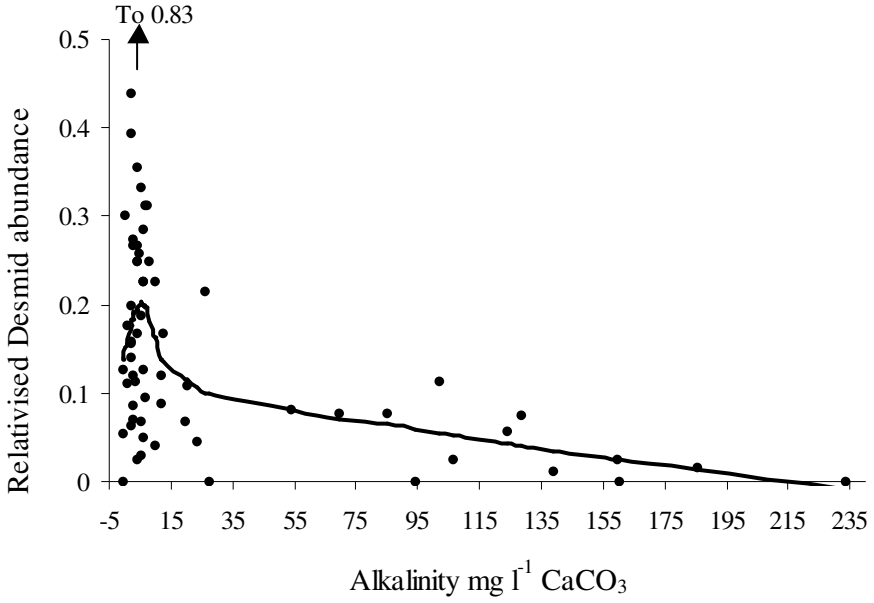


Figure 4.7 Relativised Desmid abundance against alkalinity in 63 reference lakes sampled with a 53 µm net in April or May 2001 or 2002. Lowess smoothed line fitted.

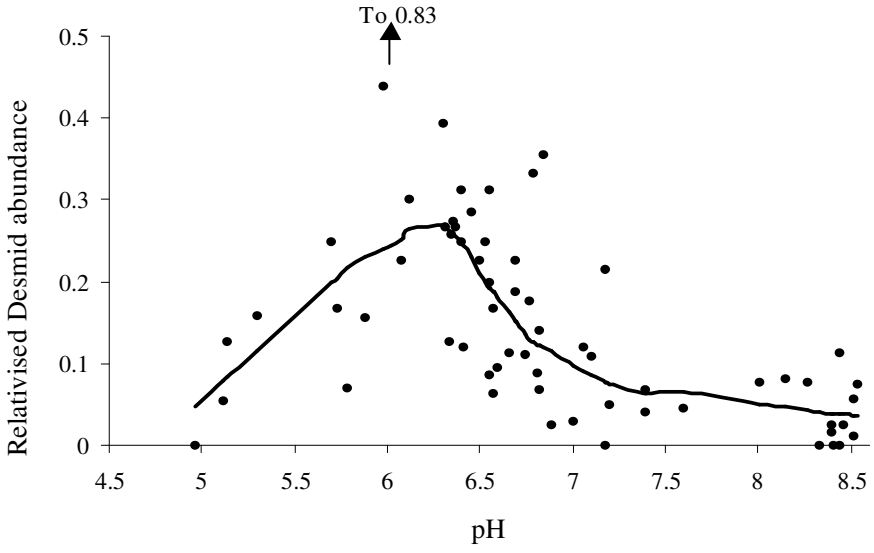


Figure 4.8 Relativised Desmid abundance against pH in 63 reference lakes sampled with a 53 µm net in April or May 2001 or 2002. Lowess smoothed line fitted.

Testing the proposed typology and comparing it with the System A typology

In order to determine if the proposed typology above resulted in types that were biologically distinct, pair-wise multi-response permutation procedure (MRPP) tests were carried out on phytoplankton abundance. Table 4.7 shows the A values (chance-corrected within-group agreement) of the pair-wise tests. The A values indicate the homogeneity within a group to that expected by chance: 1 equals complete within group homogeneity whereas an A of 0 equals within group heterogeneity equal to that expected by chance (McCune *et al*., 2002). Table 4.7 shows that most significant differences were found between the low alkalinity type (< 10 mg l^{-1} $CaCO_3$) and the high alkalinity type (> 100 mg l^{-1} $CaCO_3$).

The Water Framework Directive requires that if a system B typology is developed by a member state, it must achieve at least the same degree of differentiation as would the application of the default system A typology (CEC, 2000). A detailed statistical comparison of both typology systems would be difficult, but only a broad comparison is required (REFCOND, 2003). MRPP tests may allow some comparison of the proposed typology with that of system A (Table 4.7, Table 4.8). An indication of the degree of differentiation achieved by each typology system is given by comparing the overall A values from the MRPP analysis. The proposed typology for phytoplankton had an overall A value of 0.12 ($p < 0.001$) whereas the default system A typology had an overall A of 0.14 ($p < 0.001$). This actually indicates that the default system A would be better at partitioning natural variation than the proposed typology. However, if the proposed typology is simplified even further by removing depth, the overall A value is 0.16, which is a slight improvement on the system A typology. Such a typology may be overly simple, having just 3 alkalinity bands (< 10, 10-100, > 100 mg l^{-1} $CaCO_3$) and two colour bands (< 40, > 40 mg l^{-1} PtCo). It was noted earlier that even though depth was not found to be a significant factor differentiating clusters, it is likely to be important in governing the response to nutrient input. Depth may therefore be retained in the phytoplankton typology, as the purpose of a typology is to help detect ecological change.

Table 4.7 Results (A values) of pair-wise MRPP tests for the proposed types using transformed ($x^{0.5}$) phytoplankton abundance and the Sorensen (Bray-Curtis) distance measure (rank transformed matrix). Significant ($p \leq 0.05$) differences are in bold. An A of 1 = complete within group homogeneity, an A of 0 = within group heterogeneity equal to that expected by chance (McCune *et al*., 2002). Tests were carried out where group n was > 1 and total n was > 4.

Type	n	< 10 Alk < 4 m < 40 Col	< 10 Alk < 4 m > 40 Col	< 10 Alk > 4 m < 40 Col	< 10 Alk > 4 m > 40 Col	10-100 Alk < 4 m < 40 Col	10-100 Alk < 4 m > 40 Col	10-100 Alk > 4 m < 40 Col	10-100 Alk > 4 m > 40 Col	> 100 Alk > 4 m < 40 Col
< 10 Alk < 4 m < 40 Col	3									
< 10 Alk < 4 m > 40 Col	12	-0.01								
< 10 Alk > 4 m < 40 Col	16	-0.03	0.03							
< 10 Alk > 4 m > 40 Col	8	-0.02	-0.02	**0.05**						
10-100 Alk < 4 m < 40 Col	2	**0.04**	0.02	0.00	0.08					
10-100 Alk < 4 m > 40 Col	2	-0.09	0.00	-0.04	0.01	n/a				
10-100 Alk > 4 m < 40 Col	4	0.00	0.05	0.04	0.08	-0.11	-0.11			
10-100 Alk > 4 m > 40 Col	4	0.12	0.06	**0.07**	0.08	-0.05	-0.116	0.02		
> 100 Alk > 4 m < 40 Col	9	**0.15**	**0.18**	**0.15**	**0.20**	-0.02	0.09	0.05	0.06	

Table 4.8 Results (A values) of pair-wise MRPP tests for system A types using transformed ($x^{0.5}$) phytoplankton abundance and the Sorensen (Bray-Curtis) distance measure (rank transformed matrix). Significant ($p \leq 0.05$) differences are in bold. Organic = peat ≥ 50% of catchment, Calcareous = limestone ≥ 20% of catchment.

Type	n	Organic < 3 m < 50 ha < 200 m	Organic < 3 m > 50 ha < 200 m	Organic 3 - 15 m < 50 ha < 200 m	Organic 3 - 15 m < 50 ha > 200 m	Organic 3 - 15 m > 50 ha < 200 m	Organic 3 - 15 m > 100 ha < 200 m	Siliceous 3 - 15 m < 50 ha < 200 m	Calcareous 3 - 15 m > 50 ha < 200 m	Calcareous 3 - 15 m > 100 ha < 200 m	Calcareous 3 - 15 m > 1000 ha < 200 m
Organic < 3 m < 50 ha < 200 m	8										
Organic < 3 m > 50 ha < 200 m	5	-0.06									
Organic 3 - 15 m < 50 ha < 200 m	3	0.02	-0.03								
Organic 3 - 15 m < 50 ha > 200 m	4	-0.02	-0.05	0.06							
Organic 3 - 15 m > 50 ha < 200 m	7	**0.08**	0.02	-0.02	**0.11**						
Organic 3 - 15 m > 100 ha < 200 m	14	**0.05**	-0.01	-0.06	0.06	0.03					
Siliceous 3 - 15 m < 50 ha < 200 m	3	0.04	-0.07	-0.03	**0.10**	0.07	0.05				
Calcareous 3 - 15 m > 50 ha < 200 m	2	**0.18**	0.10	**0.07**	**0.16**	0.13	**0.13**	**0.12**			
Calcareous 3 - 15 m > 100 ha < 200 m	4	**0.19**	**0.12**	0.18	**0.15**	**0.20**	**0.17**	**0.22**	0.03		
Calcareous 3 - 15 m > 1000 ha < 200 m	3	**0.16**	0.05	0.14	**0.18**	0.12	0.06	0.03	**0.20**	0.08	

Describing the typology in reference condition

A description of the proposed typology was achieved by calculating indicator values of the phytoplankton across the types (Table 4.9). A high (> 50%) indicator value (IV) typically indicates that a taxon is both frequent and abundant in a type. As expected, low alkalinity types (< 10 mg l^{-1} $CaCO_3$) had more Desmids than the other types. *Merismopedia* had its highest IV recorded in low alkalinity coloured lakes. Of the dinoflagellates (Pyrrophyta), *Gymnodinium* had a higher IV in low alkalinity lakes whereas *Ceratium* had a higher IV at higher alkalinities. Higher alkalinity bands tended to have more diatoms (Bacillariophyta). The highest alkalinity band (> 100 mg l^{-1} $CaCO_3$) was typically comprised of marl lakes and was characterised by higher amounts of *Peridinium* spp. and *Dinobryon* spp.

Mean and standard deviation values for the phytoplankton index (described in the following section) and summer chlorophyll *a* are presented in Table 4.9. The phytoplankton index appeared to be similar across the reference types and also had low within-type variation. Summer chlorophyll *a* typically had a concentration of approximately 4 μg l^{-1} in the reference lakes. Chlorophyll *a* was higher but more variable in two of the moderate alkalinity types (10 - 100 mg l^{-1} $CaCO_3$). This may indicate that some of these lakes deviated from reference condition: some difficulty was encountered in finding reference lakes for this type.

Table 4.9 Description of phytoplankton typology (summer samples). Indicator values, average phytoplankton index score and chlorophyll *a* for the 9 types where n was > 1. Indicator values are presented for the most common taxa in the reference lakes (occurring in ≥ 40% of lakes in an alkalinity band but including all Desmids).

	< 10 Alk < 4 m > 40 Col	< 10 Alk < 4 m < 40 Col	< 10 Alk > 4 m > 40 Col	< 10 Alk > 4 m < 40 Col	10-100 Alk < 4 m > 40 Col	10-100 Alk < 4 m < 40 Col	10-100 Alk > 4 m > 40 Col	10-100 Alk > 4 m < 40 Col	> 100 Alk > 4 m < 40 Col
n	12	3	8	16	2	2	4	4	9
Cyanophyta									
Anabaena spp.	2	1	1	1	3	1	3	83	0
Merismopedia spp.	25	3	27	3	0	0	0	0	0
Coelosphaerium/Gomphosphaeria spp.	1	0	0	0	19	3	12	0	7
Anabaena flos-aquae	1	0	1	0	0	0	12	3	20
Bacillariophyta									
Melosira/Aulacoseira spp.	0	2	1	0	51	0	16	1	3
Cyclotella/Stephanodiscus spp.	2	28	2	7	4	16	22	5	14
Asterionella formosa	10	2	1	5	9	32	18	2	6
Synedra spp.	2	1	2	2	1	25	12	9	26
Tabellaria fenestrata	4	0	6	3	0	8	26	0	1
Tabellaria flocculosa	19	10	19	7	3	2	0	0	0
Cryptophyta									
Cryptomonas spp.	8	5	8	10	8	8	17	23	14
Chrysophyta									
Dinobryon spp.	3	4	2	9	2	17	2	6	46
Pyrrophyta									
Ceratium spp.	1	0	0	2	3	32	11	16	7
Peridinium spp.	1	1	1	5	0	12	4	1	30
Gymnodinium spp.	16	12	6	21	7	1	2	5	8
Chlorophyta									
Botryococcus spp.	20	10	4	4	0	36	1	6	4
Scenedesmus spp.	1	11	0	3	6	1	31	9	7
Quadrigula/Elakatothrix spp.	4	33	10	1	2	3	1	10	0
Monoraphidium spp.	4	0	1	1	1	23	28	6	6
Ankistrodesmus spp.	9	2	14	3	3	2	26	1	4
Oocystis spp.	8	0	0	1	7	8	1	12	1
Chlorophyta - Desmids									
Spondylosium planum	0	0	0	38	0	0	0	0	0
Closterium spp.	3	2	25	1	1	1	9	0	0
Cosmarium spp.	27	2	0	1	0	6	0	0	0
Staurastrum spp.	19	0	12	3	0	0	4	0	0
Staurodesmus spp.	5	4	3	10	0	0	3	0	0
Phytoplankton index	0.94	1.00	0.92	0.92	0.90	0.96	0.84	0.96	0.91
Standard deviation	(0.11)	(0.00)	(0.10)	(0.05)	(0.14)	(0.06)	(0.09)	(0.04)	(0.06)
Chlorophyll *a* µg l^{-1}	4.3	3.2	3.9	4.1	3.4	6.8	7.9	4.0	4.4
Standard deviation	(2.3)	(1.1)	(2.2)	(1.7)	(1.5)	(4.6)	(1.2)	(1.4)	(2.8)

4.3.2 Development of a phytoplankton index for ecological assessment

An assessment system was developed for summer phytoplankton based on quantitative samples. For the purpose of metric development, summer was considered to be June, July, August and early September. Summer was chosen as it is the period when the excessive growth of phytoplankton is most often manifested. In addition, the response of phytoplankton to nutrient enrichment in spring may be complicated by the availability of silica and light. The assessment system was developed so that it meets the requirements of the Water Framework Directive in that it incorporates information on the composition, abundance and biomass of the phytoplankton. The latter was indicated by chlorophyll *a* as an overall measure of biomass. The steps taken to produce the index were:

1) Selected 9 'eutrophic' taxa or groups of taxa that showed a positive growth response to TP as: *Scenedesmus* spp., *Pediastrum* spp., *Anabaena* spp., *Cryptomonas* spp., *Rhodomonas* spp., *Aphanizomenon* spp., *Oocystis* spp., *Sphaerocystis* spp. and *Melosira* + *Aulacoseira* spp.
2) Defined (in cells or colonies ml^{-1}) and scored the response of each of the above groups from 1 to 0.1, descending with increasing TP concentration (Table 4.10).
3) Scored the response of chlorophyll *a* to TP from 1 to 0.1 (descending with anthropogenic pressure (TP) as required by the WFD) (Table 4.10).
4) Averaged the scores to produce a phytoplankton index.

The relationship between the phytoplankton index and TP is shown in Figure 4.9 and Figure 4.10. The r^2 between the phytoplankton index and transformed (log x + 1) TP was 0.67 ($p \leq 0.0001$, n = 129) with 3 outlying lakes removed. Reference lakes had higher values of the index (Figure 4.9). Although the index includes chlorophyll *a*, which has a well-known relationship with TP (Dillon and Rigler, 1974), the phytoplankton index is not overly influenced by chlorophyll *a* as it is averaged alongside up to 9 scores derived from phytoplankton abundance. Averaging the score of chlorophyll *a* with the other scores had the beneficial effect of reducing variation in the phytoplankton index in reference condition (cf. Figure 4.9 with Figure 4.11).

The phytoplankton index also had a significant (r^2 = 0.61, $p \leq 0.0001$, n = 132) relationship with transformed (log x + 1) sample chlorophyll *a* (Figure 4.12). The data used to test the relationship between the phytoplankton index and TP and chlorophyll *a* (Figure 4.9 to Figure 4.12) were used to develop the index. While it is useful to run a model's prediction on the data used in its generation, it is better to test the model with independent data. Summer phytoplankton data were available for 30 lakes sampled in summer (mainly July) 1996 (Irvine *et al*., 2001). Thirteen of these lakes were also sampled by this project but the 4 to 6 year gap between samples should still allow a relatively independent test of the model. Figure 4.13 shows that the phytoplankton index had reasonable success in detecting eutrophication in the set of independent lakes (r^2 = 0.62, $p \leq 0.0001$, n = 30). However, Lough Ramor had a higher value of the phytoplankton index than would be expected for its very high mean TP concentration (88 $\mu g\ l^{-1}$, Figure 4.13). This would caution against the uncritical use of the phytoplankton index. Examination of the phytoplankton count data for Lough Ramor revealed high concentrations of *Melosira/Aulacoseira* spp. (431 colonies ml^{-1}) and *Anabaena* spp. (122 colonies ml^{-1}), which may allow a lower value of the index to be awarded based on expert opinion.

Ideally the phytoplankton index should be similar over the summer months so that an assessment from one or two samples would be sufficient to classify the lake. Table 4.11 shows that the index was similar during the summer months in the three lakes examined, with the exception of June in Lough Gowna, which had a high value. This was probably caused by the uncharacteristically low chlorophyll *a* concentration at this time. Zooplankton grazing may have caused this, as zooplankton total abundance was at a maximum in the preceding month (Caroni, 2000). The assessment system would be likely to be improved by the incorporation of information on zooplankton.

The phytoplankton index appeared to work across the typologies although the importance of typology factors may be more apparent in a larger dataset. It might be expected that deeper lakes would have a higher phytoplankton index than shallow lakes for a given TP owing to less light availability with full water-column mixing. It was not possible to investigate this with the current dataset because most deep lakes tended to be located in the uplands and have low TP concentrations. Ideally, more depth profiles from summer months would allow calculation of the mixing depth.

Table 4.10 Scores that are awarded and averaged to give the phytoplankton index. Scores are applied to the natural unit of occurrence (cells or colonies ml^{-1}) when taxa are present only. The index is only applied to samples from June, July, August and early September. Melo/Aul = *Melosira* and *Aulacoseira* spp.

SCORE	1.0	0.9	0.8	0.7	0.6	0.5	0.4	0.3	0.2	0.1
Scenedesmus spp.	>0 < 3.6	4.2	8.4	11.6	15.2	16.8	19.4	22.1	26.0	> 26.0
Pediastrum spp.	>0 < 2.1	2.3	4.4	6.8	13.7	16.0	19.4	20.3	21.2	> 21.2
Anabaena spp.	>0 < 8.4	12.3	19.4	24.0	30.3	34.8	37.2	39.7	41.0	> 41.0
Melo/Aul spp.	>0 < 2.6	5.8	9.0	15.2	19.4	21.2	24.0	26.0	28.1	> 28.1
Cryptomonas spp.	>0 < 28.1	42.3	77.4	121.0	196.0	265.7	324.0	400.0	449.4	> 449.4
Rhodomonas spp.	>0 < 64.0	114.5	187.7	259.2	313.3	364.8	396.0	412.1	436.8	> 436.8
Aphanizomenon spp.		>0 < 4.0	5.8	9.0	12.3	16.0	19.4	22.1	> 22.1	
Oocystis spp.	>0 < 1.9	2.9	4.4	5.8	7.3	8.4	10.2	11.6	13.0	> 13.0
Sphaerocystis spp.	>0 < 2.0	3.8	4.8	5.1	> 5.1					
Summer Chl *a* µg l^{-1}	>0 < 5.2	8.5	10.8	12.9	14.5	16.8	17.5	18.1	19.4	>19.4

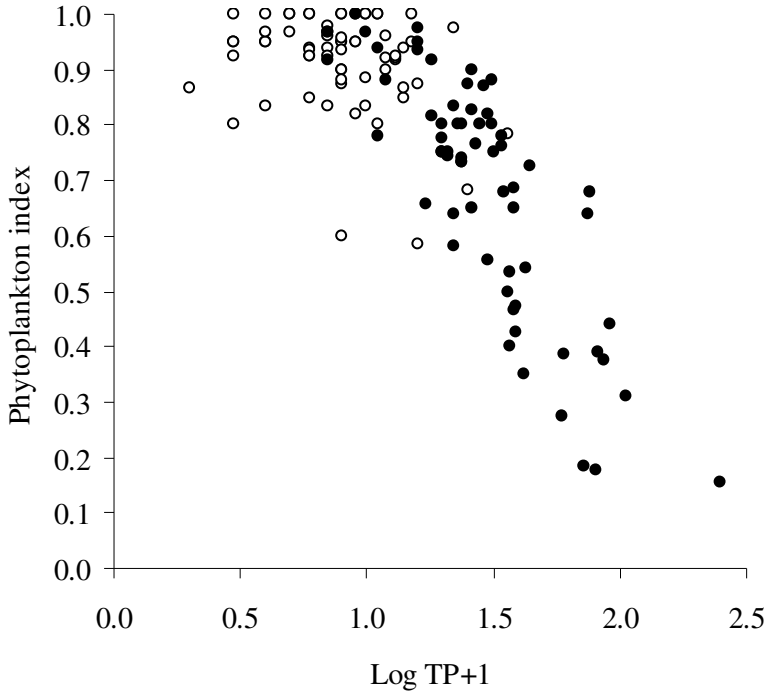


Figure 4.9 Relationship between the phytoplankton index and sample TP µg l^{-1} (n = 129). ○ = reference lakes, ● = non-reference lakes.

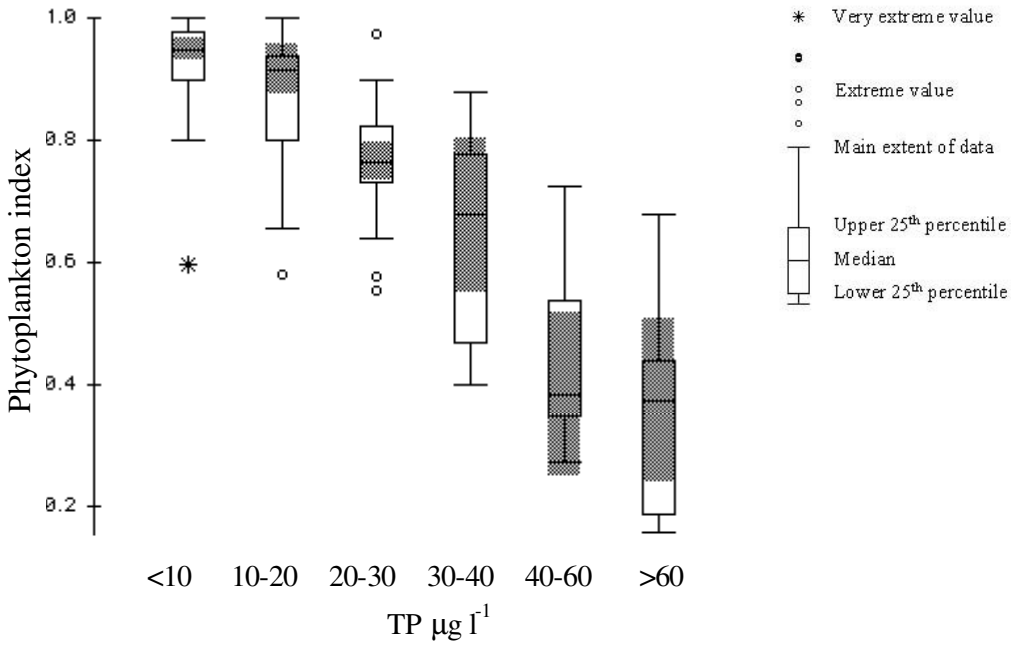


Figure 4.10 Box plot of the phytoplankton index and sample (summer) TP µg l^{-1} (n = 129). Shaded areas represent 95% confidence intervals.

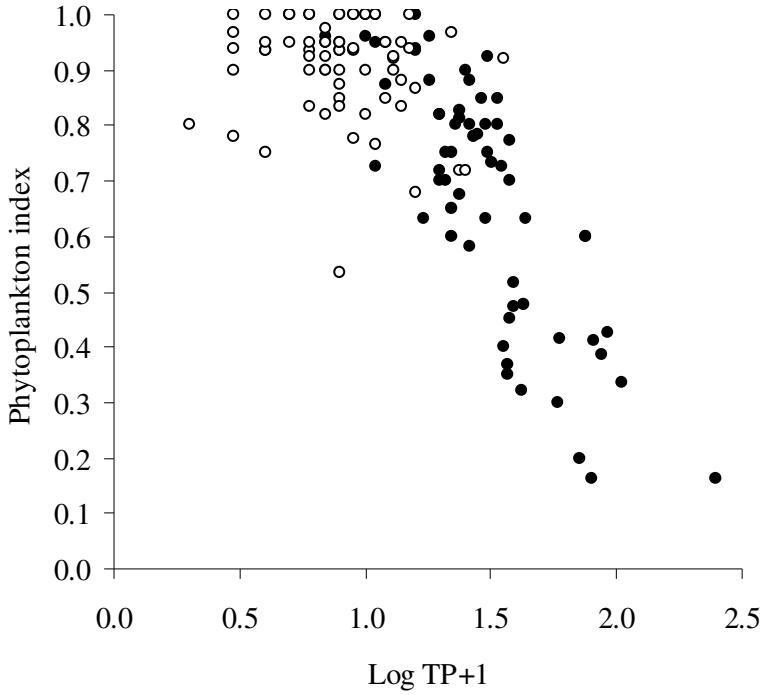


Figure 4.11 Relationship between the phytoplankton index (excluding chlorophyll *a* score) and sample TP µg l^{-1} (n = 129). ○ = reference lakes, ● = non-reference lakes.

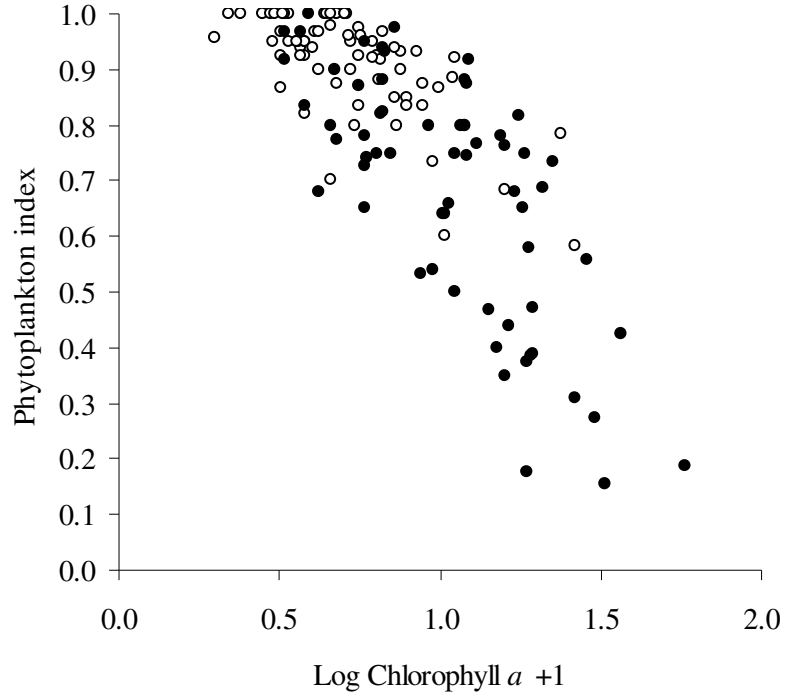


Figure 4.12 Relationship between the phytoplankton index and sample chlorophyll *a* µg l^{-1} (n = 132). ○ = reference lakes, ● = non-reference lakes.

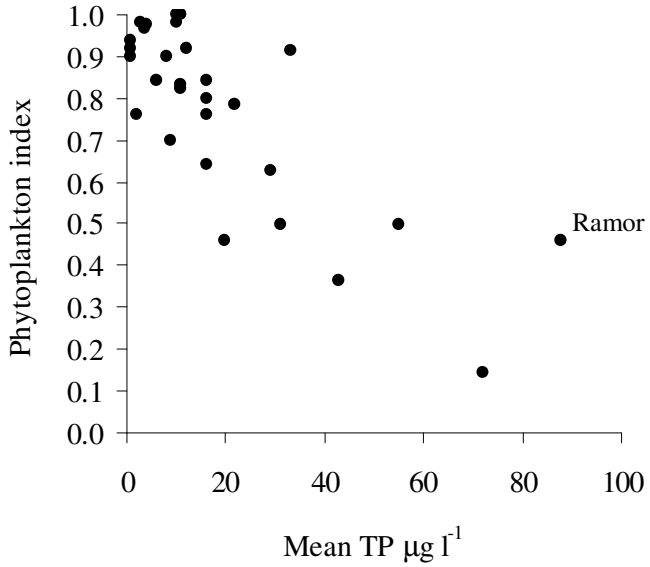


Figure 4.13 Relationship between the phytoplankton index and mean (1996-7) TP µg l^{-1}. Phytoplankton data are from 30 lakes sampled in June or July 1996 (Irvine *et al*., 2001).

Table 4.11 Variation in the phytoplankton index and chlorophyll *a* in 3 lakes over 4 'summer' months in 1996.

Lake	June	July	Aug.	Sep.
	Phytoplankton index			
Feeagh	0.90	n/a	0.86	0.96
Ramor	0.53	0.48	0.36	0.56
Gowna	0.80	0.30	0.40	0.42
	Chlorophyll *a* $\mu g\ l^{-1}$			
Feeagh	2.0	1.2	4.2	1.7
Ramor	88.1	59.8	127.2	87.4
Gowna	6.8	26.1	36.4	25.1

The phytoplankton index was developed based on a univariate examination of 'eutrophic' taxa along a pressure gradient (TP). This may be criticised as a system that loses information by not incorporating information on the whole phytoplankton population. In order to see if the phytoplankton index was useful in summarising changes in communities, a non-metric multidimensional scaling ordination (NMS) was carried out using transformed ($x^{0.5}$) phytoplankton abundance. Figure 4.14 shows the ordination with the environmental vectors overlain. The phytoplankton index is represented in the ordination by a vector and by assigning a colour to each lake based on a division of the index into 5 evenly spaced classes. Figure 4.14 indicates that differences in phytoplankton composition and abundance that were related to a nutrient gradient (as indicated by the TP vector) were also related to the phytoplankton index (as shown by the gradation in colour and also the positioning of the phytoplankton index vector). Hence, the index seems effective in expressing information about the phytoplankton community along a pressure gradient.

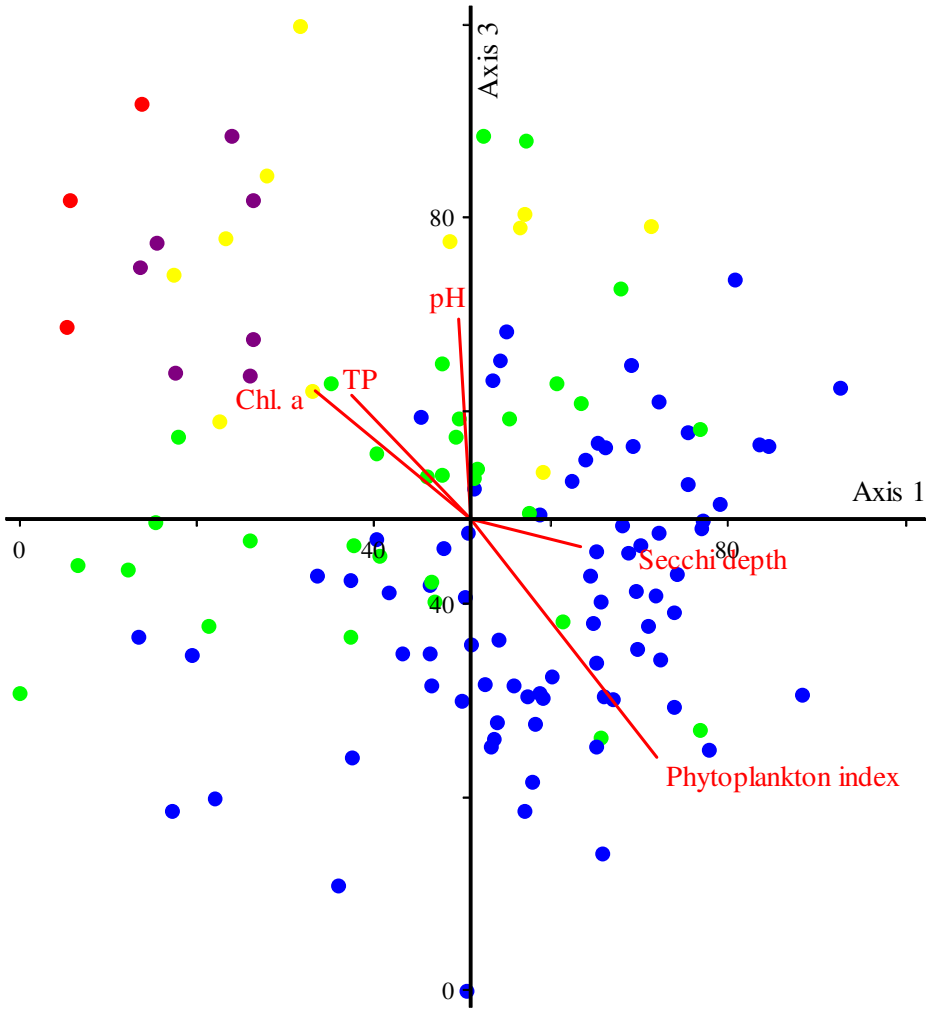


Figure 4.14 Axis 1 and 3 of NMS ordination of lakes using transformed ($x^{0.5}$) summer phytoplankton abundance. Symbols are coloured by 5 divisions of the phytoplankton index: 1-0.8 blue, 0.8-0.6 green, 0.6-0.4 yellow, 0.4-0.2 purple, 0.2-0.0 red. Stress: 19. The proportion of variance represented by axis 1 was 28.6%, axis 2: 22.3% and axis 3: 25.1%. The orientation and length of vectors indicate the direction and strength of the relationship of environmental factors and the phytoplankton index with ordination scores. n = 131.

4.4 Discussion

4.4.1 Typology

The development of a biologically based typology is the first step in developing an ecological assessment system for phytoplankton. Alkalinity, colour and depth were chosen as factors to form a typology for the phytoplankton. Alkalinity is not a parameter prescribed by the WFD system A or B but has been adopted as a convenient surrogate of geology. Colour was included as it is likely to have an influence on phytoplankton owing to its influence on the quantity and quality of light. Although depth was not found to have a major influence on reference phytoplankton

communities, it was included in the typology as it is likely to help partition metric response to pressure. Increased nutrient input would be likely to lead to a higher phytoplankton biomass in shallow rather than deep lakes.

Interestingly, the proposed typology achieved no greater success in partitioning variation in reference phytoplankton communities than the default system A. This may indicate that the system A of the WFD may be a suitable typology for phytoplankton. Alternatively, the multitude of factors that may affect phytoplankton, that are not included in the WFD system A or B, may make drawing relationships between environmental variables and reference phytoplankton communities difficult. Examples were listed in Table 4.1 and include zooplankton grazing and a highly variable seasonal composition typical of the phytoplankton.

The purpose of a typology is to separate lakes into types based on their natural characteristics and to allow ecological change to be detected more easily (CEC, 2000; REFCOND, 2003). As a metric may be used to detect and express ecological change, it follows that a typology should help partition metric variability between types. However, Table 4.9 shows that the phytoplankton index appeared to be similar across the reference types and also had low within-type variation. This may indicate that the phytoplankton index may successfully work across different lake types. This may simplify the use of the phytoplankton index in the management of a river basin district. However, a larger dataset should be examined to see how the phytoplankton index varies with nutrient enrichment and the mixing depth of the water-column.

4.4.2 Ecological assessment - phytoplankton index

The phytoplankton index was successful in describing the response of phytoplankton to pressure (TP) and may therefore prove a useful tool for the ecological assessment of lakes. The assessment system was developed so that it meets the requirements of the Water Framework Directive (WFD) in that it incorporates information on the composition, abundance and biomass of the phytoplankton. Chlorophyll *a* was used as an overall measure of biomass: the Swedish EPA has estimated that chlorophyll *a* concentration corresponds to 0.5% of plankton volume (SEPA, 2000). The WFD also defines ecological quality classes based on the frequency and intensity of planktonic blooms. Work on this was outside the scope of the project although the phytoplankton

index may be useful in this respect in that it includes examples of the bloom forming taxa: *Aphanizomenon* spp. and *Anabaena* spp.

The index was developed to work on summer samples of phytoplankton (June, July, August and early September). Summer was chosen as it is the period when excessive growth of phytoplankton is most often manifested. However, the WFD requires a minimum sampling frequency of 6 months for monitoring of phytoplankton, which may require further work to be done using spring phytoplankton. The response of phytoplankton, particularly diatoms, to nutrient enrichment in spring may be complicated by the availability of silica and light. The WFD allows sampling frequencies to be modified if justification is provided. Further work on metrics based on spring phytoplankton is recommended and consideration should be given to focusing sampling on at least two occasions in summer to give greater confidence in assessment.

The phytoplankton index was also developed over a limited trophic gradient (TP < 100 μg l^{-1}) and would benefit from the addition of more eutrophic lakes. One eutrophic taxon that is clearly missing is *Planktothrix agardhii* (*Oscillatoria agardhii*), which can dominate in shallow eutrophic lakes (Reynolds *et al*., 2002). The phytoplankton index, and the WFD by omission, fails to account for the influence of predation by zooplankton or other filter feeders, which may reduce overall phytoplankton abundance or selectively alter community structure. For example, the phytoplankton index awards scores for Cyanophytes and Cryptomonads among other groups (Table 4.10), but zooplankton may selectively filter the smaller Cryptomonads thereby reducing their abundance and giving a higher score.

It may also be noted that the phytoplankton index, like many metrics, may be slightly method dependent. The phytoplankton index is based on the average of scores of up to 9 taxa and chlorophyll *a*. As the scores are only awarded when a taxon is present, there is a potential for a higher score to be awarded when a higher volume of sample is counted. For example, the higher than expected phytoplankton index recorded in Lough Ramor (Figure 4.13) may in part be influenced by the fact that counting took place in the 1996 study (Irvine *et al*., 2001)at transect level for common species and at

whole chamber level for rarer taxa. Eutrophic taxa, occurring at low concentration may therefore have been awarded a high score. This study developed the phytoplankton index based on counts from one or more transects only of a settling chamber

One criticism of the phytoplankton index is that it sums taxa at genus level or higher and species indicative of oligotrophic conditions may be included with those indicative of eutrophic conditions. This may not be a major drawback as the system mainly works on abundance and species typical of oligotrophic conditions do not typically occur in high abundance. Another criticism is that the system is applied to cells or colonies rather than biomass. Owing to the variable number and size of cells in a colony within a species (Irvine *et al*., 2001) and between species amalgamated in the index (Table 4.10) there may not be a consistent relationship between biomass and colonies ml^{-1}. This may weaken the relationship of the index with indicators of overall biomass (chlorophyll *a*). However, counting in units of occurrence (cells or colonies) is much less time consuming then producing biomass estimates based on cell measurements.

The phytoplankton index was based on defining univariate relationships of eutrophic taxa - taxa which show strong positive growth response to eutrophication - with TP. Effectively, the phytoplankton index attempts to integrate evidence for eutrophication in terms of abundance, composition of selected taxa and overall biomass of phytoplankton. Such an approach to the ecological assessment of phytoplankton has clear limitations. It tends to ignore broader scale community change in the phytoplankton which may be influenced by many factors as well as phosphorus (Reynolds, 1998). Despite this, a NMS ordination (Figure 4.14) showed that there was a good relationship between changes in phytoplankton composition and abundance along a pressure gradient (TP) and the phytoplankton index. So the index seemed to provide a useful summary of changes in the phytoplankton community along a pressure gradient.

5. Macrophytes

5.1 Introduction

The Water Framework Directive (WFD) requires that the composition and abundance of macrophytes are used as one of the biological quality elements to determine the ecological status of lakes. Macrophytes are an important biological group and many of the communities found in Irish lakes are unique and worthy of conservation in both an Irish and European context (Heuff, 1984). Macrophytes are an integral component of lake ecosystems playing a key role in the functioning of other biological groups. They provide a structured habitat for invertebrates and have been linked to macroinvertebrate diversity (Palmer, 1981) and plankton dynamics (Scheffer, 1999).

5.1.1 The influence of eutrophication on macrophytes

Macrophytes are strongly affected by eutrophication (Lachavanne, 1985). In Ireland, eutrophication has been attributed to nutrient export from agricultural and municipal sources (McGarrigle *et al*., 2002). The response of macrophytes to eutrophication may be simplified into three stages as shown in Figure 5.1 for high alkalinity (> 100 mg l^{-1} $CaCO_3$) marl lakes as an example. At low nutrient concentrations (TP < 10 µg l^{-1}) the lake may be termed oligotrophic (OECD, 1982) and the community composition is dominated by charophytes, which colonise the lake to a deep depth owing to high water transparency. Under mesotrophic conditions the nutrient concentrations are higher (10 - 35 µg l^{-1} TP) leading to increased growth of planktonic algae, which reduces transparency and leads to a shallower depth of colonisation (Figure 5.1). The community composition also changes with the addition of species such as *Potamogeton lucens* and *Elodea canadensis*. In eutrophic lakes the high nutrient concentrations (35 - 100 µg l^{-1} TP) lead to substantially increased planktonic algae populations which severely limit transparency. In addition, epiphytic algae may coat the surface of the macrophytes further limiting light availability (Phillips *et al*., 1978). This results in a severe decrease in macrophyte abundance, with only a few species surviving at shallow depths or, with severe pollution, almost complete loss of submerged macrophytes from the lake (Figure 5.1).

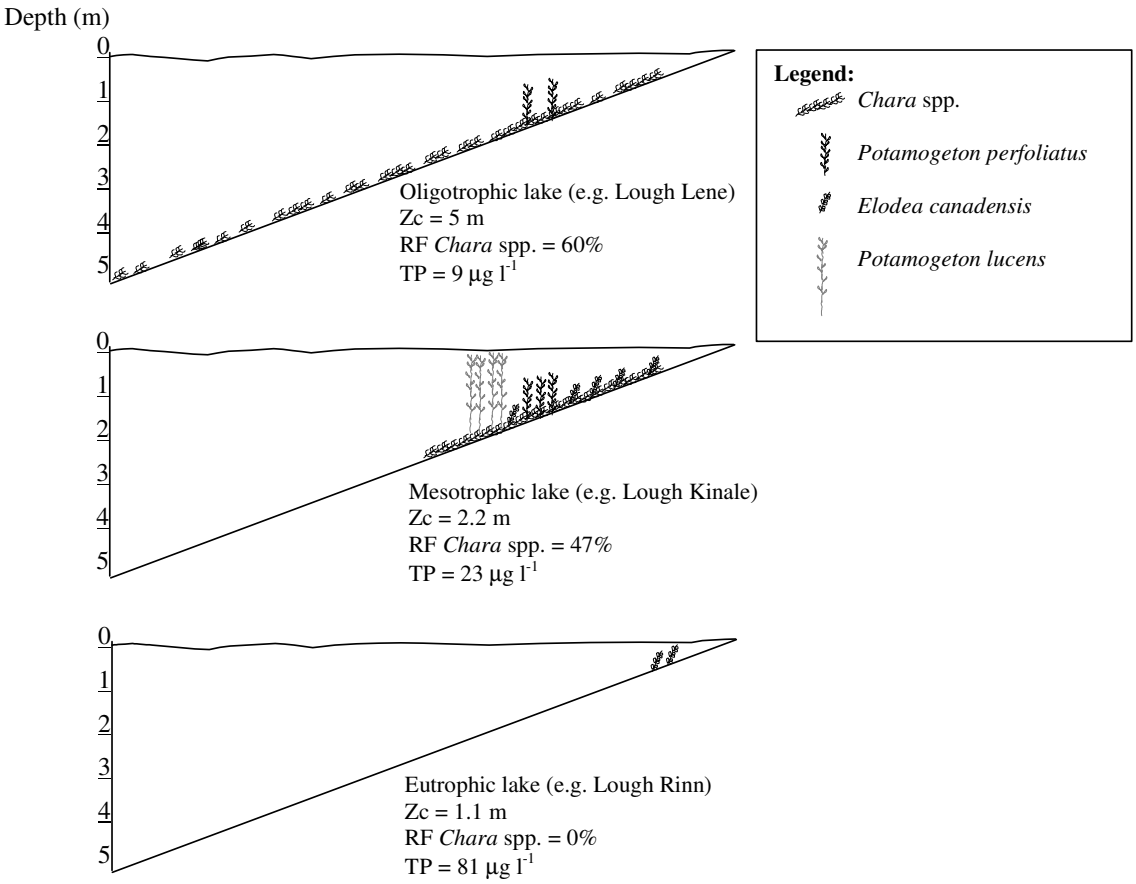


Figure 5.1 Simplified response of macrophyte species composition and depth of colonisation (Zc) to eutrophication in high alkalinity lakes (> 100 mg l^{-1} $CaCO_3$). RF = relative frequency expressed as %occurrence of all transect positions with plants, TP = total phosphorus.

5.1.2 The importance of typology

One of the problems in trying to measure how macrophyte populations change as a result of eutrophication is that there are different types of lakes, which may respond differently to eutrophication. Therefore, natural variation in relatively undisturbed or 'reference' lakes must first be taken into account before the effects of eutrophication can be ascertained. The WFD recognises this and requires member states to separate lakes into types based on their natural characteristics to allow ecological change to be detected more easily (CEC, 2000; REFCOND, 2003).

There are several factors that affect macrophytes under natural conditions that could be used to define lake types. One of these factors is light or transparency which determines the depth of colonisation (Chambers and Prepas, 1988). Substrate type is

also important as most macrophytes derive the majority of their nutrients from lake sediments (Spence, 1982). A larger lake area may lower abundance through increased wind exposure but increase species richness, either as a result of an increase in habitat diversity or by virtue of its broader euphotic zone (Rørslett, 1991; Vestergaard and Sand-Jensen, 2000a). Depth can affect macrophytes through its influence on light penetration, mitigation of the effects of wave exposure at deeper depths and by its relationship with sediment type (Spence, 1982). Alkalinity exerts a strong influence on the natural distribution of macrophytes. Certain plant groups such as elodeids and charophytes increase with alkalinity owing to their ability to use HCO_3^- as a carbon source for photosynthesis, whereas species typical of soft-water lakes such as *Isoetes lacustris* do not have this ability (Vestergaard and Sand-Jensen, 2000b).

In recognition of the factors thought to define lake types naturally, the WFD lists ecoregion, altitude, depth, area and geology (of which alkalinity is a surrogate) as obligatory factors to be used to define lake types (CEC, 2000). One of the objectives of this study and reported in this chapter is to define a typology for Irish reference lakes using macrophytes. The approach taken is to examine macrophytes in 58 candidate reference lakes (data for 11 candidate reference lakes was not available for macrophytes) and to define boundaries to distinct biological types in terms of alkalinity, depth and area. Ecoregion and altitude are excluded from the typology as they mainly form one type, respectively the island of Ireland (ecoregion 17) and less than 200 m.

5.1.3 Assessment of ecological quality using macrophytes

Following the establishment of lake types, the ecological quality of a lake is then assessed by comparing it to a set of minimally impacted (i.e. reference) lakes of the same type. The next question, and the second objective of this chapter, is how to assess the ecological quality of a lake by comparison with its type-specific reference condition.

Metrics and multimetric indices

The use of multimetric indices for ecological assessment is practiced widely in the US and, through production of a European standard, is now one of the recommended approaches for the WFD (CEN, 2004). A metric is a measure of one or more

properties of a biological group. For example, metrics can be based on abundance, composition, tolerance or sensitivity to pollution, functional groups such as feeding guilds, life-form groups, reproductive ability and health (US EPA, 2004). A multimetric index is an index that is the combination of the results of three or more such metrics. The combination of several metrics is done to obtain a more comprehensive assessment of a biological group. In addition, combining metrics should also increase the reliability of an assessment (CEN, 2004). The draft CEN (2004) guidance recommends five steps in the development of a multimetric index.

- Selection of candidate metrics
- Exclusion of redundant metrics
- Selection of core metrics
- Transformation of core metrics into a 0 - 1 score (where 0 = bad and 1 = good ecological quality)
- Averaging of transformed core metrics to give a multimetric index

Multimetric indices can be developed to measure biological response to one or more pressures. Where separate metrics are developed for different pressures then the worst result can be used to assign an ecological quality class (CEN, 2004). Multimetric indices can also be developed specifically for a type. However, a balance needs to be achieved, as too many or too few types can prevent effective metrics being developed (US EPA, 2004). Attempting to develop a separate index for each of many different types may be problematic owing to low sample size and a limited pressure gradient; using too few types may not be appropriate as taxa may be present or absent owing to natural factors rather than anthropogenic pressure. It may be preferable to initially develop a multimetric index across broad types and then seek to refine it using a more detailed typology. This has the benefit of helping the interpretation of a multimetric index across types.

Nichols *et al*. (2000) developed a multimetric index comprised of seven metrics: maximum depth of plant growth (m), proportion of littoral area vegetated (%), submersed species (%), taxa number, exotic species (%), Simpson's diversity index and sensitive species (%). Each of these metrics was scaled from 1 to 10, based on the

distribution of data for the metric along a pressure gradient, and then summed to give a multimetric index. The upper-quartile, inter-quartile and lower-quartile of the multimetric index were taken to correspond to high quality, moderately impacted and severely impacted plant communities. The index was found to detect regional differences in water quality. One drawback was the tendency for some of the metrics to show a non-linear response to pressure. For example, contrary to the expected trend, an increase in nutrients in one lake was found to increase the depth of colonisation and diversity. It was concluded that the metric could not be used uncritically.

Although the compilation of metrics into a multimetric index can be done by a simple averaging (CEN, 2004) or addition of the metrics (Nichols *et al*., 2000) an alternative approach may be to use multiple regression to develop a multimetric index. This has the advantage that only metrics significant in explaining the variation in an anthropogenic pressure gradient (e.g. TP) will be included in the model. The problem of redundant metrics is therefore dealt with statistically. In addition, stepwise multiple regression also indicates the relative importance of different metrics and combines them in a way that increases the predictive power along a pressure gradient. One of the drawbacks of multiple regression is that the variables must fit the assumptions of multiple regression. In addition, a value for all predictive metrics must be used in the regression. In contrast, one of the benefits of the CEN (2004) approach, which averages several metrics, is that it compensates for missing values or allows metrics that are inappropriate for certain types to be excluded. For example, if the depth of colonisation is used along with other metrics in an index, it may be appropriate to exclude it from the calculation of an average multimetric index for shallow lakes. A further benefit of the multimetric approach is that a metric with a response limited to part of the pressure gradient may be used.

5.2 Methods

5.2.1 Sampling

The method used for macrophyte sampling consisted of samples being taken by grapnel along a transect perpendicular to the shoreline and is described in Chapter 3.

5.2.2 Species richness

Sampling was designed to measure ecological quality and while species richness is a part of this, time did not permit an inventory of the species in the lakes. However, it is useful to estimate the proportion of the macrophyte taxa detected by the sampling strategy. Species-area curves may be used to estimate whether the sampling effort recovered an adequate amount of species. Typically, such curves show a steep increase in species collected followed by a plateau where additional effort (transects) only results in a few additional species being collected (McCune *et al*., 2002). Figure 5.2 shows that the typical sampling effort of between three and six transects per lake would, as expected, fall short of providing a correct estimate of species number. One method of estimating the 'true' number of species in a lake from a sample is to calculate a second order jack-knife estimate. This works by using the number of species that occurred once and twice in the sample (McCune *at al*., 2002). This method estimated that there was likely to be as much as double the number of aquatic species as detected by the sampling strategy (Table 5.1).

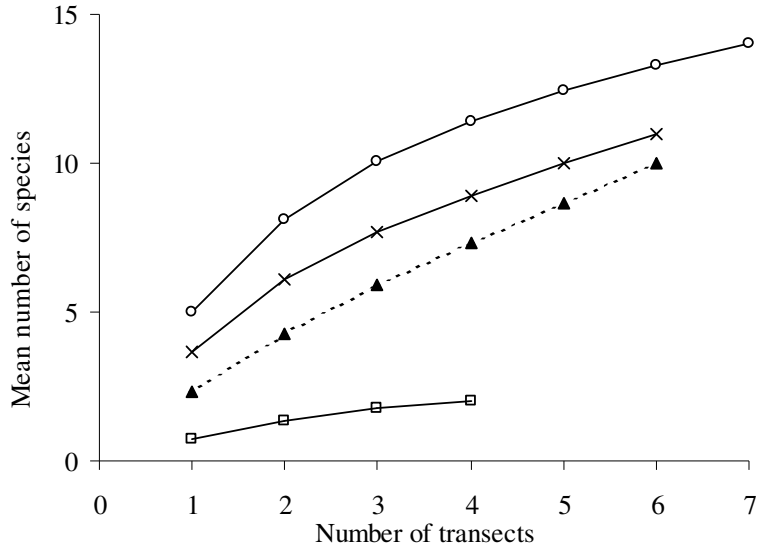


Figure 5.2 Species area curves for Loughs Derravaragh (—○—), Sheelin (—×—), Caragh (- - -▲- - -) and Bunny (—□—).

Table 5.1 Number of taxa recorded and Jack-knife estimate of number of macrophyte taxa likely to be present in four lakes.

Lake	n taxa	2nd order Jack knife
Derravaragh	14	20
Sheelin	11	20
Caragh	10	22
Bunny	2	3

5.2.3 Usefulness of underwater cameras in macrophyte assessment

One of the best methods of conducting a macrophyte survey is by SCUBA diving (Melzer, 1999). This is preferable to a grapnel-based method (as used in this survey) which tends to under-represent rooted taxa with a growth-form close to the sediment (e.g. *Isoetes* spp.). The usefulness of underwater cameras was investigated as a safe and cost efficient alternative to SCUBA diving. Transects were filmed using a pole-mounted PAL Seaview Super Mini-Color 50 camera with a 130 watt halogen light and recorded on a Panasonic NV-GS5B digital video camera. Transects were filmed in three contrasting lakes: Loughs Ballycullinan, Brackley and Sheelin. The following observations were made on the usefulness of underwater cameras in macrophyte assessment:

- It was difficult to identify taxa from film playback. Species lists derived from film were typically only half those generated by transects using grapnels.
- It was difficult to maintain sufficient control of the camera from a boat to allow the percentage cover of taxa to be determined.
- The area visible from the camera was dependent on transparency. As transparency varies with colour and phytoplankton density, camera-use may be restricted to oligotrophic and mesotrophic lakes of low colour.
- It was difficult to determine the depth of colonisation using cameras owing to poor screen visibility, motion inherent in the use of a small boat and the often sparse growth of aquatic plants.
- Underwater cameras would be useful in providing a permanent record of macrophyte communities in lakes with high transparency. This would be best achieved where SCUBA divers operate a camera along transects. The drop camera could also be used as an adjunct to sampling with a rake.

5.2.4 Calculation of metrics

The metrics calculated in order to assess their potential use in a multimetric index are listed in Table 5.2. Six metric types were identified (modified from the draft CEN (2004) guidance on multimetric indices): depth metrics, composition metrics, abundance metrics, diversity metrics, life-form groups and sensitivity/tolerance metrics. Most of the depth-based metrics were based on the depth of colonisation. One novel index in this group is the average taxon depth as a proportion of taxa maximum depth. The maximum depth at which a taxa was found in a lake was expressed as a proportion of the taxa's maximum depth recorded for the survey. These proportions were averaged for all the taxa in each lake. This effectively expresses the maximum depth of occurrence of a taxon in a lake in terms of its 'reference' maximum depth of occurrence. For the composition metrics the ratio of *Litorella* 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).

The life-form groups identified from two previous studies (Jensen, 1979; Moss *et al*, 2003) were examined and additional similar life-forms encountered during this study were also included. Jensen's (1979) system separates the life-forms as Isoetids (taxa with a low 'rosette' growth form such as *Isoetes*, *Chara* and *Litorella*); Elodeids (taxa capable of growing up into the water column, such as *Elodea* and *Potamogeton lucens*); Nymphaeids (rooted taxa with floating leaves such as *Nuphar*) and Lemnids (free-floating taxa such as *Lemna minor*). With a reduced light environment caused by eutrophication, a shift in relative frequency may be expected from rosette forms to upright growth forms to those with floating leaves. The system developed by the ECOFRAME project (Moss *et al*., 2003) followed a similar approach as Jensen (1979).

Five sensitivity/tolerance metrics were calculated (Table 5.2). The relative frequency of tolerant taxa was calculated as the relative frequency of taxa that occurred in lakes with a weighted average TP above 25 $\mu g\ l^{-1}$ (Table 5.13). The mean trophic rank score was also calculated (Palmer *et al*., 1992). A similar metric to that of Palmer *et al*. (1992): the plant trophic score was developed from the 159 lakes sampled by this

project. This was based on weighted average TP concentration and is described in the results section. The combination of metrics into a multimetric index followed a modified approach of Nichols *et al*. (2000) and CEN (2004).

Table 5.2 Metrics examined and estimated response to eutrophication pressure. Responses with increasing pressure: ↓ = decrease, ↑ = increase and ∩ = unimodal. Zc =max. depth of colonisation,TRS=trophic rank score. See Moss *et al*. (2003) for a fuller description of the following abbreviations; ALG=Algae, ISO=Isoetids, Char=Charophyte dominant community, Sphag=Sphagnum, ElPo=Elodeids and pondweeds, CanNym = communities of canopy forming plants and nymphaeids. References 1 = Jensen (1979), 2 = Moss *et al*. (2003), 3 = Palmer *et al*. (1992), 4 = after Chao (1984).

Metric	Response	Metric	Response
DEPTH BASED METRICS		**DIVERSITY**	
Zc Angiosperms	↓	Species Richness (transect)	∩
Zc *Chara* and *Nitella*	↓	Eveness	∩
Zc All taxa	↓	Shannon index (H)	∩
Mean depth of presence	↓	Simpson's diversity	∩
Mean depth of absence	↑	Species Richness (transect & shoreline)	∩
Average taxa depth as proportion of taxa max depth	↓	No single occurences	∩
Mean taxa max depth in lake · taxa max depth recorded	↓	No double occurences	∩
Zc Isoetes (m)	↓	No single+double occurences	∩
COMPOSITION METRICS		Once or twice as % of taxa found[4]	∩
Frequency - all taxa	↑/↓/∩	Once as % of taxa found[4]	∩
Relative frequency - All taxa	↑/↓/∩	Once or twice as % specimens found[4]	∩
Chara % occurrence	↓	**LIFE-FORM METRICS**	
Isoetes % occurrence	↓	$ELODEIDS_{fn\ grp\ RF\%}$[1]	↑
% of taxa found with algae	↑	$NYMPHAEIDS_{fn\ grp\ RF\%}$[1]	↑
Inverse Litorella+ 0.2 / (Litorella + Lobelia+Eriocaulon)	↓	$ISOETIDS_{fn\ grp\ RF\%}$[1]	↓
% of sites with litorella present	↓	$LEMNIDS_{fn\ grp\ RF\%}$[1]	↑
% *Phragmites*	↑	$ALG_{Ecoframe\ RF\%}$[2]	↑
% *Scirpus*	↑	$ISO_{Ecoframe\ RF\%}$[2]	↓
ABUNDANCE METRICS		$Char_{Ecoframe\ RF\%}$[2]	↓
Number specimens found in lake	∩	$Sphag_{Ecoframe\ RF\%}$[2]	↑
Number of sites with plants	∩	$ElPo_{Ecoframe\ RF\%}$[2]	↑
Relative density of specimens	∩	$CanNym_{Ecoframe\ RF\%}$[2]	↑
Plant Frequency %	∩	$Other_{Ecoframe\ RF\%}$[2]	
Specimen Density	∩	$ElPo\&CanNym_{Ecoframe\ RF\%}$[2]	↑
Taxa Density	∩	**SENSITIVITY/TOLLERANCE METRICS**	
Mean Weight (g)	∩	RF Tolerent taxa	↑
		Plant trophic score	↑
		Palmer's TRS (mean)[3]	↑
		Frequency x Palmer's TRS (mean)	↑
		Tall (>1 m) taxa (RF%)	∩

5.2.5 Analysis

The methods used in data analysis are described in Chapter 3. Analysis was performed on submerged and floating species as defined by Palmer *et al*. (1992).

5.3 Results

5.3.1 General description of macrophytes found in reference lakes

A total of 47 submerged and floating (as defined by Palmer *et al*., 1992) macrophyte taxa were identified from the 58 canidate reference lakes. The average number of taxa found in a lake was 9; the maximum was 16 and the minimum 2 (Figure 5.3). *Littorella uniflora* was the most ubiquitous, occurring in 97% of the reference lakes. Twenty-eight percent of the taxa were rarely encountered, occurring in less than 5% of the reference lakes. The occurrence and abundance of many macrophyte taxa was strongly influenced by alkalinity. Figure 5.4 shows the relative frequency of selected macrophytes for low, medium and high alkalinity lakes. Angiosperms and *Isoetes* spp. were the most frequently encountered macrophytes in low alkalinity lakes (< 20 mg l^{-1} $CaCO_3$). At intermediate alkalinity (20-100 mg l^{-1} $CaCO_3$) *Isoetes* spp. were less frequent whereas *Chara* spp. and *Nitella* spp. were more frequent. In high alkalinity 'marl' lakes (> 100 mg l^{-1} $CaCO_3$) *Chara* spp. dominated the reference lakes sampled.

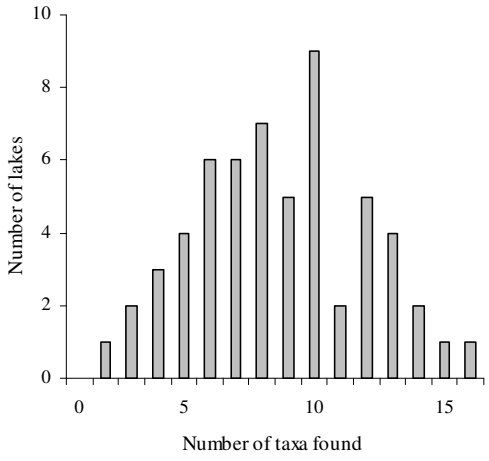


Figure 5.3 Histogram of number of taxa found in reference lakes.

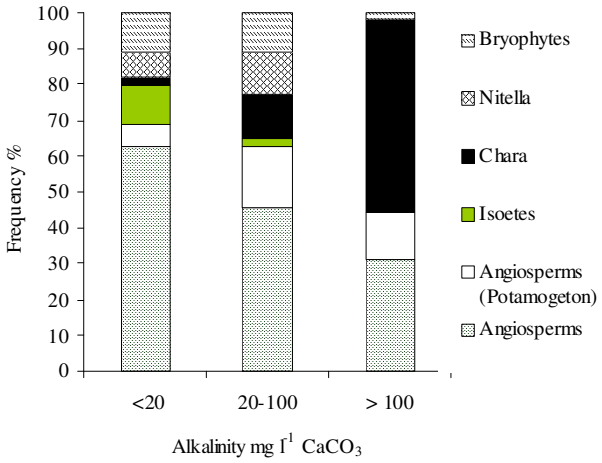


Figure 5.4 Relative frequency (%) of selected macrophyte groups in 58 reference lakes by alkalinity band.

5.3.2 Development of a typology using macrophyte composition and abundance

The approach taken to develop the typology had the following objectives:

1) To determine if distinct types were evident in macrophyte community composition and abundance using 58 reference lakes.
2) To see if such 'biological types' were also distinct in terms of measured environmental variables.
3) To assign environmental boundaries that are useful in defining distinct biological types.
4) To test and describe the resulting lake types in reference condition.

In order to perform an initial search for biological types; cluster analysis was carried out on transformed ($x^{0.5}$) macrophyte abundance (g) (Figure 5.5). An indicator species analysis was used to prune the dendrogram to six clusters. Figure 5.6 shows that six clusters had a high number of significant indicator species (11) and that over all taxa the indicator values were most significant (minimum p) between the six clusters. In order to support the findings of the cluster analysis, a non-metric multidimensional scaling (NMS) ordination was carried out using the Sorensen (Bray-Curtis) distance measure. Figure 5.7 shows that four clusters (same symbols as Figure 5.5 encircled) were reasonably well separated along the most important axes (2 and 3) and two clusters were more distinct along axis 1. Using two multivariate techniques like this provides confidence that the underlying biological variation has been adequately represented by the cluster analysis.

The next step was to see if the six clusters were distinct in terms of macrophytes and measured environmental variables. To do this, an indicator species analysis was carried out on the six clusters to identify taxa that were significant indicators of a cluster in terms of macrophyte abundance and composition (Table 5.3). In order to visualise differences in environmental variables between clusters, box-plots were drawn (Figure 4.4). Pair-wise post-hoc tests (with Bonferroni adjustment) identified where these differences were significant (Table 5.4). Table 5.5 provides a brief description of the clusters with reference to Table 5.3, Figure 5.8 and Table 5.4.

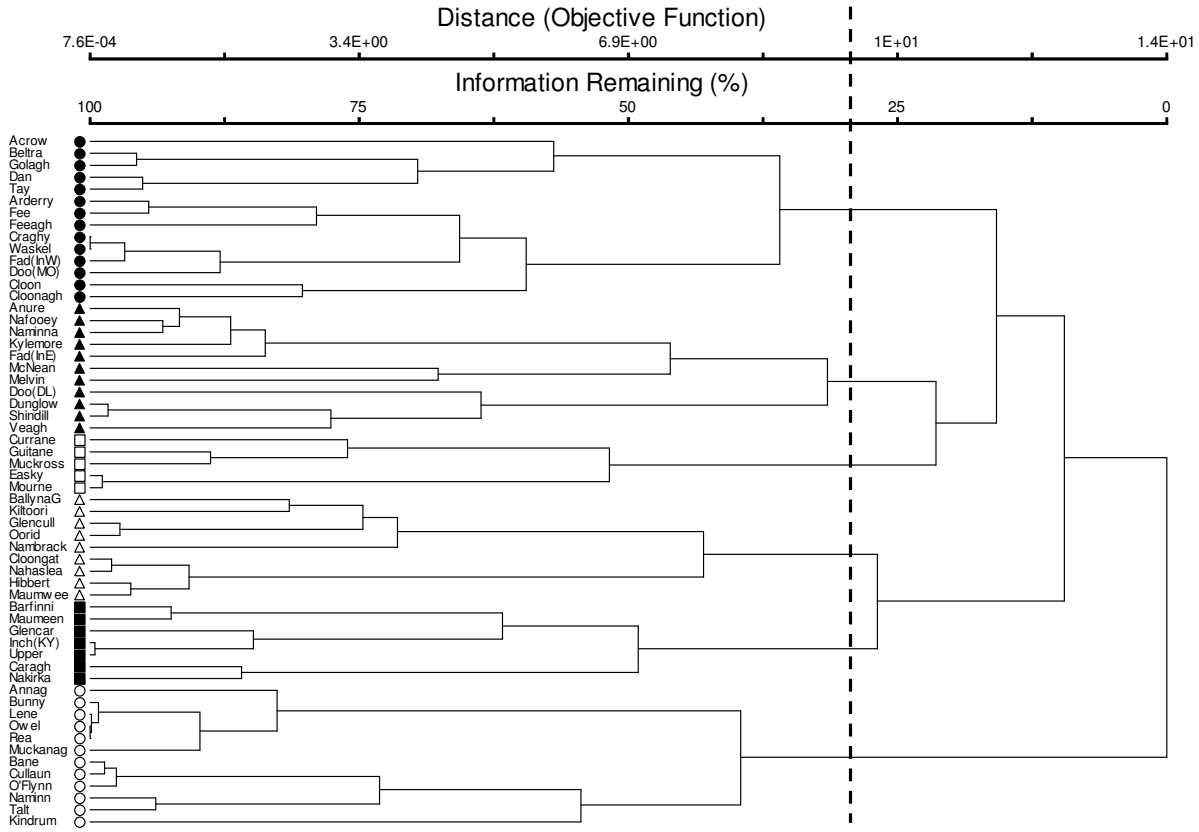


Figure 5.5 Dendrogram from cluster analysis of transformed ($x^{0.5}$) macrophyte abundance in reference lakes (n = 58). Sorensen (Bray-Curtis) distance measure was used with flexible beta (-0.25) linkage. Dashed line represents cut-off point for six clusters.

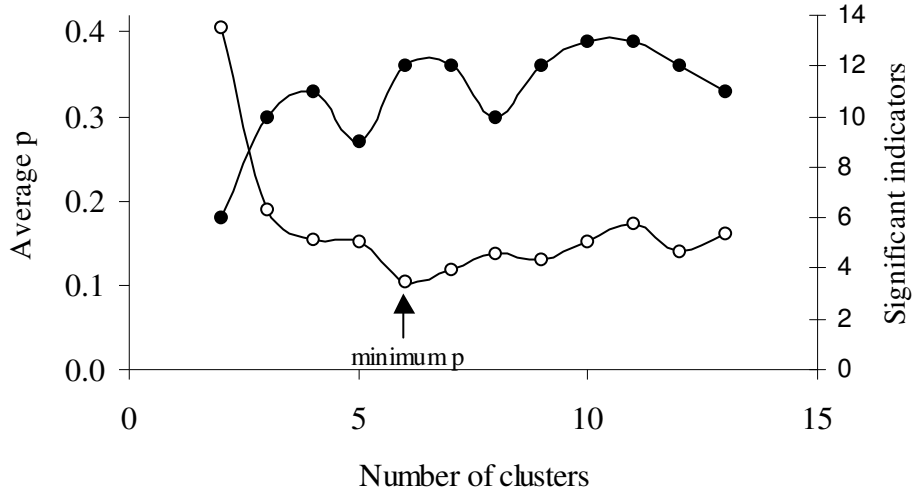


Figure 5.6 Average p for all taxa (○) and number of significant ($p < 0.05$) indicator species identified (●) from an indicator species analysis of clusters 2 to 13. Minimum p (0.10) was reached after six clusters.

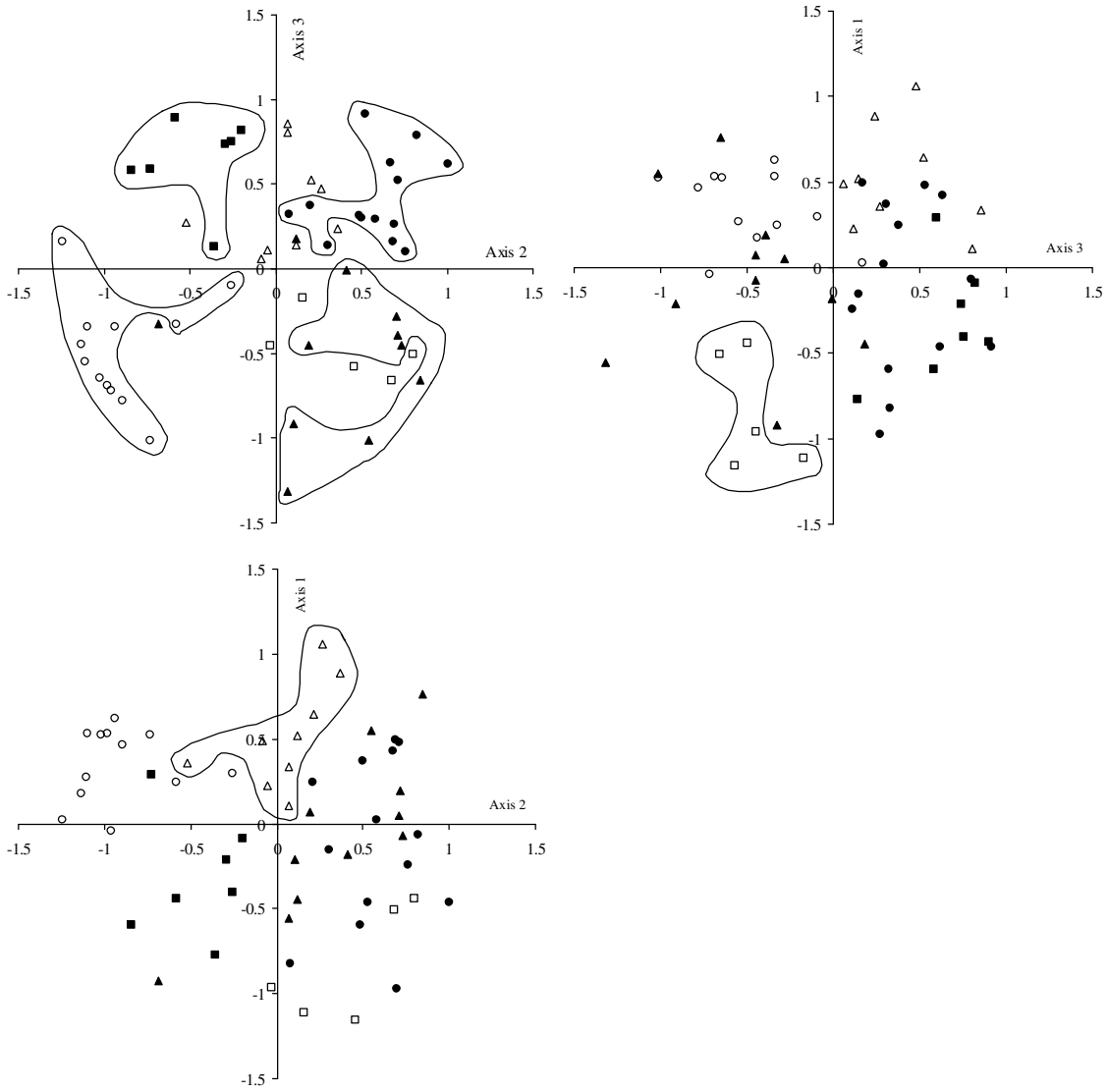


Figure 5.7 NMS ordination of transformed ($x^{0.5}$) macrophyte abundance (g) in reference lakes (n = 58). Sorensen (Bray-Curtis) distance measure used; stress: 17.8. The proportion of variance explained by axis 2 was 35%, axis 3: 22% and axis 1: 13%. Symbols and overlays identify the six clusters found in Figure 5.5.

Table 5.3 Taxa that were found to be significant indicators of clusters. %F = % frequency of occurrence in cluster, %RA = % relative abundance in cluster. p was determined by Monte Carlo test - proportion of 1000 randomised trials where the observed indicator value was equalled or exceeded (McCune *et al*., 2002).

Taxa	Cluster taxa indicative of	Indicator value (%)	%F	%RA	p
Chara spp.	2	70	100	70	0.001
Elodea canadensis	2	33	33	100	0.038
Myriophyllum alterniflorum	3	53	91	58	0.002
Eriocaulon septangulare	4	88	89	99	0.001
Isoetes lacustris	4	69	100	69	0.001
Lobelia dortmanna	4	65	78	84	0.002
Littorella uniflora	4	63	89	71	0.014
Myriophyllum spicatum	4	33	33	97	0.024
Juncus bulbosus	4	42	78	54	0.030
filamentous algae	5	66	71	93	0.001
Nitella spp.	5	80	86	93	0.001
Potamogeton perfoliatus	5	39	43	91	0.032

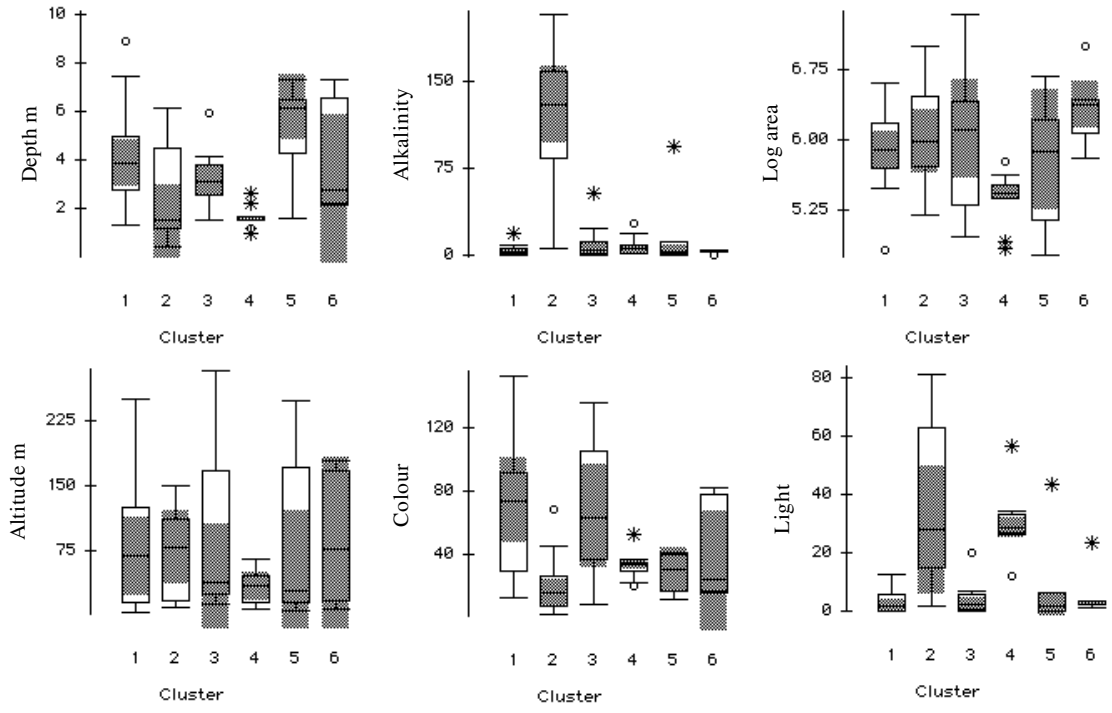


Figure 5.8 Box plots of clusters by mean transect depth (m), alkalinity (mg l^{-1} $CaCO_3$), log area (m^2), altitude (m), colour (mg l^{-1} PtCo) and predicted % light remaining at mean transect depth in reference condition (Free *et al*., 2005). See Figure 5.17 for legend.

Table 5.4 Results (p) of Bonferroni post-hoc tests for significance differences between clusters in transformed (Log x + 2) environmental variables.

Cluster	Mean transect depth m	Alkalinity mg l^{-1} $CaCO_3$	Area m^2	Altitude m	Colour mg l^{-1} PtCo	% light at mean transect depth
2 - 1	0.112	**< 0.001**	1.000	1.000	**< 0.001**	**< 0.001**
3 - 1	0.998	0.906	1.000	1.000	1.000	1.000
3 - 2	0.802	**< 0.001**	1.000	1.000	**0.002**	**< 0.001**
4 - 1	**0.007**	0.901	0.325	0.901	0.625	**< 0.001**
4 - 2	0.988	**< 0.001**	0.101	0.931	0.375	1.000
4 - 3	0.142	1.000	0.115	0.894	0.803	**< 0.001**
5 - 1	0.955	0.993	1.000	1.000	0.312	1.000
5 - 2	**0.011**	**< 0.001**	0.997	1.000	0.891	**< 0.001**
5 - 3	0.469	1.000	0.998	1.000	0.475	1.000
5 - 4	**0.001**	1.000	0.885	1.000	1.000	**< 0.001**
6 - 1	1.000	1.000	0.853	1.000	0.942	0.998
6 - 2	0.567	**< 0.001**	0.994	1.000	0.535	**0.006**
6 - 3	1.000	0.996	0.995	1.000	0.980	1.000
6 - 4	0.106	0.995	**0.031**	0.993	1.000	**0.006**
6 - 5	0.990	1.000	0.725	1.000	1.000	1.000

Table 5.5 Description of clusters using Table 5.3, Table 5.4 and Figure 5.8.

Cluster	Description
Cluster 1 (●)	Low alkalinity (median = 4 mg l^{-1} $CaCO_3$) of medium transect depth ($\bar{x}$ = 4.2 m) with no significant indicator taxa but consistently had *Isoetes lacustris*, *Litorella uniflora* and *Fontanalis antipyretica* in low abundance.
Cluster 2 (○)	Alkalinity was significantly higher than all other clusters (median = 131 mg l^{-1} $CaCO_3$), mean depth was variable, colour was significantly lower than 2 other clusters. *Chara* spp. and *Elodea canadensis* were significant indicator taxa. *Chara* spp. occurred in 100% of this clusters lakes and the majority (70%) of the abundance of *Chara* spp. in the 58 reference lakes was concentrated into this cluster (Table 5.3).
Cluster 3 (▲)	Low alkalinity (median = 6 mg l^{-1} $CaCO_3$) of medium transect depth ($\bar{x}$ = 3.3 m) with *Myriophyllum alterniflorum* as a significant indicator taxa occurring in 91% of lakes in this cluster.
Cluster 4 (△)	Low alkalinity (median = 6 mg l^{-1} $CaCO_3$) with a shallow transect depth ($\bar{x}$ = 1.6 m) that was significantly shallower than 2 other clusters, and had higher estimated light levels (Figure 5.8). Significant indicators were *Eriocaulon septangulare*, *Lobelia dortmanna*, *Isoetes lacustris*, *Litorella uniflora*, *Juncus bulbosus* and *Myriophyllum spicatum*. In addition to these taxa being frequent in this cluster there was also a notable concentration of abundance into this group (Table 5.3 and Table 5.4).
Cluster 5 (■)	Low alkalinity (median = 3 mg l^{-1} $CaCO_3$) with a deep transect depth ($\bar{x}$ = 5.5 m) that was significantly deeper than 2 other clusters. *Nitella* spp., filamentous algae and *Potamogeton perfoliatus* were significant indicator taxa.
Cluster 6 (□)	Low alkalinity (median = 4 mg l^{-1} $CaCO_3$) with a medium transect depth ($\bar{x}$ = 4.2 m) and tended to have a higher lake area (Figure 5.8). No significant indicator taxa were found, both diversity and abundance were low in this cluster.

Table 5.4 shows that no significant differences were found in the selected environmental variables among clusters 1, 3, 5 and 6. These groups could all be classified as having low alkalinity lakes with a medium to deep transect depth. In contrast, clusters 2 and 4 had a number of significant differences with the other clusters in terms of environmental variables as well as indicator species (Table 5.3, Table 5.4). Cluster 4 had a low alkalinity but appears distinct from the other lakes of low alkalinity by having a shallow transect depth, high light level and several Isoetid growth forms as significant indicator species. Cluster 2 had a significantly higher alkalinity and its most important indicator taxa were *Chara* spp.. It is noteworthy that there were few significant differences between the clusters for altitude, colour and lake area (Table 5.4).

Examining the differences in environmental factors between clusters is useful in that factors that exert a strong influence on the biological groups may be readily identified (e.g. alkalinity, Figure 5.8). However, it must also be considered that environmental factors may have compounding effects resulting in a distinct environmental type that is reflected in macrophyte abundance and composition. For example, it may be expected that a combination of low colour, a shallow littoral and a small lake area may lead to a higher abundance of macrophyte taxa by reduced exposure and high light levels (Figure 5.8, Cluster 4). One way to examine clusters in terms of a combination of environmental variables is to perform a discriminant analysis – also known as canonical variates analysis (CVA). CVA determines which linear combination of environmental factors discriminates best between clusters and can indicate if clusters are different in terms of environmental factors (ter Braak and Smilauer, 2002).

Axes 1 and 2 of the CVA represented 80% of the variation in the relationship between clusters (referred to as species in CVA) and the environment (Table 5.6). Figure 5.9 shows the group centroids for each cluster. In support of the univariate examination of environmental variables, the group centroids for clusters 1, 3, 5 and 6 are located close together indicating that they are not distinct environmentally. In contrast, the centroids for clusters 4 and 2 are more distinct and are more closely surrounded by each clusters lakes (encircled). The arrows (Figure 5.9, Figure 5.10) and standardised canonical coefficients (Table 5.7) give an indication of the relative importance of each

environmental factor in cluster separation. Alkalinity, area and mean transect depth were the most important discriminant factors along axis 1, 2 and 3 respectively. In contrast, colour and altitude were not found to be significant ($p > 0.05$) in separating clusters in the model (Table 5.8).

In summary, the analysis of 58 reference lakes indicates that 3 groups were distinct in terms of both macrophytes and environmental characteristics. The first is a high alkalinity group characterised by *Chara sp*. (cluster 2), the second is a low alkalinity shallow type characterised by rich growth of several Isoetid growth forms (cluster 4). The third group are low alkalinity lakes of medium to deep depth comprising clusters 1, 3, 5 and 6, which although they had some biological differences (Table 5.5) were not clearly distinct environmentally. Alkalinity, area and mean transect depth were the most important factors separating clusters and will be used to form type boundaries.

Selection of lake type boundaries

Environmental type boundaries must be defined in order to separate types of lakes that are distinct biologically. Although the preceding analysis was useful in defining some types and the environmental factors that were important in this, some potential types were underrepresented in reference site selection. For example, only four moderate alkalinity lakes (30 – 100 mg l^{-1} $CaCO_3$) were included owing to the unavailability of reference conditions over this alkalinity range. However, in terms of defining boundaries, the reference sites available may allow upper and lower alkalinity boundaries to be set which are relevant to macrophytes. Plant communities of low nutrient status lakes may be characterised by *Chara* species in hard-water lakes and *Isoetes* species in soft-water lakes (Rørslett and Brettum, 1989; John *et al*., 1982). In fact, for the two clusters visible in Figure 5.5 (distance 1.3E+01) these taxa had the strongest indicator values (*Chara sp.* IV = 93%, p = 0.001; *Isoetes lacustris* IV = 71%, p = 0.003). The distribution of such key taxa may be useful in defining type boundaries. Figure 5.11 shows the frequency of occurrence of *Isoetes lacustris* and *Chara* species in relation to alkalinity for 100 lakes with summer total phosphorus less than 20 $\mu g\ l^{-1}$.

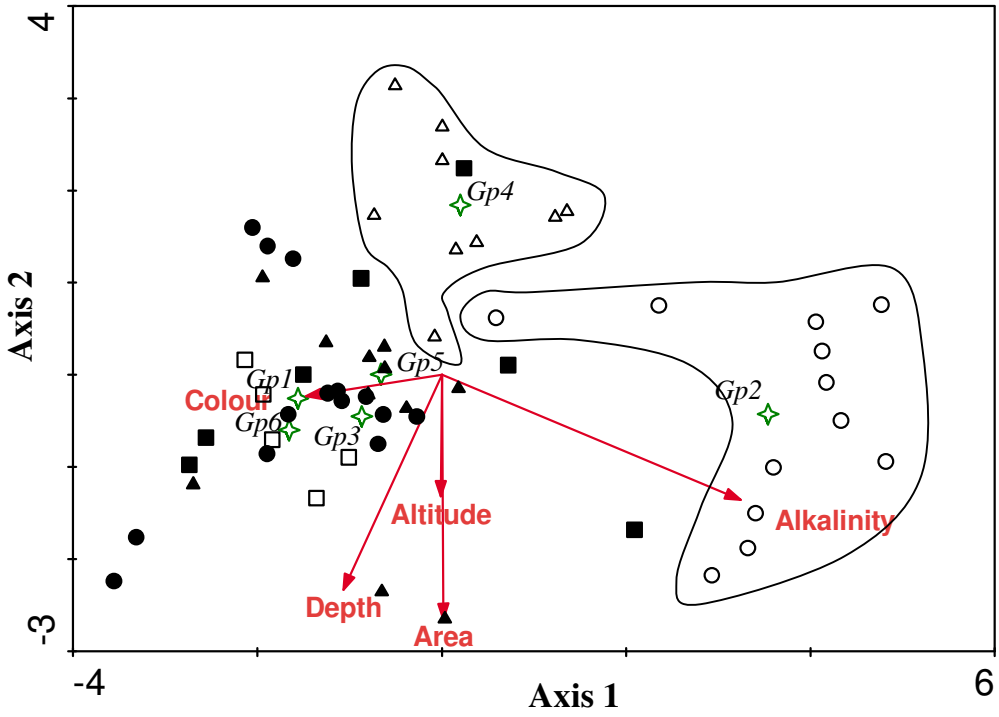


Figure 5.9 Axis 1 and 2 of CVA plot of six clusters identified in Figure 5.5. Group centroids (✧) are labelled. Groups 2 and 4 are encircled.

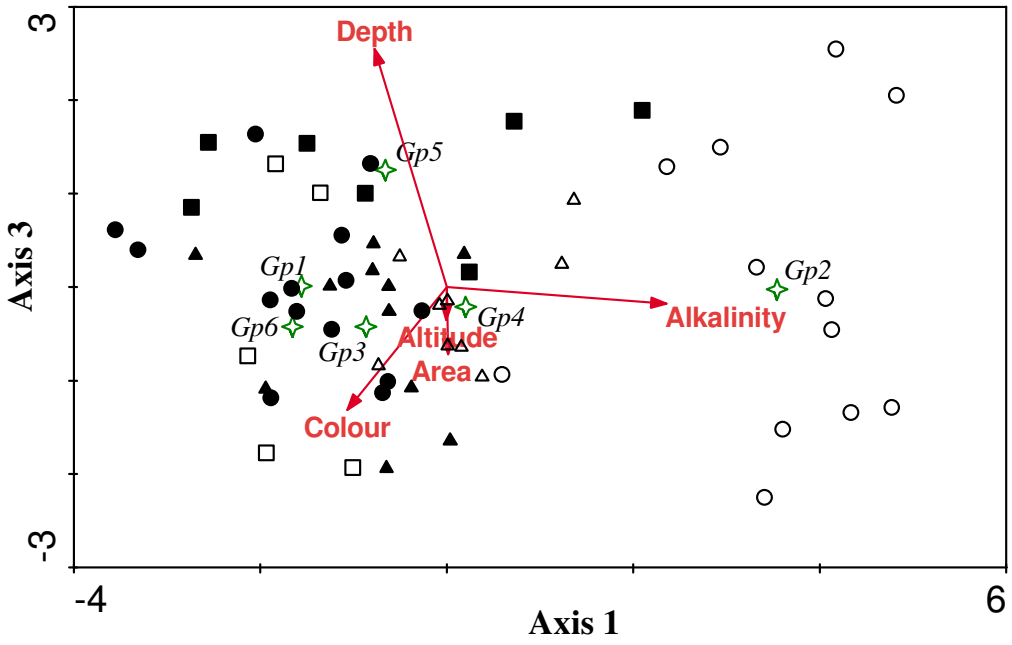


Figure 5.10 Axis 1 and 3 of CVA plot of six clusters identified in Figure 5.5. Group centroids (✧) are labelled.

Table 5.6 Summary statistics of CVA axes.

	Axis 1	Axis 2	Axis 3	Axis 4
Eigenvalues	0.783	0.394	0.197	0.090
Species-environment correlations	0.885	0.627	0.444	0.300
Cumulative percentage variance				
of species data	15.7	23.5	27.5	29.3
of species-environment relation	53.5	80.4	93.8	100

Table 5.7 Standardised canonical coefficients for transformed (Log x + 2) variables.

	Axis 1	Axis 2	Axis 3	Axis 4
Depth	-0.284	-0.619	1.144	-0.392
Alkalinity	1.922	-0.561	0.367	-0.876
Altitude	0.404	-0.632	-0.494	0.051
Area	-0.592	-0.825	-0.744	0.663
Colour	-0.505	-0.439	-0.252	-1.080

Table 5.8 Summary of automatic forward selection in CVA. Marginal effects lists variables in order of variance explained by each variable alone. Conditional effects lists variables in order of inclusion in model along with additional variation explained and whether it was significant ($p \leq 0.05$). Variables were transformed (Log x + 2).

Marginal Effects		Conditional Effects			
Variable	Lambda-1	Variable	Lambda-A	p	F
Alkalinity	0.67	Alkalinity	0.67	0.002	8.61
Transect depth	0.35	Transect depth	0.29	0.006	3.94
Colour	0.34	Area	0.22	0.006	3.19
Area	0.21	Colour	0.15	0.074	2.13
Altitude	0.06	Altitude	0.13	0.084	2.00

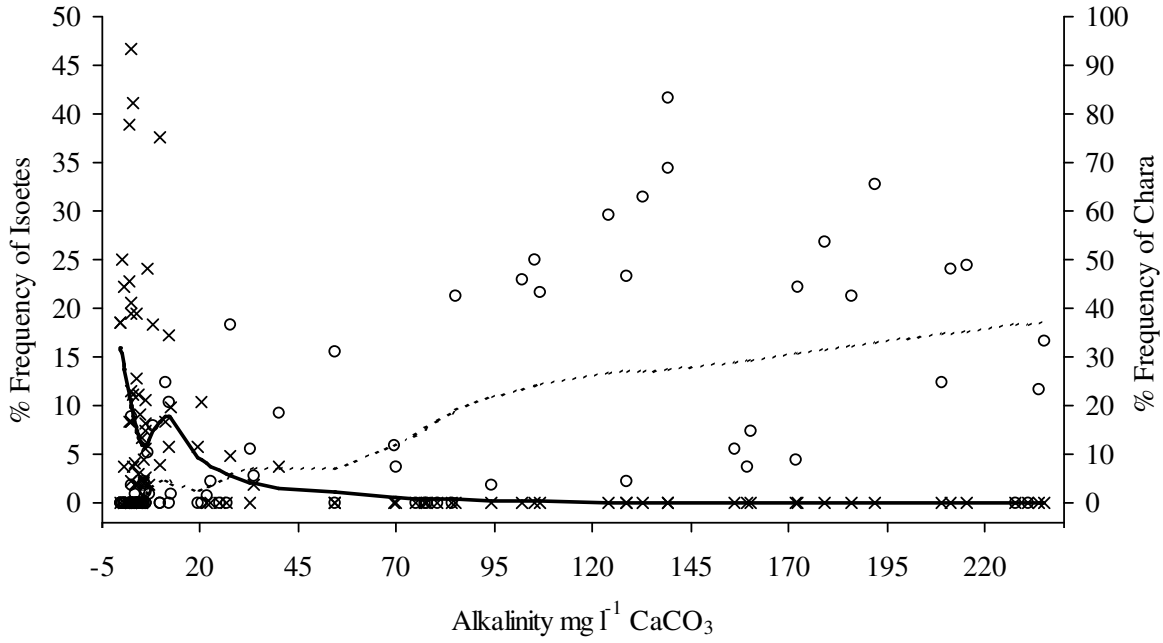


Figure 5.11 Percentage frequency of occurrence with fitted smoothed line for *Isoetes lacustris* (×,——) and *Chara* (○,– –) against alkalinity in lakes with summer TP < 20 µg l^{-1} (n = 100). Lowess smoothed line was fitted (Velleman, 1997).

Isoetes lacustris tends to be largely absent when alkalinity is greater than 20 mg l^{-1} $CaCO_3$ whereas *Chara* species increase markedly between 85 and 100 mg l^{-1} $CaCO_3$. These levels also broadly correspond to the upper 25th percentile alkalinity found in siliceous catchments (26 mg l^{-1} $CaCO_3$) and the lower 25th percentile (108 mg l^{-1} $CaCO_3$) found in catchments with 100% limestone (Free *et al*., 2005). Therefore alkalinities of 20 and 100 mg l^{-1} $CaCO_3$ may be useful for defining biological and environmental types for Irish lakes. This would effectively leave a third type (20-100 mg l^{-1} $CaCO_3$) by default. There may be some biological support for this default type. Lakes in this alkalinity band have previously shown some separation in an NMS ordination of macrophyte abundance (Free *et al*., 2005).

In order to select a type boundary for depth, an average was calculated between the upper and lower 25th percentiles of two shallow clusters (3 and 4) as 2.2 m (Figure 5.8). Cluster 4 was chosen, as it appeared to be strongly influenced by depth in having a distinctly higher amount of littoral rosette species. For the purposes of a reporting typology it was necessary to convert the mean transect depth of 2.2 m into a mean lake depth of 4 m using equation 5.1.

Mean lake depth = 1.526 + 1.168 · mean transect depth **Equation (5.1)**
(r^2 = 0.61, p = 0.0002, n = 17) mean depth data from Irvine *et al*. (2001)

The type boundary for lake area was selected as 50 ha based on the Water Framework Directive System A (CEC, 2000). An additional reason was that cluster 4, whose macrophytes may have been influenced by lake area (Figure 5.9, group centroid 4 at opposite end of area vector) were mostly smaller than 50 ha (upper 25th percentile = 37 ha).

Testing the proposed typology and comparing it with the system A typology

In order to determine if the proposed typology above resulted in types that were biologically distinct, pair-wise multi-response permutation procedure (MRPP) tests were carried out on macrophyte abundance. Table 5.9 shows the A values (chance-corrected within-group agreement) of the pair-wise tests. The A values indicate the homogeneity within a group to that expected by chance: 1 equals complete within group homogeneity whereas an A of 0 equals within group heterogeneity equal to that expected by chance (McCune *et al*., 2002). The main groups previously found to be distinct following CVA and cluster analysis were largely encompassed by the typology and again found to have significant differences in macrophytes (Figure 5.12). For example, cluster 4 is largely (67%) represented in the type "< 20 mg l^{-1} $CaCO_3$ alkalinity, < 4 m mean depth, <50 ha" and was found to be significantly different from all the other low alkalinity lake types (Table 5.9). The high alkalinity (> 100 mg l^{-1} $CaCO_3$) lake types tested were found to be significantly different from all the low alkalinity (< 20 mg l^{-1} $CaCO_3$) lake types. Mid alkalinity lakes (20 - 100 mg l^{-1} $CaCO_3$) were not found to cluster separately in earlier analysis; however, the MRPP analysis did show evidence for significant differences with most of the low and high alkalinity lake types (Table 5.9). Several of the types in the moderate-alkalinity range were underrepresented owing to the unavailability of reference conditions.

The Water Framework Directive requires that typologies developed by member states must achieve at least the same degree of differentiation as would the application of the default system A typology (CEC, 2000). A detailed statistical comparison of both typology systems would be difficult, but only a broad comparison is required

(REFCOND, 2003). MRPP tests may allow some comparison of the proposed typology with that of system A (Table 5.9 and Table 5.10). An indication of the degree of differentiation achieved by each typology system is given by comparing the overall A values from the MRPP analysis. The proposed typology for macrophytes had an overall A value of 0.35 ($p < 0.001$) whereas the default system A typology had an overall A of 0.21 ($p < 0.001$). This provides some evidence that the biologically derived typology was better at partitioning natural variation than the default system A typology. Comparing Table 5.9 with Table 5.10 it can also be seen that the proposed typology was more successful at partitioning variation in reference macrophyte communities in soft water lakes than system A.

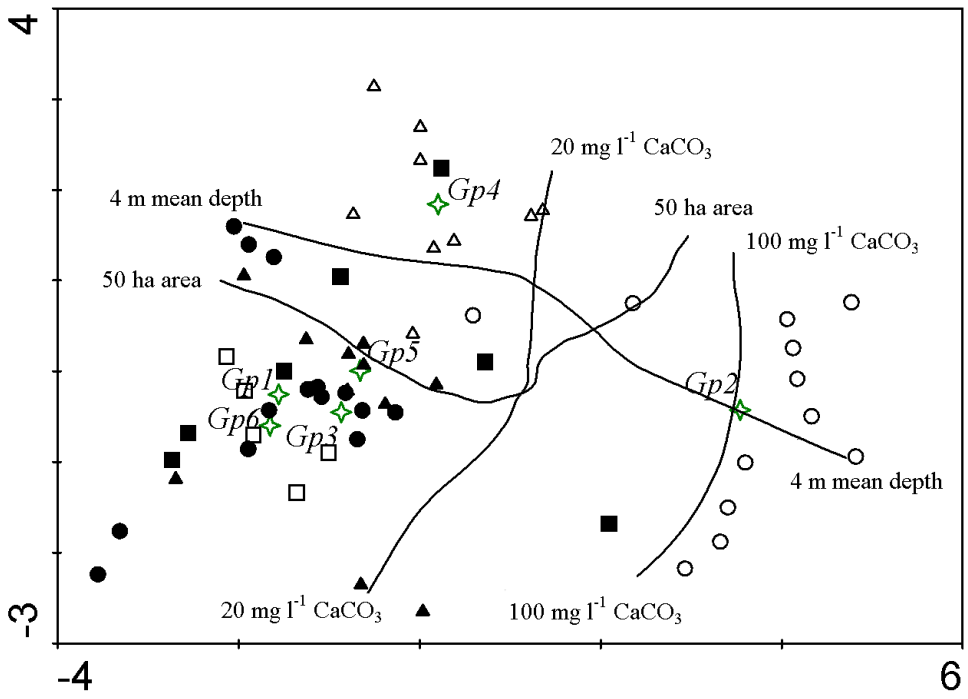


Figure 5.12 Figure 5.9 redrawn with smoothed lines of proposed typology overlain. Axis 1 and 2 of CVA plot of six clusters identified. Group centroids (✧) are labelled.

Table 5.9 Results (A values) of pair-wise MRPP tests for the proposed types using transformed ($x^{0.5}$) macrophyte abundance and the Sorensen (Bray-Curtis) distance measure (rank transformed matrix). Significant ($p \leq 0.05$) differences are in bold. An A of 1 = complete within group homogeneity, an A of 0 = within group heterogeneity equal to that expected by chance (McCune *et al*., 2002). Tests done where group n was > 1 and total n was > 4.

Type	n	< 20 alk < 4 m < 50 ha	< 20 alk < 4 m > 50 ha	< 20 alk > 4 m < 50 ha	< 20 alk > 4 m > 50 ha	20 -100 alk < 4 m < 50 ha	20 -100 alk < 4 m > 50 ha	20 -100 alk > 4 m < 50 ha	20 -100 alk > 4 m > 50 ha	>100 alk < 4 m < 50 ha	>100 alk < 4 m > 50 ha	>100 alk > 4 m < 50 ha	>100 alk > 4 m > 50 ha
< 20 alk < 4 m < 50 ha	8												
< 20 alk < 4 m > 50 ha	4	**0.13**											
< 20 alk > 4 m < 50 ha	9	**0.12**	0.03										
< 20 alk > 4 m > 50 ha	21	**0.11**	-0.01	0.01									
20 - 100 alk < 4 m < 50 ha	1												
20 - 100 alk < 4 m > 50 ha	0												
20 - 100 alk > 4 m < 50 ha	1												
20 - 100 alk > 4 m > 50 ha	5	**0.27**	**0.19**	**0.13**	**0.13**								
>100 alk < 4 m < 50 ha	0												
>100 alk < 4 m > 50 ha	6	**0.42**	**0.35**	**0.45**	**0.36**				**0.26**				
>100 alk > 4 m < 50 ha	1												
>100 alk > 4 m > 50 ha	2	**0.33**	**0.37**	**0.32**	**0.18**				0.10		0.09		

Table 5.10 Results (A values) of pair-wise MRPP tests for system A types using transformed ($x^{0.5}$) macrophyte abundance and the Sorensen (Bray-Curtis) distance measure (rank transformed matrix). Significant ($p \leq 0.05$) differences are in bold. Organic = peat ≥ 50% of catchment, Calcareous = limestone ≥ 20% of catchment.

Type	n	Organic < 3 m < 50 ha < 200 m	Organic < 3 m > 50 ha < 200 m	Organic 3 - 15 m < 50 ha < 200 m	Organic 3 - 15 m < 50 ha > 200 m	Organic 3 - 15 m > 50 ha < 200 m	Organic 3 - 15 m > 100 ha < 200 m	Siliceous 3 - 15 m < 50 ha < 200 m	Calcareous < 3 m > 100 ha < 200 m	Calcareous 3 - 15 m > 50 ha < 200 m	Calcareous 3 - 15 m > 100 ha < 200 m	Calcareous 3 - 15 m > 1000 ha < 200 m
Organic < 3 m < 50 ha < 200 m	8											
Organic < 3 m > 50 ha < 200 m	3	-0.06										
Organic 3 - 15 m < 50 ha < 200 m	4	0.05	0.07									
Organic 3 - 15 m < 50 ha > 200 m	4	0.01	-0.08	0.09								
Organic 3 - 15 m > 50 ha < 200 m	8	0.00	-0.06	0.06	0.01							
Organic 3 - 15 m > 100 ha < 200 m	12	0.05	-0.08	0.08	-0.02	0.03						
Siliceous 3 - 15 m < 50 ha < 200 m	3	0.02	-0.05	0.16	**0.15**	0.10	**0.11**					
Calcareous < 3 m > 100 ha < 200 m	2	**0.27**	0.09	**0.38**	**0.35**	**0.25**	**0.25**	**0.39**				
Calcareous 3 - 15 m > 50 ha < 200 m	2	**0.27**	0.16	**0.36**	**0.32**	**0.27**	**0.24**	**0.39**				
Calcareous 3 - 15 m > 100 ha < 200 m	3	**0.20**	0.08	**0.32**	0.16	**0.15**	**0.19**	0.24	-0.07	-0.09		
Calcareous 3 - 15 m > 1000 ha < 200 m	2	0.05	-0.12	0.11	-0.01	0.02	0.03	**0.09**			-0.19	

Describing the typology in reference condition

A description of the proposed typology for macrophytes was achieved by calculating indicator values of the macrophytes across the types (Table 5.11). A high indicator value (IV > 50%) typically indicates that a taxon is both frequent and abundant in a type. As expected, there is a clear distinction between low and high alkalinity bands in terms of species composition as seen by the higher IV for *Chara* at high alkalinity. In the low alkalinity, small shallow lake type, littoral rosette species are both frequent and abundant but as area and depth increase across the low alkalinity band their IV decreases. It is difficult to describe the moderate alkalinity (20-100 mg l^{-1} $CaCO_3$) type owing to the poor representation across depth and area bands. The type may be distinguished by its lack of key low alkalinity species such as *Lobelia dortmanna* and *Isoetes lacustris* while taxa such as *Nitella* and *Myriophyllum alterniflorum* may still be present.

Mean and standard deviation values for six metrics and the final multimetric index are also presented in (Table 5.11). In high alkalinity lakes in reference condition *Chara* tends to have a relative frequency of occurrence greater than 40%. The plant trophic score appears consistent within alkalinity bands but was higher in the high alkalinity types. The percent of tolerant taxa was highest in the moderate alkalinity type. This may indicate that some of these lakes deviated from reference condition: some difficulty was encountered in finding reference lakes for this type. The final macrophyte index (see next section) appeared to be one of the most similar across the reference types and also had low within type variation. Depth of colonisation (Zc) was highest in the deeper lakes (> 4 m mean depth) indicating that it may only be valid to apply this metric in deeper lakes.

Table 5.11 Description of typology. Indicator values (IV) and metric scores for the 12 types. Indicator values are presented for the most common taxa in the reference lakes (occurring in ≥ 40% of the low or high alkalinity lakes). Mean and standard deviation (in parenthesises) are presented for the metrics.

IV and metric scores	< 20 alk < 4 m < 50 ha	< 20 alk < 4 m > 50 ha	< 20 alk > 4 m < 50 ha	< 20 alk > 4 m > 50 ha	20 -100 alk < 4 m < 50 ha	20 - 100 alk < 4 m > 50 ha	20 -100 alk > 4 m < 50 ha	20 -100 alk > 4 m > 50 ha	> 100 alk < 4 m < 50 ha	> 100 alk < 4 m > 50 ha	> 100 alk > 4 m < 50 ha	> 100 alk > 4 m > 50 ha
n	8	4	9	21	1	0	1	5	0	6	1	2
Isoetes lacustris	69	5	10	6				0		0		0
Lobelia dortmanna	73	1	0	0				0		0		0
Juncus bulbosus	26	12	8	14				0		0		0
Eriocaulon septangulare	62	0	0	0				0		0		0
Potamogeton natans	26	5	13	2				1		0		0
Litorella uniflora	56	8	2	9				1		0		0
Nitella sp.	1	31	15	2				5		0		0
Myriophyllum alterniflorum	1	19	8	2				24		0		0
Fontinalis antipyretica	18	10	2	2				0		0		0
Filamentous algae	0	0	22	5				6		0		5
Potamogeton perfoliatus	0	22	0	0				3		0		6
Chara sp.	2	2	0	0				7		26		47
Macrophyte index	0.86(0.11)	0.7(0.06)	0.75(0.11)	0.77(0.1)				0.67(0.11)		0.78(0.11)		0.89(0.04)
RF% Tolerent taxa	25(12)	35(8)	42(17)	42(15)				60(23)		35(15)		28(2)
RF% Chara sp.	4(5)	3(4)	0(0)	2(4)				14(22)		48(18)		62(2)
Plant trophic score	21(3)	22(4)	23(3)	22(3)				30(3)		31(3)		32(1)
RF% Elodeids	25(19)	35(28)	45(23)	34(15)				63(22)		37(20)		34(5)
Average depth of presence	1.5(0.4)	0.9(0.7)	1.8(0.9)	1.3(0.6)				1.6(0.6)		1.4(0.3)		2.3(0.4)
Zc (m)	2.6(0.4)	2.7(0.8)	3.4(1.6)	3.4(1)				5.1(3.5)		2.2(0.9)		7.5(3)

5.3.3 Development of a multimetric index for ecological assessment based on linear metrics

An assessment system was initially developed for lakes with an alkalinity greater than 20 mg l^{-1} $CaCO_3$ alkalinity. Lakes of lower alkalinity were excluded as they are in a separate reporting typology, naturally have a lower diversity, and the sampling strategy revealed only a limited trophic gradient over which to develop an assessment system. The steps taken to produce the multimetric index were:

1) Identified metrics that had a linear (or log linear) response to TP as: maximum depth of colonisation (Zc), relative frequency of Elodeids (functional group), mean depth of macrophyte presence, relative frequency of tolerant taxa, relative frequency of *Chara* (for lakes > 100 mg l^{-1} $CaCO_3$ alkalinity only)
2) Developed a plant trophic score based on TP concentration.
3) Scaled each of the metrics into deciles ranging from 1 to 0.1 (descending with anthropogenic pressure (TP) as required by the WFD).
4) Averaged the metrics to produce a linear macrophyte index.

The relationships between the selected metrics and transformed TP (Log (x+1)) are shown in Figure 5.13. Correlations with transformed TP (Log (x+1)) ranged from 0.36 for the relative frequency (RF) of Elodeids (functional group) to 0.69 for the RF of tolerant taxa (Table 5.12). Correlations among the metrics were largely below 0.8, the level above which metrics should be excluded owing to redundancy (CEN, 2004). One exception was the RF of *Chara* which had a -0.87 correlation with the RF of tolerant taxa. However, the RF of *Chara* metric was retained as it is only applied to a high alkalinity (> 100 mg l^{-1} $CaCO_3$) subset of the lakes and it was felt important to include *Chara* owing to its conservation importance (John *et al*., 1982). It is also noteworthy that the RF of *Chara* showed a rapid decline with relatively low increases in TP across the lakes (approximately from 10 to 20 µg l^{-1} TP, Figure 5.13).

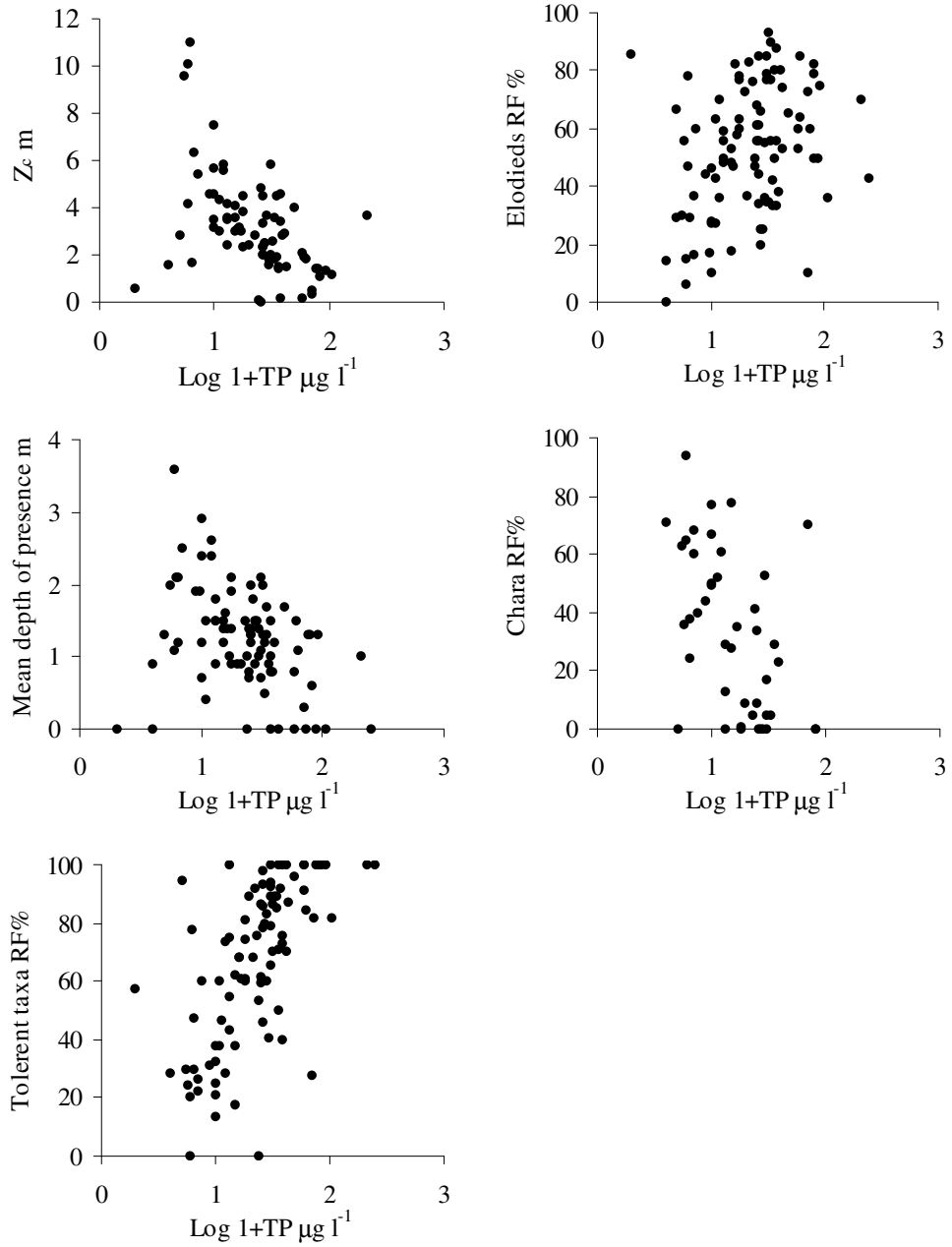


Figure 5.13 Linear relationships found between macrophyte metrics and transformed TP (Log (x+1)) in lakes with alkalinity > 20 mg l^{-1} $CaCO_3$. Chara RF% was only examined for lakes > 100 mg l^{-1} $CaCO_3$ (n = 44).

Table 5.12 Pearson correlation coefficients for the macrophyte index, its components and transformed TP (Log (x+1)). Loughs Cloonagh and Salt were removed (n = 93).

	TP (Log (x+1))	Mean depth of presence	Zc	RF% Elodeids	RF% Chara	Plant score	Macrophyte index	RF% Tolerant
Log TP+1	1.00							
Mean depth of presence	-0.51	1.00						
Zc	-0.53	0.76	1.00					
RF% Elodeids	0.36	-0.19	-0.20	1.00				
RF% Chara	-0.60	0.33	0.39	-0.70	1.00			
Plant trophic score	0.68	-0.34	-0.31	0.14	-0.46	1.00		
Macrophyte index	-0.77	0.61	0.62	-0.61	0.81	-0.64	1.00	
RF% Tolerant taxa	0.69	-0.31	-0.37	0.59	-0.87	0.62	-0.87	1.00

The plant trophic score was developed using data from all lakes sampled (n = 159). Firstly, the lakes were divided into TP bands (<10, 10-20, 20-30, 30-40, 40-60, > 60). For each taxon the mean TP i.e. the average TP of all the lakes combined where the taxon in question was recorded was calculated within these bands. A weighted average TP concentration for each taxon was then calculated based on the percentage of sites where the taxon was present in each TP band. This allowed the score to take into account the different sampling intensities across the trophic scale (i.e. TP bands). Weighted averages were only calculated where a species was found in five or more lakes. For each lake the weighted average scores in Table 5.13 were averaged to give a plant trophic score. The relationship between the plant trophic score and transformed spring TP (Log (x+1)) is shown in Figure 5.14 for lakes with an alkalinity greater than 20 mg l^{-1} $CaCO_3$ and for all lakes. High values of the plant trophic score (above 50) were caused by the co-occurrence of *Lemna minor* and *Lemna polyrrhiza* which tended to occur in lakes with high TP (Table 5.13).

Using the full set of lakes the plant trophic score was slightly better correlated (r_S = 0.70) to transformed TP (Log (x+1)) than Palmer *et al.*'s (1992) mean trophic rank score (r_S = 0.61). However, for lakes with alkalinity > 20 mg l^{-1} $CaCO_3$, the plant trophic score was much better correlated (r_S = 0.61) with transformed TP (Log (x+1)) than Palmer *et al.*'s (1992) mean trophic rank score (r_S = 0.25) (Figure 5.14 and Figure 5.15).

In order to place the metrics on a comparable scale, each of the metrics was scaled (and inverted if necessary) into deciles ranging from 1 (perceived high status) to 0.1 (perceived poor or bad status) (Table 5.14). The metrics were then averaged to give a macrophyte index. A score was not assigned for Zc when it was less than 3 m and between 80 and 100% of the maximum transect depth recorded. Similarly, a score was not assigned for the average depth of presence if it was less than 1.8 m and was within 50% of the maximum transect depth. This was done to prevent a low score being assigned to shallow lakes. The score RF% Chara was developed only for lakes with an alkalinity greater than 100 mg l^{-1} $CaCO_3$. In addition, lakes were randomly removed from this alkalinity band so that the distribution of TP was similar to that of the 95 lakes with alkalinity > 20 mg l^{-1} $CaCO_3$. Otherwise the scaled values would not have referred to a similar pressure gradient (TP).

The relationship between the macrophyte index and TP is shown in Figure 5.16 and Figure 5.17 for lakes with alkalinity > 20 mg l^{-1} $CaCO_3$. Reference lakes clearly have higher values of the index. A linear regression between the macrophyte index and transformed TP (Log (x+1)) resulted in an r^2 of 0.59 ($p = 0.0001$, $n = 93$, Salt and Cloonagh removed). Independent data should be used to test the index when it becomes available.

In order to see if the macrophyte index also corresponded to a pressure gradient detected from an ordination, an NMS ordination of transformed ($x^{0.25}$) abundance was carried out. The ordination was rotated to maximise correlation between transformed TP (Log (x+1)) and axis 1 ($r = 0.52$, $p < 0.01$). Figure 5.18 shows the relationship between the macrophyte index and the pressure gradient apparent from the ordination. The macrophyte index had an r^2 with the rotated axis of 0.50 ($p \leq 0.0001$, $n = 87$, some lakes excluded owing to lack of abundance data).

A multimetric index to detect nutrient enrichment in low alkalinity lakes was not attempted owing to the low numbers of lakes sampled that had high nutrient concentrations (only 3 lakes > 20 μg l^{-1} TP). A temporary measure would be to use the metric developed for the lakes with higher alkalinity (> 20 mg l^{-1} $CaCO_3$) as this metric correctly identified these lakes as being of low TP (Figure 5.19). It is possible that the index may work better when applied separately to types. In such cases the index may be used in terms of the ratio of expected (reference condition) to observed values.

Table 5.13 Weighted spring TP (µg l^{-1}) where taxa present. The score was calculated using data from all lakes sampled for macrophytes (n = 159). Tolerant taxa are those with a TP > 25 µg l^{-1}.

Taxa	Weighted TP of lakes where taxa present	n
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 spp.	17	8
Myriophyllum alterniflorum	17	46
Hippuris vulgaris	20	12
Nitella spp.	20	62
Nymphea alba	21	19
Utricularia vulgaris	21	25
Sagittaria sp	22	13
Chara spp.	23	70
Other Moss *spp.*	23	31
Potamogeton gramineus	23	16
Fontinalis antipyretica	26	78
Potamogeton perfoliatus	28	43
Potamogeton pectinatus	31	17
Lemna trisulca	31	35
Myriophyllum spicatum	32	27
Litorella uniflora	34	109
Potamogeton natans	34	51
Callitriche hamulata	34	6
Potamogeton lucens	35	32
Potamogeton berchtoldii	37	34
Filamentous algae	39	96
Sparganium emersum	40	46
Nuphar lutea	43	66
Elodea canadensis	48	62
Potamogeton obtusifolius	54	14
Potamogeton crispus	59	10
Ceratophyllum demersum	62	9
Polygonum amphibium	67	12
Other *Callitriche spp.*	68	13
Lemna minor	88	11
Lemna polyrrhiza	145	5

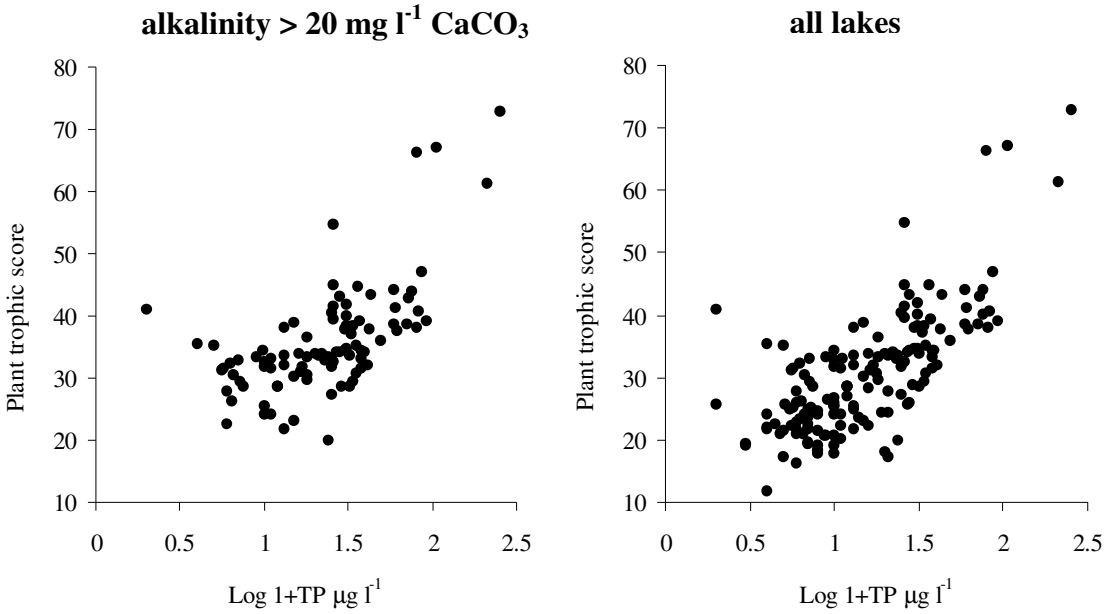


Figure 5.14 Relationship between plant trophic score and transformed TP (Log (x+1)) in lakes with alkalinity > 20 mg l^{-1} $CaCO_3$ (n = 95) and for all lakes (n = 158).

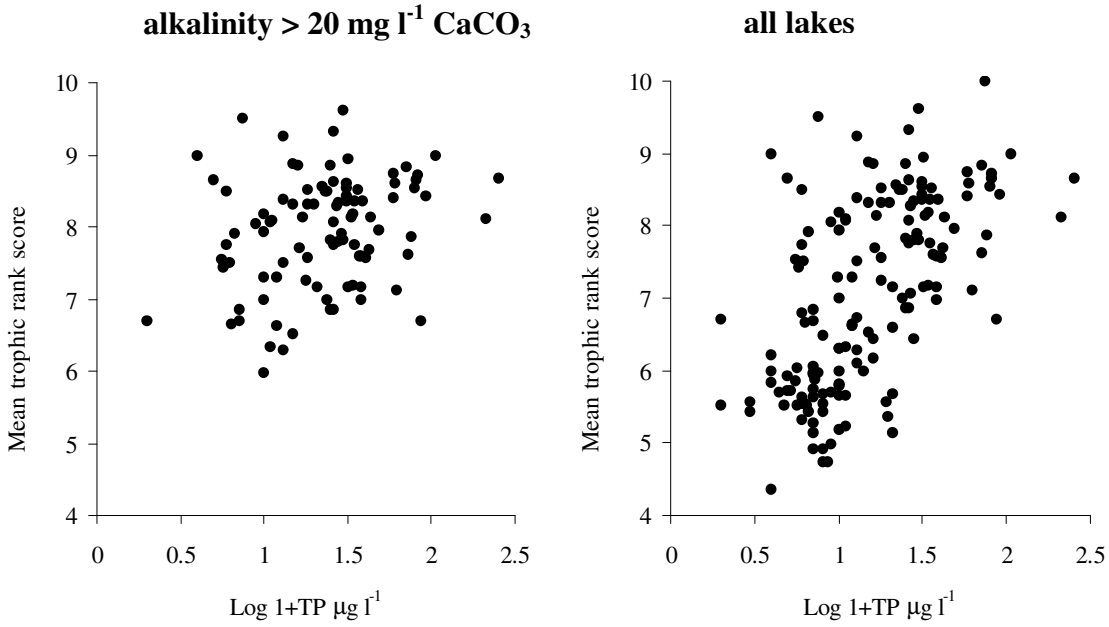


Figure 5.15 Relationship between the mean trophic rank score (Palmer *et al.*, 1992) and transformed TP (log (x+1)) in lakes with alkalinity > 20 mg l^{-1} $CaCO_3$ (n = 95) and for all lakes (n = 158).

Table 5.14 Table of scaled deciles for six metrics that had a log-linear response to spring TP.

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

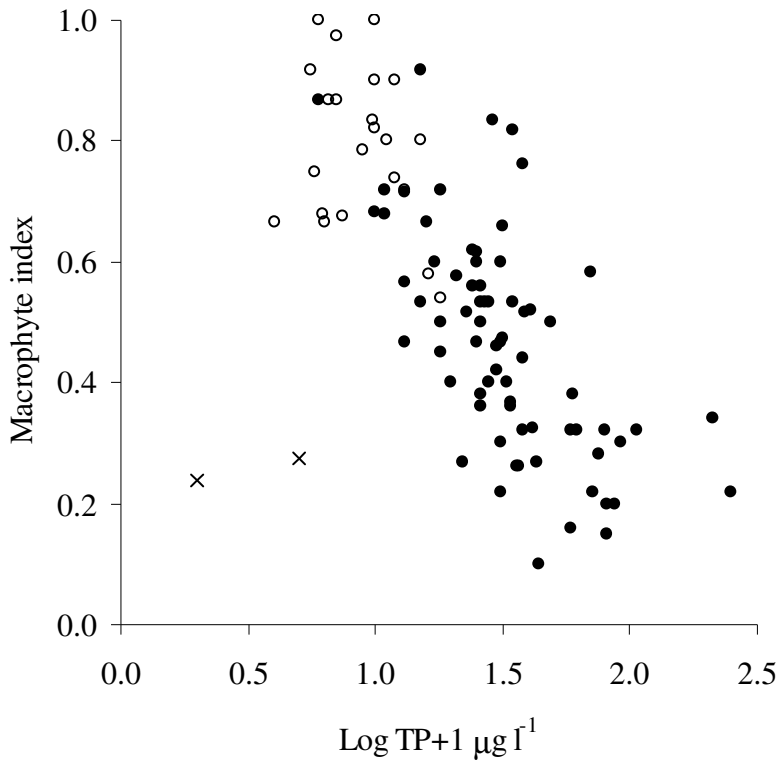


Figure 5.16 Relationship between the macrophyte index and transformed TP (Log (x + 1)) µg l^{-1} in lakes with alkalinity > 20 mg l^{-1} $CaCO_3$. ○ = reference lakes (n = 22) ● = non reference lakes (n = 71). Outliers (×) are Loughs Cloonagh and Salt.

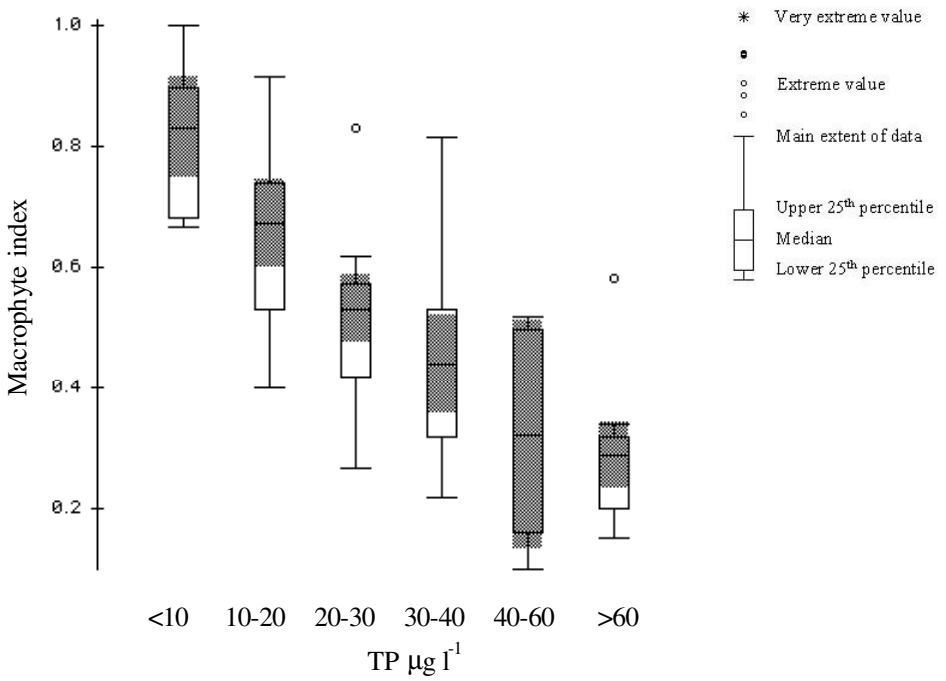


Figure 5.17 Box plot of the macrophyte index and TP µg l^{-1} in lakes with alkalinity > 20 mg l^{-1} $CaCO_3$. Loughs Cloonagh and Salt removed. Shaded areas represent 95% confidence intervals.

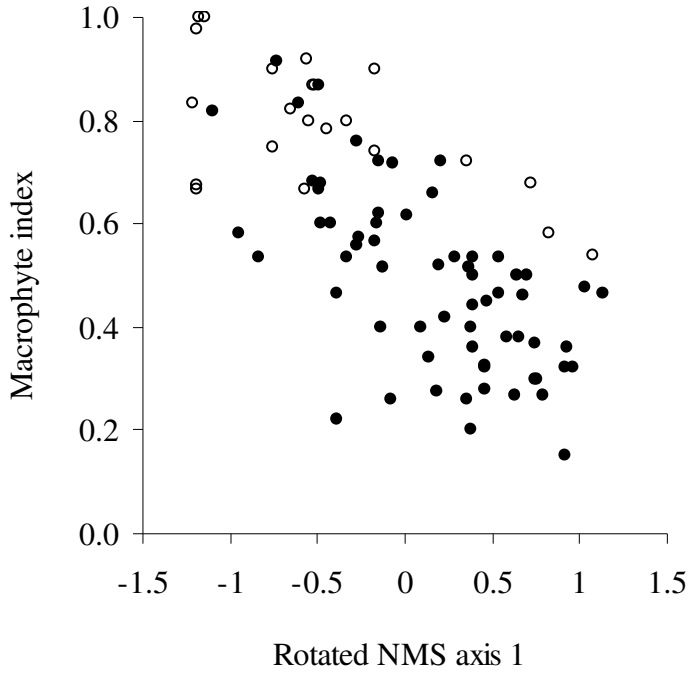


Figure 5.18 Relationship between the macrophyte index and rotated NMS ordination axis 1 (for lakes with alkalinity > 20 mg l^{-1} $CaCO_3$). Ordination was rotated to maximise correlation with transformed TP (Log (x+1)). ○ = reference lakes (n = 22) ● = non-reference lakes (n = 65).

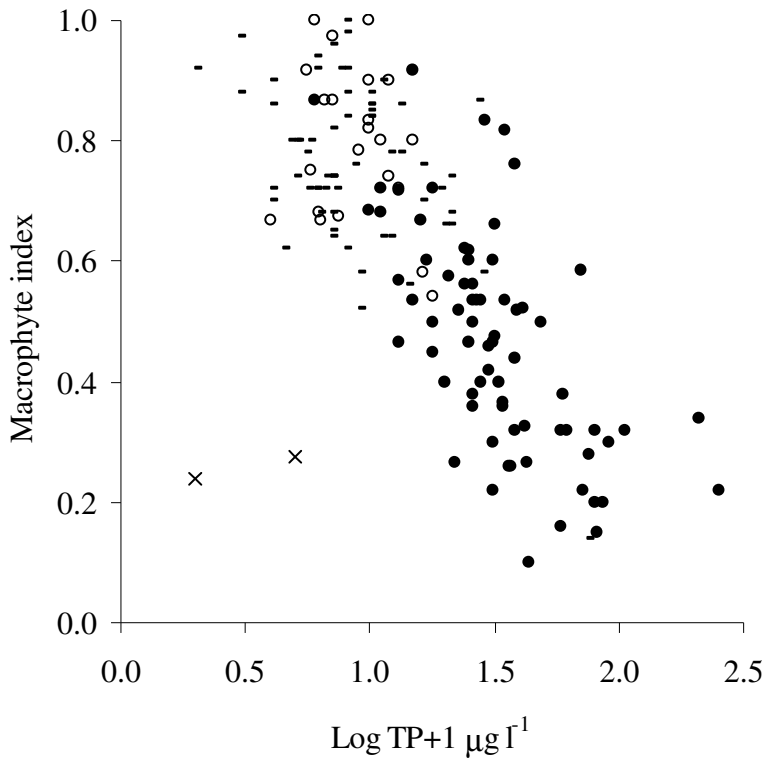


Figure 5.19 Relationship between the macrophyte index and transformed TP (Log (x + 1)) µg l^{-1}. ○ = reference lakes > 20 mg l^{-1} $CaCO_3$ (n = 22), ● = non reference lakes > 20 mg l^{-1} $CaCO_3$ (n = 71) and - = lakes with alkalinity < 20 mg l^{-1} $CaCO_3$ (n = 64). Outliers (×) are Loughs Cloonagh and Salt.

5.3.4 Development of an assessment tool for aquatic macrophytes based on multiple linear regression

The multimetric index from the preceding section was developed by averaging several indexes that had a linear response to anthropogenic pressure (TP). Averaging may not be the most effective way to combine metrics. In addition, it is difficult to determine whether the addition of a metric improves an index or whether it is redundant – simply conveying the same information as metrics already present in the index. In an attempt to address this, stepwise multiple regression models were developed for all the lakes and lake sub-types (based on alkalinity and depth typology bands) (Table 5.15). Figure 5.20 shows the relationship between TP and the model predictions transformed to a 1-0 score to correspond to high and bad status using equation 5.2 modified from CEN (2004).

Equation (5.2)

$$Score = \frac{Metric\ result\ (i.e.\ model\ prediction) - worst\ value\ of\ metric}{Upper\ 10^{th}\ percentile\ of\ metric\ for\ reference\ sites - worst\ value\ of\ metric}$$

The models explained between 57 and 80% of the variation in TP (Table 5.15). Relative metrics such as RF tolerant taxa, RF filamentous algae and taxa found once or twice as a percentage of all taxa found were selected more frequently in the models produced for the high alkalinity lakes (> 100 mg l^{-1} $CaCO_3$). In contrast, the plant trophic score and depth of angiosperm colonisation were more important in the lakes of intermediate alkalinity (20 – 100 mg l^{-1} $CaCO_3$).

Table 5.15 Multiple regression models of transformed (Log x + 1) TP µg l^{-1} against macrophyte metrics for all lakes and lake sub-types.

Typology	Model	n	r^2	p	Outliers
All lakes	-0.558+1.525 · log 1+Plant trophic score-0.673 · log 1+Zc-0.003 · RF Isoetids	153	**0.67**	< 0.001	Cloonagh, Salt
>20 Alkalinity	-0.576+0.0056 · RF tolerant taxa-0.525 · log 1+Zc Angiosperms+1.182 · log 1+Plant trophic score	92	**0.64**	< 0.001	Cloonagh, Salt
20-100 Alkalinity	-0.6114+1.539 · log 1+Plant trophic score-0.651 · log 1+Zc Angiosperms	46	**0.73**	< 0.0001	Egish, Salt
> 4 m	1.111+0.0178 · Plant trophic score-0.609 · log 1+Zc Angiosperms	25	**0.77**	< 0.0001	Egish, Salt
< 4 m	1.886-0.023 · Species richness-0.523 · log 1+Zc Angiosperms	21	**0.64**	< 0.05	
> 100 Alkalinity	1.056+0.007 · RF tolerant taxa-0.0045 · once or twice as % of taxa found	44	**0.64**	< 0.01	Cross, Cloonagh
> 4 m	1.982-0.671 · Mean taxa max depth in lake · taxa max depth recorded in survey	22	**0.57**	< 0.0001	
< 4 m	1.016+0.0056 · RF tolerant taxa-0.0045 · once or twice as % of taxa found+0.0104 · RF filamentous algae	22	**0.80**	< 0.05	Cross, Cloonagh

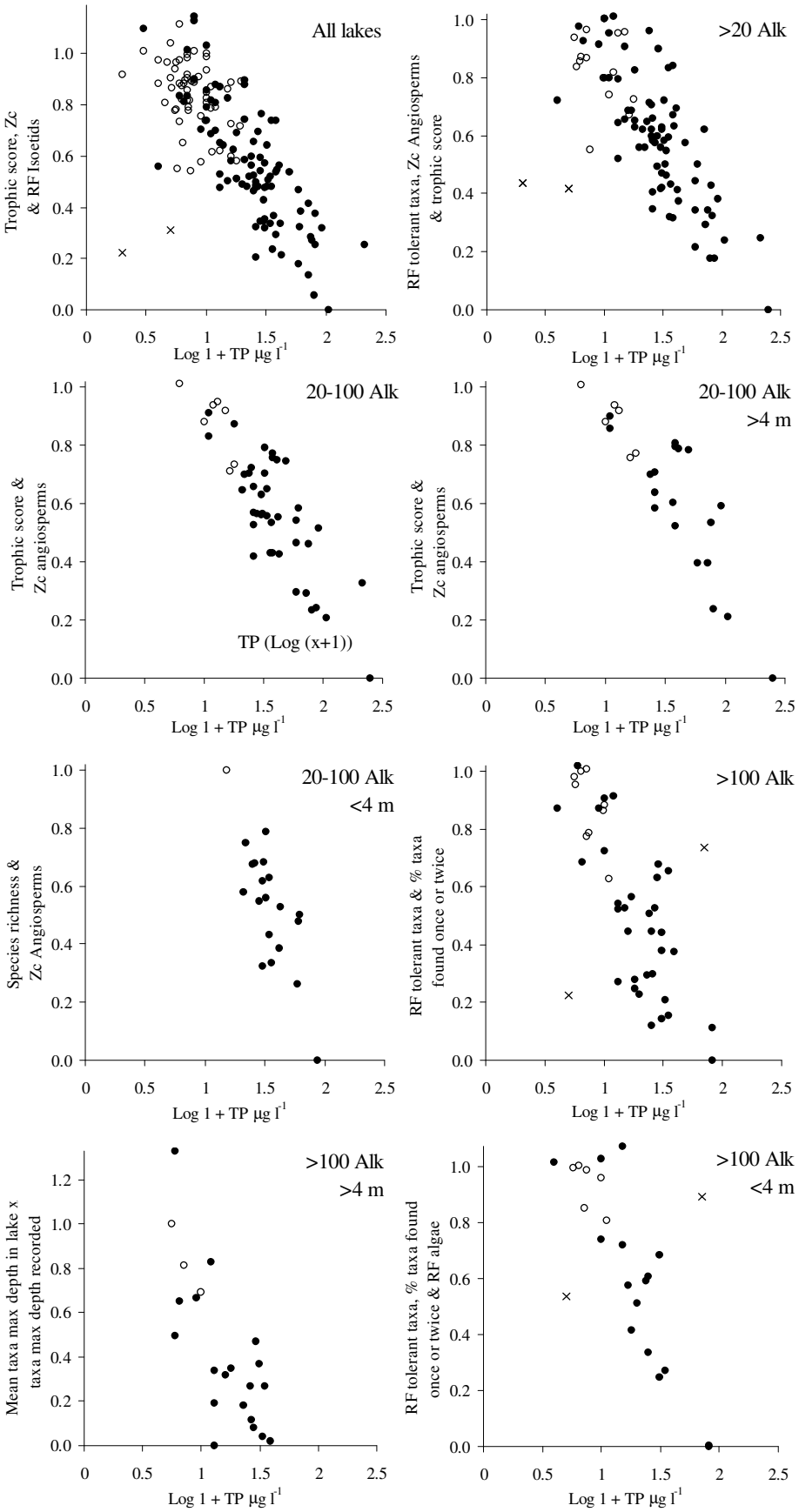


Figure 5.20 Relationship between multiple regression predictions (Table 5.15) and transformed TP (log (x + 1)) for all lakes and lake sub-types. Predictions were converted into a 1-0 score (high to bad status) following CEN (2004). ○ = reference lakes, ● = non-reference lakes, outliers = ×.

5.3.6 Epilithic chlorophyll a

Phytobenthos are required to be monitored by the Water Framework Directive. This project carried out an initial examination of the abundance of epilithic algae, as indicated by chlorophyll *a*, in the littoral zone of 99 lakes. Epilithic chlorophyll *a* was most highly related to alkalinity ($r^2 = 0.43$, $p < 0.001$, Figure 5.21). Total phosphorus was not significant in explaining additional variation in chlorophyll *a* in multiple regression. This indicates that the abundance of epilithic algae may not be related in a straightforward way to mid-lake nutrient concentrations. The higher chlorophyll *a* found at higher alkalinities might be related to the habit of particular species. For example, high chlorophyll *a* was recorded from some marl-encrusted stones with which colonies of *Schizothrix* spp. can be naturally associated (John *et al*., 1982). This strong influence of alkalinity on abundance underlines the need for a typology to be developed for epilithic algae in Irish lakes.

Figure 5.21 compares values recorded in 1997 (Irvine *et al*., 2001) with those recorded from this study (n = 15). Many of the lakes deviate from a 1:1 relationship indicating that there is a high degree of inter-annual variation in abundance or that several sites within a lake should be sampled to better account for natural variation. The high variation in abundance between years and the lack of a relationship between abundance and pressure (TP) indicate the need to focus attention on the use of species composition in ecological assessment.

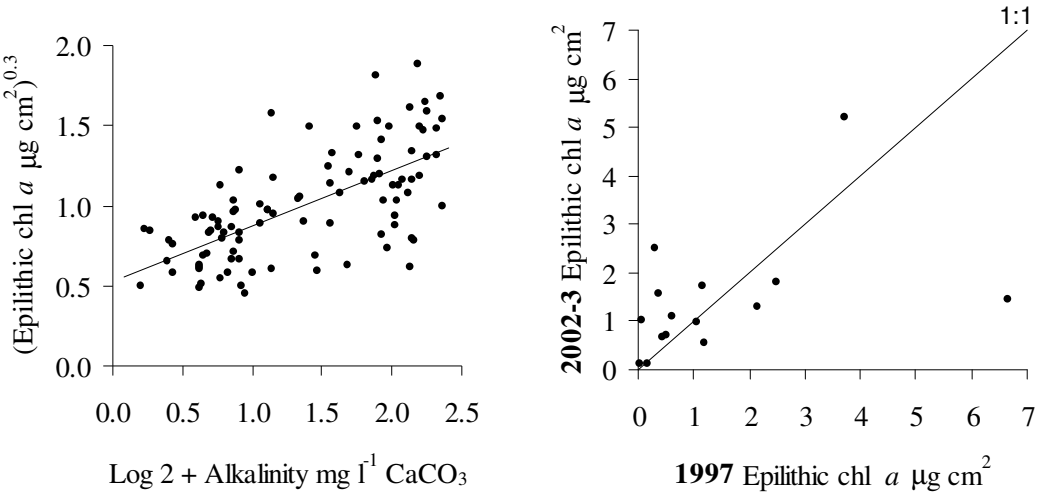


Figure 5.21 Relationship between transformed epilithic chlorophyll *a* and alkalinity (n = 99) and comparison between levels found in 1997 (Irvine *et al*., 2001) and this survey (n = 15).

5.4 Discussion

The development of a biologically based typology is the first step in developing an ecological assessment system for macrophytes. Six groups were initially identified from the cluster analysis and largely supported by the NMS ordination. Three groups were then identified as being distinct both biologically and in terms of the environmental variables: alkalinity, depth and area. No evidence was found for a significant effect of altitude and colour in discriminating between the macrophyte groups. Altitude is unlikely to be important in Ireland because most lakes are at less than 200 m. Its inclusion in the WFD will obviously be of greater relevance to other member states e.g. in the Spanish Pyrenees, altitude and its relationship with ice cover has been found to be an important factor affecting macrophytes (Gacia *et al*, 1994). Colour was not found to be a significant factor separating the clusters. Some influence was expected, as colour is likely to be the principal determinant of light extinction in reference condition (Free *et al*., 2005).

Typology boundaries were selected for the significant environmental variables: alkalinity, depth and area (Table 5.8). The boundaries selected largely encompassed those groups found to be different following CVA and cluster analysis. In addition, MRPP analysis indicated that the majority of types defined were significantly different in terms of macrophyte composition and abundance. The proposed typology was therefore successful in partitioning variation in macrophyte composition and abundance in reference condition. This should allow ecological change to be detected more easily (REFCOND, 2003).

There was a significant under-representation of lakes in the 20 – 100 mg l^{-1} $CaCO_3$ alkalinity band owing to the scarcity of reference conditions. This was a result of reference lake selection, which aimed to select genuine reference lakes rather than 'best available'. One of the benefits of the multimetric and multiple regression models developed, is that once a clear linear response to pressure is demonstrated then reference conditions may be confirmed or extrapolated by the model with few or no reference sites. For example, Figure 5.20 shows that only one reference site was available for the '20 – 100 mg l^{-1} $CaCO_3$, < 4 m' typology, but the linear response (r^2 = 0.64, $p < 0.05$) to pressure found helps to confirm that the site is in reference condition. The site may therefore be used to ecologically assess other lakes of that

type by departure from reference condition. If subsequent paleolimnological evidence indicates that reference lakes are impacted, then reference conditions may be extrapolated. Care must be taken to ensure that a linear response is found over the range to which reference conditions are extrapolated.

Spring total phosphorus (TP) was chosen as the sole pressure against which the multimetric and multiple regression models were developed. Unfortunately, annual values of TP were not available but as TP shows a sinusoidal annual pattern in Irish lakes - being higher in winter and lower in summer (Gibson *et al*., 1996), samples taken in the months of April and May correspond closest to annual averages (Irvine *et al*., 2001). Therefore, spring samples may provide a rough approximation of annual averages for lakes that are only slightly affected by the release of P from sediments or from point-source discharges. For the models developed, 75% of the TP samples were taken during spring with the remainder largely taken in June. One-off measurements of chlorophyll *a* were not considered suitable to use as a surrogate for pressure to develop models owing to the high variation in summer months (Irvine *et al*., 2001).

The multimetric index was successful in describing the response of macrophytes to pressure and should therefore prove a useful tool for the ecological assessment of lakes (Figure 5.16). Figure 5.22 compares the response to eutrophication (MRP and TP) of both the macrophyte multimetric index and the Irish EPA's Q-value assessment system for river macroinvertebrates (McGarrigle, 1998). The multimetric index developed for the macrophytes appears to have achieved similar success in detecting the effects of pressure on ecological quality as the long established Q-value system. Interestingly, a similar response to pressure is observed, despite the comparison being made for different biological groups in different aquatic systems. Figure 5.22 shows an initial rapid deterioration in ecological quality at low anthropogenic pressure (MRP and TP) followed by a more gradual decline. The similarity of the response indicates that the application of a reference-based system may help harmonisation of ecological status classification between rivers and lakes within catchments.

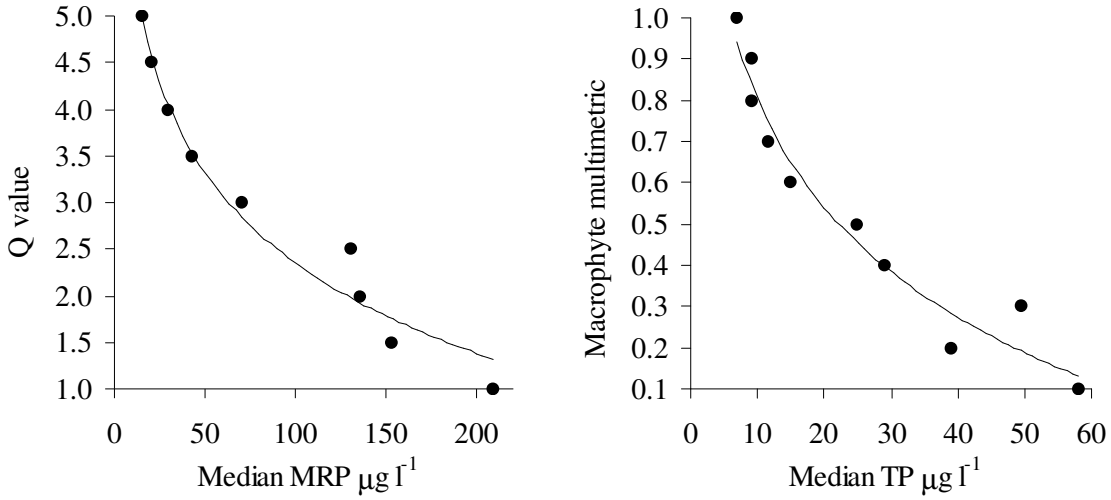


Figure 5.22 Comparison of metric response to pressure (Molybdate Reactive Phosphorus and TP) for the Irish EPA macroinvertebrate Q-Value system for rivers (McGarrigle, 1998) and the macrophyte multimetric developed for this study (> 20 mg l^{-1} $CaCO_3$ alkalinity). Logarithmic trend-lines fitted.

Despite widespread use in the US, multimetric indices have received some criticism. Reynoldson *et al*. (1997) listed the disadvantages of a multimetric approach as: 1) they discard information in comparison to multivariate analysis, 2) redundancy of some metrics, 3) tendency to compound errors and 4) lack of transparency on metric composition. Some of the advantages are that they are easy to interpret and redundancy can be partly beneficial in that it can provide additional evidence for trends. Metrics can also be developed to distinguish different types of pressure. In addition multivariate methods typically use abundance and composition whereas a multimetric index can incorporate several types of biological information such as depth of occurrence or plant health, which hold important ecological information (US EPA, 2004).

The model developed using multiple regression performed slightly better (r^2 = 0.64) than the multimetric index (r^2 = 0.59) for lakes greater than 20 mg l^{-1} $CaCO_3$. Separating the lakes into sub-types generally improved model performance (r^2 = 0.57 to 0.80, Table 5.15). Having models for each type or sub-type may complicate model comparison and interpretation if there is uncertainty whether true reference conditions are available for a type. On the other hand, developing models for specific types will help to take natural variation into account. For example, relative metrics such as RF tolerant taxa and RF filamentous algae were selected more frequently in the models

produced for the high alkalinity lakes (> 100 mg l^{-1} $CaCO_3$). It is likely that relative metrics will be more important in such lakes where there is a large amount of key taxa such as *Chara* in reference condition. In contrast, the plant trophic score and depth of angiosperm colonisation were more important in models of the intermediate alkalinity lakes (20 – 100 mg l^{-1} $CaCO_3$). It is probably more appropriate to use depth of angiosperm colonisation in this alkalinity band because deep colonisers such as charophytes may be naturally infrequent in this group.

In conclusion, a typology was developed that was successful in partitioning variation in macrophyte composition and abundance in reference condition. Two potential ecological assessment systems were presented. The first based on a multimetric model that combined six metrics to assess ecological status. This metric had comparable performance to currently used assessment techniques for macroinvertebrates in rivers. The second was based on multiple regression and produced models for lake sub-types. There is a need for compatible independent data to be gathered to test the models developed.

6. Littoral Macroinvertebrates

6.1 Introduction

"The composition and abundance of benthic invertebrate fauna" as listed in Annex V of the Water Framework Directive is one of the four biological elements in lakes which must be considered for the classification of ecological status. The deviation of this benthic invertebrate community from its low impacted, type-specific reference state, together with similar assessments of the other biological elements, will determine a lake's ecological status. Consequently, the successful implementation of the directive is critically dependent on the establishment of these type specific biological reference communities.

Hynes (1975) stated that "the valley rules the stream" and was instrumental in shaping how we currently view and manage aquatic ecosystems. Water quality is no longer measured solely at a localised or reach scale; instead factors at a catchment or ecoregion scale have been recognised as critical in determining water quality at a local one. The development of GIS technologies, powerful computers and multivariate software packages has led to a wealth of studies in recent years regarding the influence of landscape on aquatic ecosystems. Landscape analysis supports the view that catchments (geology, climate and landuse) determine local ecological attributes such as water chemistry and flow which in turn influences biotic communities

The Water Framework Directive lists latitude, longitude, altitude, depth, surface area and geology as obligatory attributes to define lake types (CEC, 2000). Lake types identified by these abiotic factors require verification using the biological communities they contain. Ecological assessment requires incorporating natural variation into classification schemes and the separation of lakes into natural types ensures that the effects of pressures are assessed by comparison with appropriate reference conditions.

6.2 Methods

6.2.1 Sampling and analysis

The methods used for sampling macroinvertebrates and a brief introduction on the statistical methods and computer packages used was described in Chapter 3.

6.3 Results

6.3.1 Composition of the invertebrate communities used for typology analysis

Macroinvertebrate community data used to develop a lake typology was collected from potential reference lakes sampled in Spring 2001 and 2002, and Summer 2002. Reference lakes were chosen on the basis of existing chemical and biological data, catchment information and expert opinion following recommendations from Wallin *et al*. (2003). They were considered to be affected minimally by anthropogenic pressures, which would result in only very minor changes as outlined in the normative definitions of Annex V of the WFD. The Spring data (n = 69) comprised almost 43,000 macroinvertebrates representing 149 taxa, identified predominantly to species level, encompassing 105 genera. The combined season data set (it was not always possible to get Summer samples from all of the lakes therefore for lakes with both Spring and Summer samples, n = 58) comprised over 67,000 individuals representing 197 taxa encompassing 126 genera (Figure 6.1).

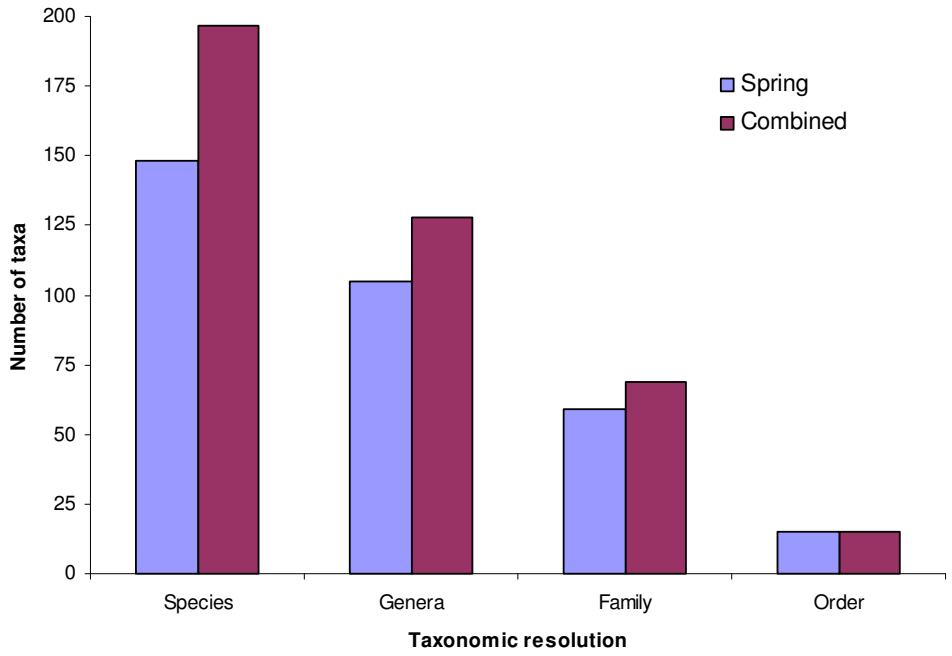


Figure 6.1 Number of taxa distributed across different taxonomic resolutions.

The caddis flies (Trichoptera) and beetles (Coleoptera) were by far the most diverse orders of invertebrates encountered with 35 and 28 taxa, respectively, recorded in Spring (Figure 6.2). The mayflies (Ephemeroptera) were the next most diverse group with 18 taxa, followed by the snails and limpets (Mollusca) and bugs (Hemiptera) with 15 and 12 taxa, respectively. Summer samples concurred with this pattern. Trichoptera and Coleoptera increased to 46 and 43, respectively; Mollusca comprised 23 different taxa; Ephemeroptera and Hemiptera comprised 19 and 15 taxa respectively. Although Trichopterans and Coleopterans comprised almost half of the taxa richness, they nevertheless, accounted for less than 4% and 9%, respectively, of total abundance. The crustacean amphipods *Gammarus* spp. were the most abundant invertebrates encountered and made up almost 30% of the invertebrate numbers in the combined data, followed by the worms (Oligochaeta) and the mayflies which comprised 15 and 14% of the total abundance, respectively. The oligochaetes were not identified to a lower taxonomic resolution due to time constraints which is why they contribute little to species diversity despite their abundance.

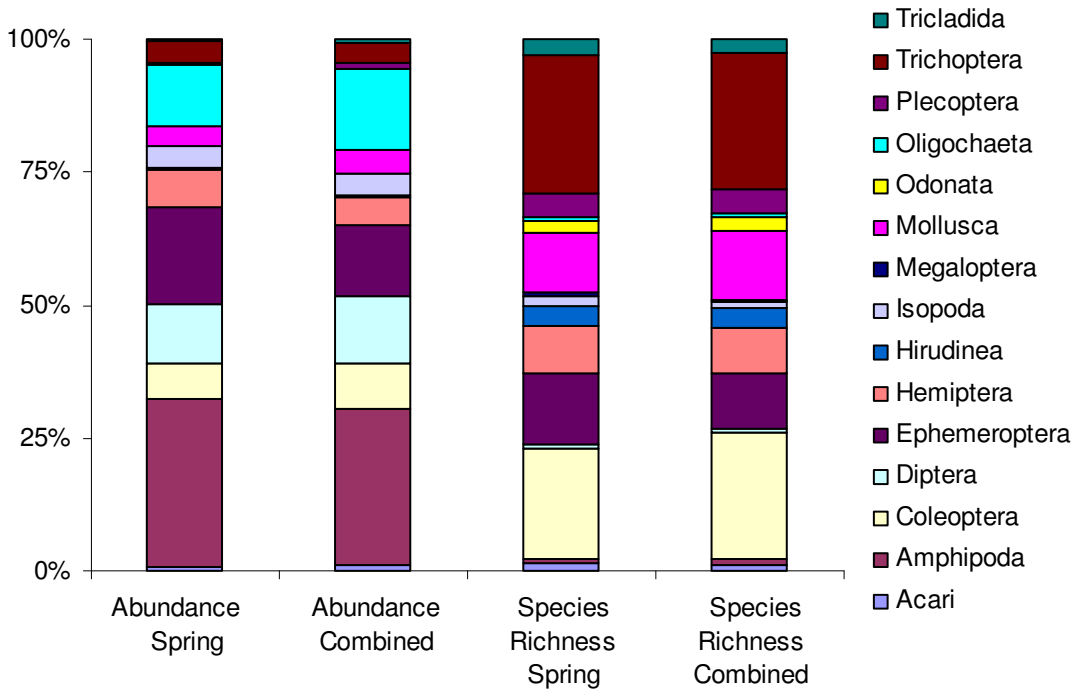


Figure 6.2 Percentage contribution in terms of abundance and species richness of each of the invertebrate orders for spring samples and all samples combined

Invertebrate species recovered from a sample ranged from 6 to 38. Maximum number of species recovered from a lake was 45. Table 6.1 shows species level data from the Spring and combined (Spring and Summer) data.

Table 6.1 Summary statistics showing number of taxa recovered at species level from reference lakes in Spring and Combined (Spring and Summer) data sets.

Data	Min.	Max.	Mean	Median	# Rare taxa	% Rare taxa	Species: genus ratio
Spring	6	38	19.2	18	63	42.2	1.42
Combined	13	45	29.6	29.5	106	53.8	1.58

The number of rare taxa (found in <5% of samples) represented between 42 - 54% of all taxa (Figure 6.3). This is typical of freshwater invertebrate communities. In this study, 49 of the 149 species encountered in Spring and 62 of the 197 species in the combined two season data were found in only one lake.

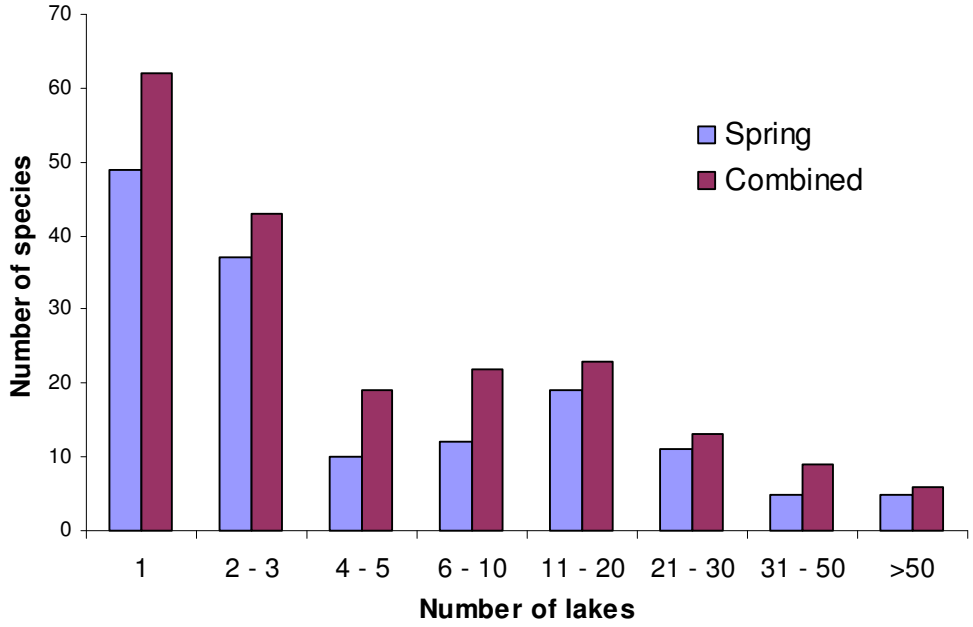


Figure 6.3 Frequency of species occurrences in lakes

6.3.2. Classification using TWINSPAN and cluster analysis.

Two different methods of classification, TWINSPAN and Cluster analysis, were used to group reference lakes on the basis of their biological communities. TWINSPAN (Hill, 1979; Gauch, 1982) is a hierarchical divisive technique that classifies species and samples based on a weighted averaging ordination using correspondence analysis (McCune *et al*., 2002). In TWINSPAN, pseudo-species cut levels of 0, 5, 20, 50, 100 and >100 or their transformed equivalents were chosen. Cluster analysis is an agglomerative technique that clusters samples that are most similar (in this case in terms of species composition). A similarity matrix assesses how similar samples are to each other. City-block (Manhattan) distances were used to calculate the dissimilarities (or distances) between samples and Ward's method was used to evaluate these distances. Manhattan distances are the average distances across dimensions and have the effect of dampening single large outliers. Ward's method uses an analysis of variance to evaluate the distance between clusters which are then joined to minimise the within group variance (Statsoft©, 1998).

Biological invertebrate data at three taxonomic resolutions - species, species excluding rare species and genus level - were subjected to a square root and a presence-absence transformation, the latter of which had the effect of converting a quantitative dataset to a qualitative one. The analysis was completed on single season (Spring) and combined season (Spring and Summer) data as a result of which there are 12 different treatments – 3 taxonomic resolutions x 2 transformations x 2 seasons. Analysis of Similarities (ANOSIM) was undertaken on the end groups generated for each of these 12 treatments to test whether or not these groups differed significantly ($p < 0.05$) from each other (Table 6.2). The strength of the ANOSIM test lies in the fact that the values of the test statistic, R, close to unity are indicative of complete separation of the groups, and small values close to zero imply little or no segregation (Clarke and Warwick, 1994). While Global R values were significant ($p < 0.05$) it should be remembered that it is possible for R values to be significant yet inconsequentially small owing to the number of replicates and the nature of the univariate test (Clarke and Warwick, 1994).

Table 6.2 Global R values from ANOSIM test on different classifications

Data Set	Taxonomic level	No. of taxa	Transformation	TWINSPAN Global R	CLUSTER Global R
Spring	Species	149	Square root	0.372	0.222
Spring	Species	149	Pres/Absc	0.510	0.295
Spring	Species excl. Rare	86	Square root	0.361	0.175
Spring	Species excl. Rare	86	Pres/Absc	0.573	0.469
Spring	Genus	105	Square root	0.427	0.144
Spring	Genus	105	Pres/Absc	0.390	0.508
Combined	Species	197	Square root	0.367	0.556
Combined	Species	197	Pres/Absc	0.417	0.281
Combined	Species excl. Rare	92	Square root	0.352	0.42
Combined	Species excl. Rare	92	Pres/Absc	0.403	0.239
Combined	Genus	125	Square root	0.443	0.424
Combined	Genus	125	Pres/Absc	0.311	0.413

The two different classification procedures used on identical data sets did not return the same groups of lakes. Choosing different levels of taxonomic resolution or different data transformations also resulted in different classifications. While all the classifications produced some groups of lakes that were significantly different from each other in terms of their invertebrate communities, in eight out of the twelve classifications TWINSPAN performed better than the cluster analysis. Overall, and among both tests, only four treatments returned a global R value > 0.5; a value generally accepted where groups separate out reasonably without substantial overlap. The highest value achieved was global R = 0.573 obtained using TWINSPAN classification on Spring presence absence data, excluding rare species. The next highest global R value, R = 0.556, was generated using cluster analysis on the combined season square root transformed species data. Although the classification procedures was completed on 12 data sets using different transformations and varying levels of taxonomic resolution, subsequent analysis concentrated on these two treatments. The output of the cluster and TWINSPAN classifications of these treatments with the biggest global R are shown in Figures 6.4 and 6.5, respectively.

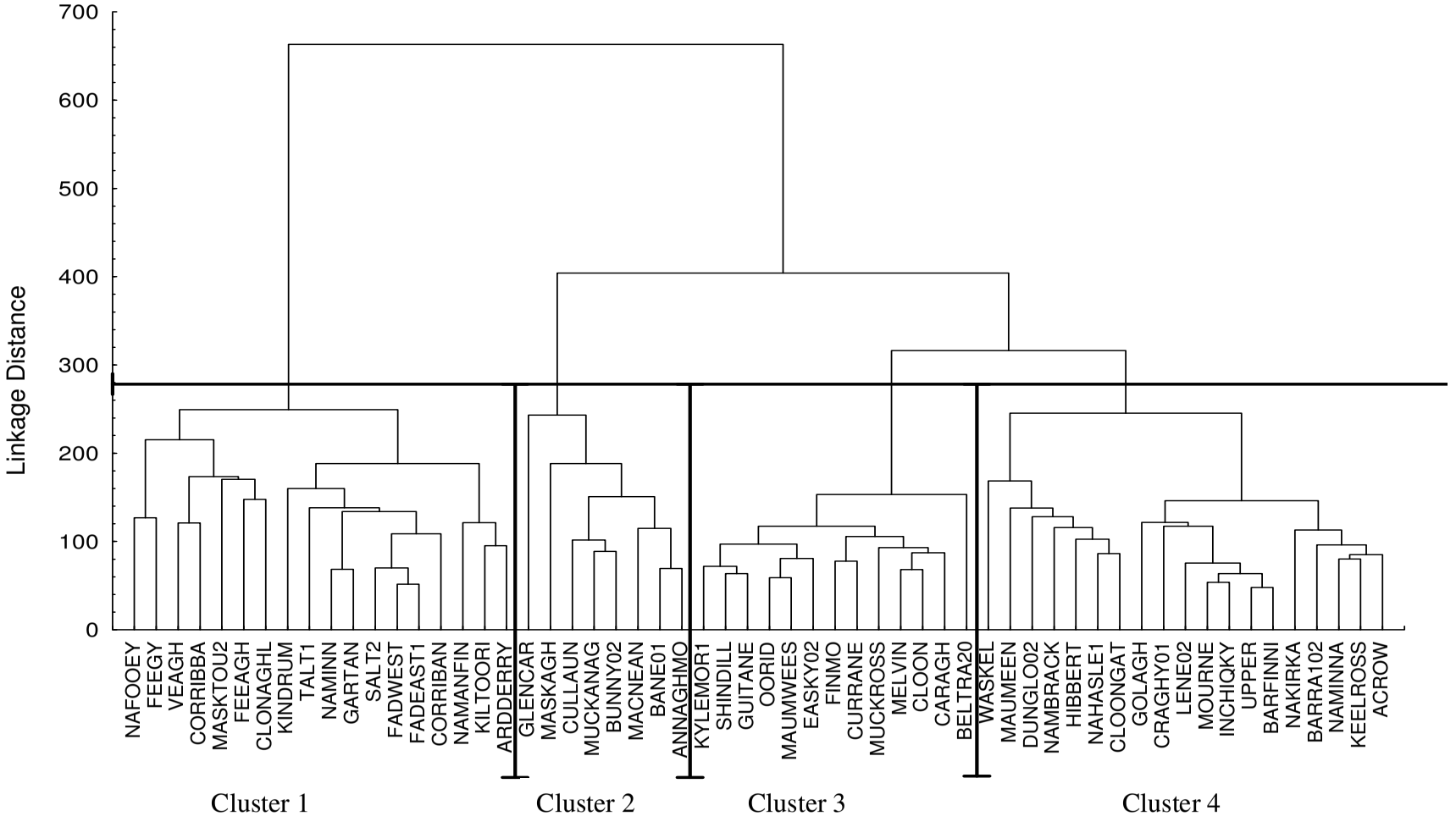


Figure 6.4 Dendrogram from cluster analysis on square root transformed species data using the combined dataset. City Block (Manhattan) distances were used with Ward's linkage method. Horizontal line indicates cut-off distance for 4 clusters.

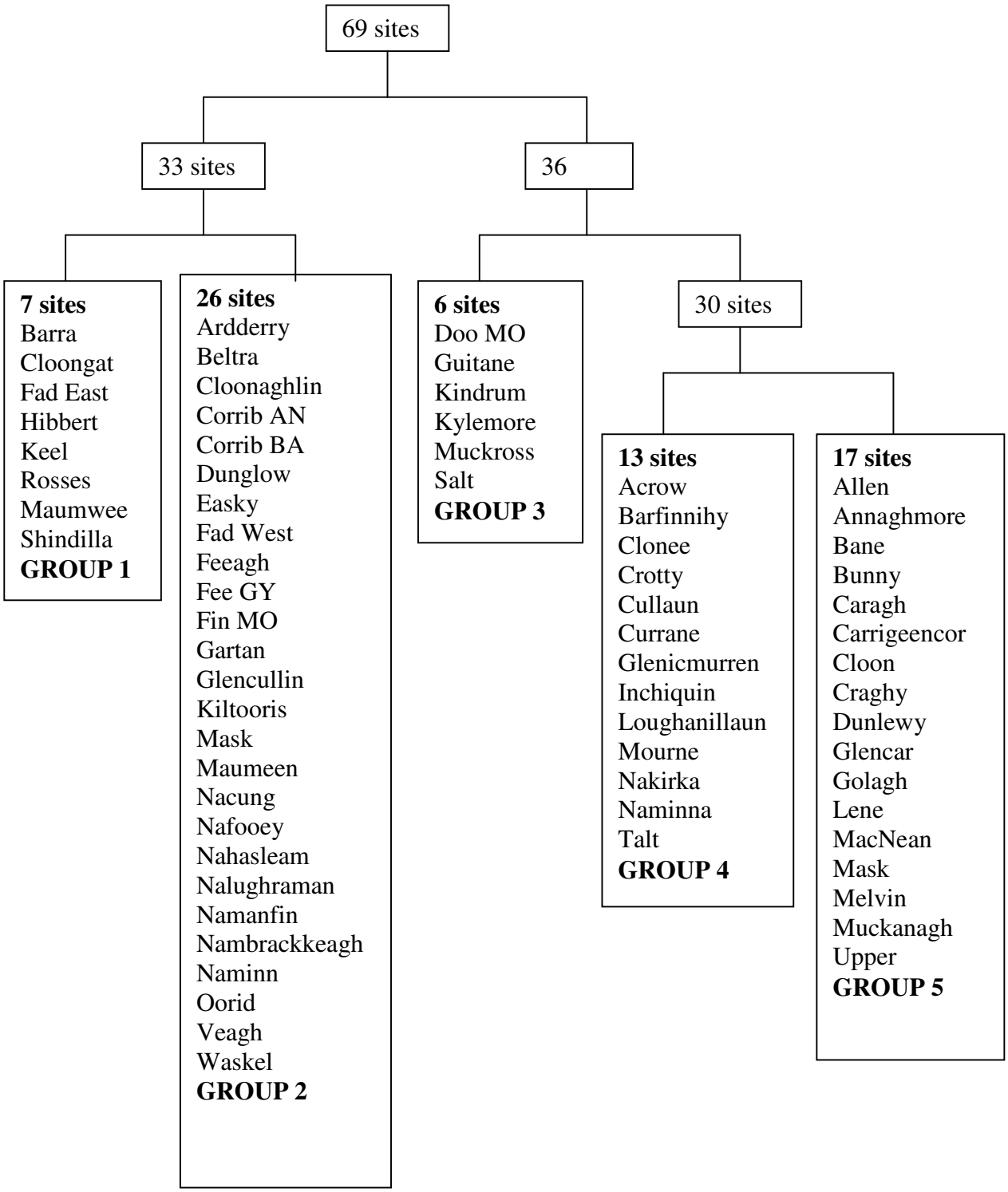


Figure 6.5 TWINSPAN classification based on Spring species presence absence data excluding rare species. Corrib AN = Corrib Annaghdown (east) Corrib BA = Corrib Baurisheen (west)

The results of the ANOSIM test and the Global R values indicate that the end groups derived using the classification techniques support statistically different invertebrate communities. To examine where the main between-group differences arose, it was necessary to examine the R values for each of the pair wise comparisons. The results of the pair-wise tests for the five end groups generated using TWINSPAN analysis and from the four groups generated using CLUSTER are listed in Table 6.3. In the TWINSPAN classification the highest pair wise values were between groups 2 & 3, 1 & 3 and 1 & 5 which all returned R values > 0.72 indicating good separation. Groups 4 & 5 in the TWINSPAN classification reported a pair wise R = 0.25 suggesting there was little to separate lakes in these groups on the basis of their invertebrate communities. However when TWINSPAN groups 4 and 5 were amalgamated and an ANOSIM run on the resulting four end groups the global R decreased to 0.453.

Table 6.3 ANOSIM R statistic results of pair wise comparisons between end groups generated using Cluster analysis and TWINSPAN. Values are significant at $p < 0.001$.

End groups	1:2	1:3	1:4	1:5	2:3	2:4	2:5	3:4	3:5	4:5
TWINSPAN	0.60	0.74	0.51	0.72	0.79	0.59	0.57	0.62	0.46	0.25
CLUSTER	0.66	0.40	0.68		0.65	0.51		0.47		

Table 6.4 shows the environmental characteristics of the end groups generated using the TWINSPAN classification procedure. The largest pair wise R values returned between groups 2 & 3 and 1 & 3 imply that invertebrate communities in these groups of lakes differed from each other. Yet groups 2 and 3 had similar median values of pH, conductivity, colour and altitude and all had relatively large amounts of peat in their catchments. They were relatively low lying, with low colour and alkalinity and median circum-neutral pH. Group 1 lakes had a much narrower alkalinity and pH range than either Group 2 or Group 3, with more peat in their catchments. Group 5 contained most of the high alkalinity lakes and had the lowest amount of peat in the catchments, although the range of values for both alkalinity and percentage peat in this group were wide and consequently low alkaline, high peat lakes were also included in this group. Often, the presence of a small number of samples in a group was responsible for expanding the range of environmental variables. For example, the presence of Lough Corrib in group 2 (alkalinity 107 mg l^{-1} $CaCO_3$) and Loughs Talt and Cullaun (alkalinity 85 and 172 mg l^{-1} $CaCO_3$ respectively) in group 4 were

responsible for greatly expanding the alkalinity ranges seen in these groups. Median alkalinity values for groups 2 and 4 were 6.55 and 6.51 mg l^{-1} $CaCO_3$, respectively.

Table 6.4 Environmental characteristics of TWINSPAN end groups.

	n =	Group 1 7	Group 2 26	Group 3 6	Group 4 13	Group 5 17
pH	**Median**	6.35	6.55	6.89	6.51	8.05
	Range	5.30 – 6.69	5.70 – 8.33	6.59 – 8.27	5.14 – 8.40	5.78 – 8.53
Alkalinity	**Median**	5.02	6.29	6.47	2.02	47.05
mg l^{-1} $CaCO_3$	Range	2.37 – 6.30	0.52 – 107	4.37 – 69.4	-0.4 – 172	2.20 – 209
Conductivity	**Median**	81	93	81	73	152
µS cm^{-1}	Range	54 – 200	33 – 284	56 – 318	40 – 393	58 – 426
Colour	**Median**	29	32	19	16	22
PtCo	Range	18 – 53	10 – 78	13 - 26	3 – 61	1 – 96
Altitude	**Median**	46	39	33	130	45
Meters	Range	14 – 233	3 – 180	8 – 246	6 – 419	14 – 112
Area	**Median**	25	61	143	57	110
Hectares	Range	8 – 70	7 - 17000	29 – 267	4 – 1035	44 – 8343
% Peat	**Median**	99.72	67.80	73.72	77.79	27.48
	Range	30 - 100	17 - 100	51 - 100	0.41 - 100	0 - 94

The environmental characteristics of the four end groups generated using Cluster analysis can be seen in Table 6.5. Again, considerable overlap in the range of environmental variables is evident. The best separation obtained in the groups generated by the cluster analysis was between groups 1 & 4, 1 & 2 and 2 & 3 respectively, all of which returned values pairwise R values > 0.65 (Table 6.3). Groups 1, 2 and 4, had highly different invertebrate communities, yet as with the TWINSPAN analysis, shared similar environmental characteristics in that they had relatively low median pH and alkalinity values and relatively large amounts of peat in their catchments. Group 4 lakes were at the lower end of these ranges and tended to be smaller, at higher altitudes with lower alkalinity and more peat in their catchments. High alkalinity lakes dominated Group 2 which contained eight lakes, seven of which had alkalinity > 90 mg l^{-1} $CaCO_3$; the one remaining lake, MacNean, had an alkalinity of 24 mg l^{-1} $CaCO_3$. Median alkalinity (144 mg l^{-1} $CaCO_3$) of this group was considerably higher than median alkalinity (<10 mg l^{-1} $CaCO_3$) of the other groups. Group 2 also had the lowest median percentage peat values of 11% compared with median percentage peat values > 60% found in the catchments of the other groups. Although groups 2 and 4 had relatively little overlap of the abiotic variables, the pairwise R value between the high alkalinity group 2 lakes and the small acid group 4 lakes was only 0.51. This ranked fourth of six possible pairwise comparisons and just

above the 0.50 cut off below which the test statistic suggests there would be considerable overlap between macroinvertebrate communities of the two groups.

Table 6.5 Environmental characteristics of cluster end groups.

	n =	Group 1 18	Group 2 8	Group 3 13	Group 4 19
pH	**Median**	6.94	8.44	6.74	6.36
	Range	6.30 – 8.33	7.60- 8.53	6.07 – 8.15	5.14 – 8.46
Alkalinity	**Median**	9.09	144.43	5.13	2.98
mg l^{-1} $CaCO_3$	Range	1.99 – 107	23 – 208	0.52 – 54	-0.42 – 104
Conductivity	**Median**	100	324	72	100
µS/cm^{-1}	Range	33 – 318	116 – 462	48 – 170	54 – 250
Colour	**Median**	27	18	19	29
PtCo	Range	15 – 78	1 – 80	10 – 73	3 – 96
Altitude	**Median**	44	24	35	65
meters	Range	6 – 246	16 – 112	6 – 180	3 – 249
Area	**Median**	113	100	132	31
hectares	Range	12 – 17291	50 – 8343	14 – 2206	6 – 416
% Peat	**Median**	67.80	11.48	67.26	77.79
	Range	18 - 100	0 – 40	28 – 100	0 - 100

Box and whisker plots were generated in order to visualise differences between the seven environmental variables among the endgroups generated using the TWINSPAN and cluster classifications. Figure 6.6 illustrates three of these plots for pH, alkalinity and percentage peat. Pair-wise post-hoc tests with Bonferroni adjustment indicated that there were few significant differences in environmental variables among the TWINSPAN groups. Group 5 was significantly different ($p < 0.05$) from groups 1, 2 and 4 in terms of pH and percentage peat and from group 4 in terms of alkalinity. None of the other groups were significantly different from each other for any of the other environmental variables. Despite the fact that Groups 4 and 5 differed significantly in pH, alkalinity and percentage peat, these two groups recorded the lowest pairwise R value ($R = 0.25$) in the ANOSIM test (Table 6.4) indicating considerable overlap in the invertebrate communities they supported.

Among the cluster groups, the post-hoc tests indicated that group 2 (the high alkalinity group) was significantly different from all other groups in terms of pH,

conductivity, alkalinity and percentage peat ($p < 0.001$). Global R values between group 2 and groups 1 and 3 were relatively high (R = 0.66 and 0.65, respectively). R values between groups 2 and 4 were lower at R = 0.51. Groups 1 and 4 were also different in terms of pH, alkalinity ($p < 0.008$) and area ($p < 0.035$). Group 4 were primarily small, acid lakes and the R between group 1 and 4 was the highest found (R = 0.68). None of the other groups were significantly different from each other for any of the other environmental variables.

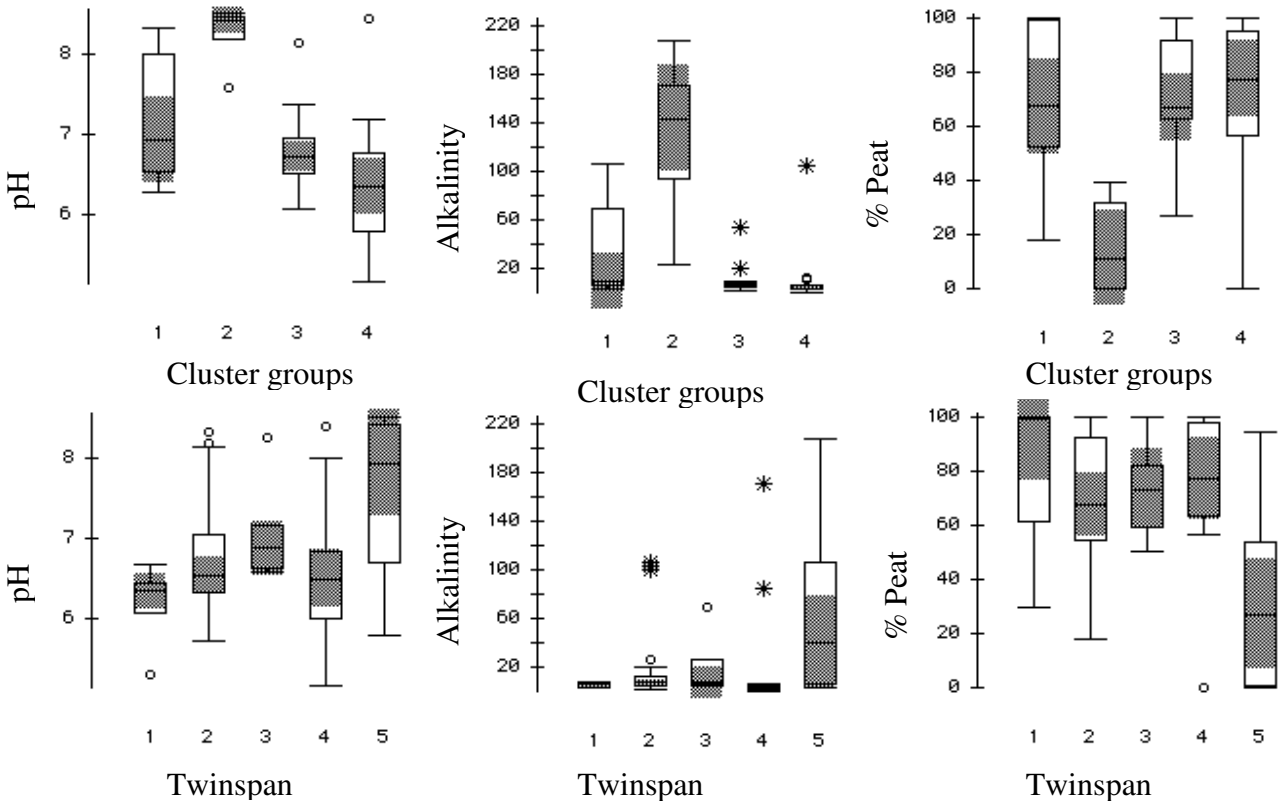


Figure 6.6 Box and whisker plots of end groups generated by TWINSPAN and cluster analysis for the environmental variables pH, alkalinity (mg l^{-1} $CaCO_3$), and % peat. Boxes represent upper and lower 25th percentile, whiskers represent range of data excluding extreme values represented by circles and asterisks. Shaded areas represent 95% confidence intervals.

As the different classification procedures produced different groups of lakes, further investigation was done using a non-metric multi-dimension scaling (MDS) ordination. This maps the samples in multi-dimensional space on the basis of species composition such that samples close together in the ordination are more similar. A Bray-Curtis similarity matrix was generated for each of the data sets and an MDS ordination performed on spring presence absence data excluding rare species, and the combined square root transformed species data. Figures 6.7 (a) and (b) shows the resulting ordinations with sites coded according to the TWINSPAN and cluster end groups, respectively.

The groups generated by the biological classifications were generally supported by the ordinations although there is some overlap among the groups. The stress values for the MDS ordinations were 0.26 and 0.23 respectively. These stress values represent the extent of the disagreement between the dissimilarity matrix and the distances between sites on the ordination plot. Stress values > 0.2 are likely to yield plots which may be difficult to interpret (Clarke 1993) and values > 0.25 indicate that the points are close to being arbitrarily placed in ordination space and any subsequent interpretation should be treated sceptically. Stress values < 0.2 would be potentially useful (Clarke and Warwick, 1994).

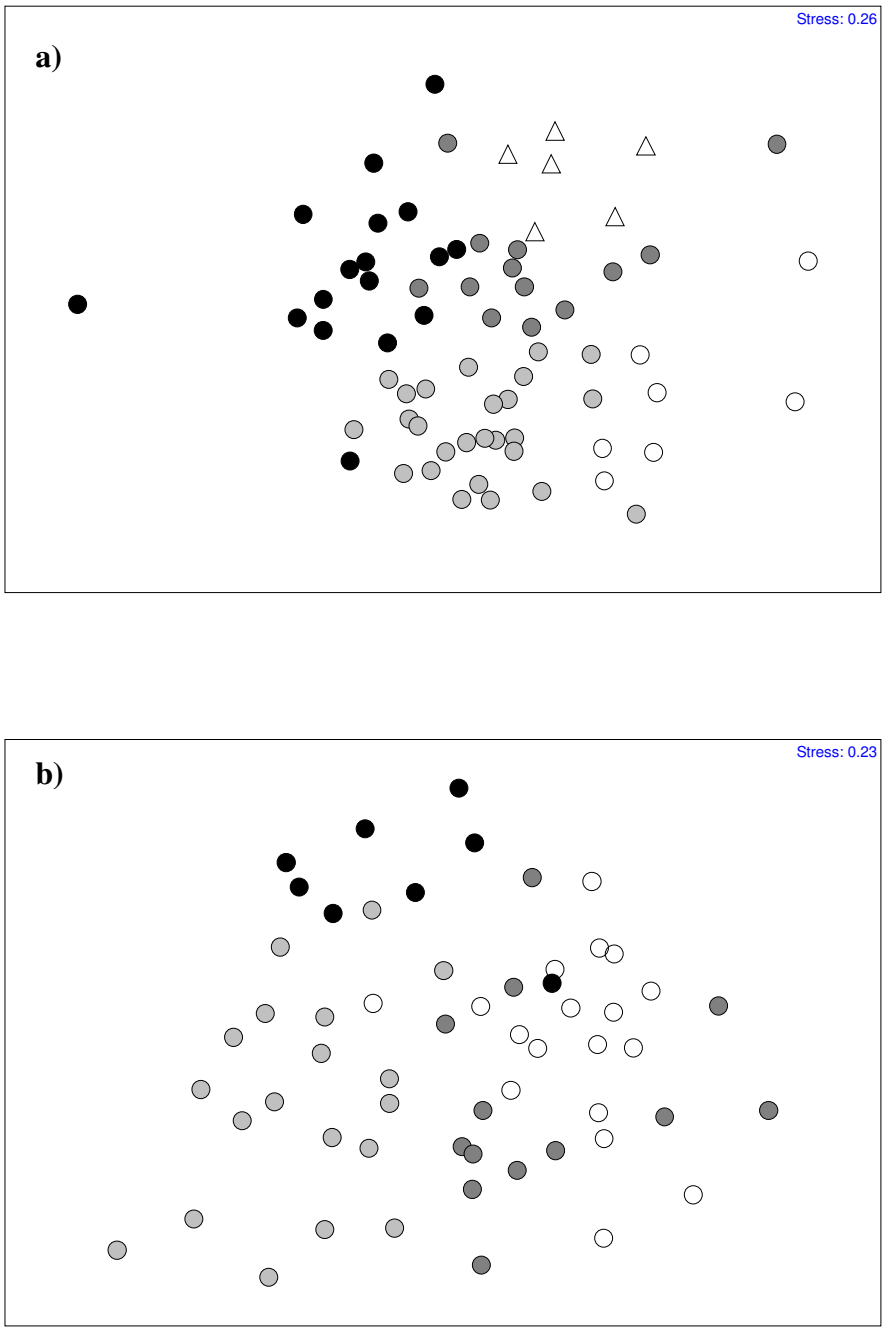


Figure 6.7 MDS ordinations of (a) Spring presence-absence data excluding rare species. Samples are coded according to the end groups generated using TWINSPAN (1○ , 2◎ , 3△ , 4● , 5●) (b) combined species square root transformed data, using Bray-Curtis distance measures. Samples coded according to endgroups generated using cluster analysis. (1○, 2●, 3◎ , 4 ◎) respectively.

6.3.3 Discriminant functions using Canonical Variates Analysis

Canonical variates analysis, an ordination method using discriminant functions, was performed on both datasets. This examines which linear combination of environmental variables can best define *a priori* identified groups.

Seven environmental variables were used to discriminate between biological groups. These were altitude, lake area, alkalinity, pH, conductivity, colour and % peat. All variables, with the exception of pH, were log (x+2) transformed. CVA was performed on the five groups identified by the TWINSPAN classification and the four groups identified by the cluster analysis. Environmental variables were chosen using automatic forward selection and their significance tested by 999 random unrestricted Monte Carlo permutations. Figures 6.8 and 6.9 illustrates the CVA ordinations for TWINSPAN and cluster end groups respectively. Axes 1 and 2 of the CVA ordination plots represented 77.7% and 88.5% of the variation in the relationship between end groups for the TWINSPAN and cluster classifications, respectively (Tables 6.6 and 6.7).

Table 6.6 Summary statistics of CVA ordination on TWINSPAN end groups.

		Axes 1	Axes 2	Axes 3	Axes 4
Eigenvalues	:	0.339	0.195	0.118	0.036
Species-environment correlations	:	0.582	0.441	0.343	0.189
Cumulative percentage variance					
of species data	:	8.5	13.3	16.3	17.2
of species environment relation	:	49.3	77.7	94.8	100.0

Table 6.7 Summary statistics of CVA ordination on cluster end groups.

		Axes 1	Axes 2	Axes 3	Axes 4
Eigenvalues	:	0.593	0.393	0.128	0.872
Species-environment correlations	:	0.770	0.627	0.358	0.00
Cumulative percentage variance					
of species data	:	19.8	32.9	37.2	66.2
of species environment relation	:	53.2	88.5	100	0.0

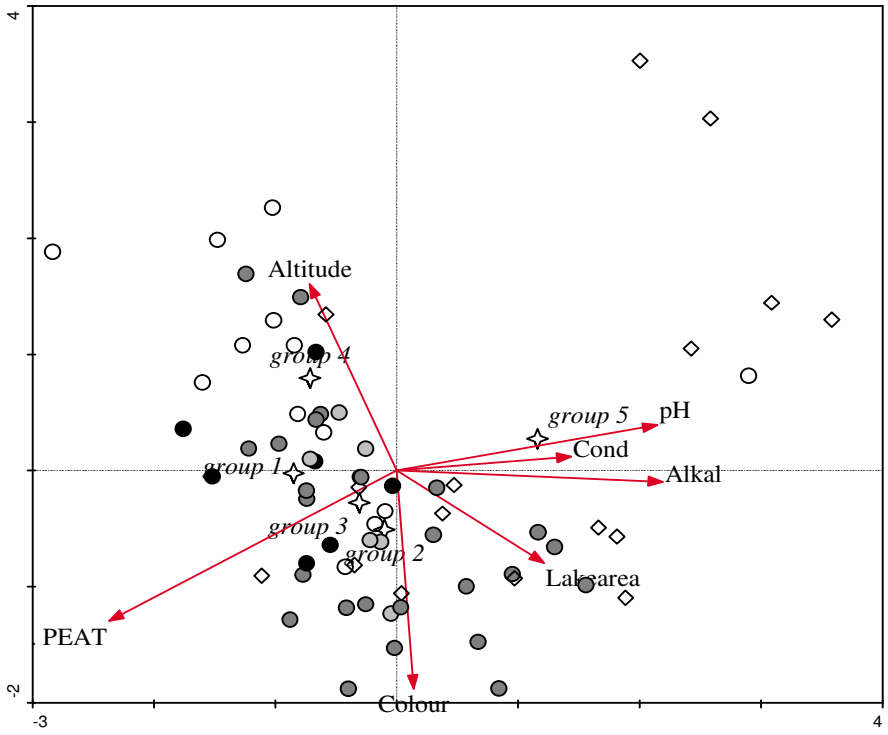


Figure 6.8 Axes 1 and 2 of CVA ordination. Sites are coded according to groups identified using TWINSPAN analysis. Group 1●, 2●, 3○, 4○, 5◇.

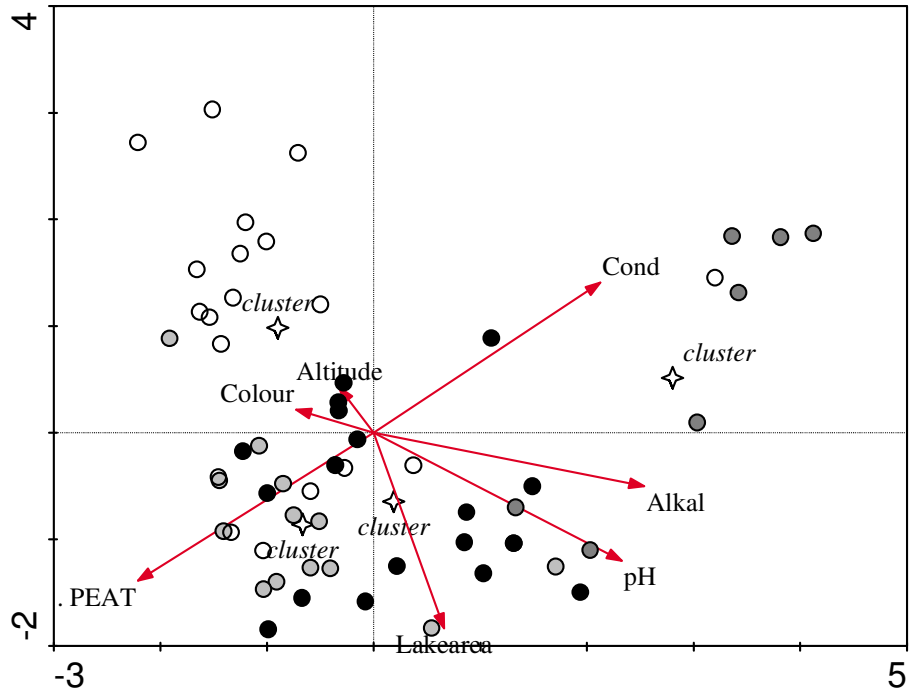


Figure 6.9 Axes 1 and 2 of CVA ordination. Sites are coded according to groups identified using cluster analysis. Group 1●, 2●, 3○, 4○.

The arrows on the CVA plot and the standardised canonical coefficients (Table 6.8) gives an indication of the relative importance of each of the environmental factors in cluster separation. Percentage peat, alkalinity and pH were the most important discriminating factors along axes 1, 2 and 3, respectively, using the TWINSPAN groups. Alkalinity and conductivity were the most important discriminating factors separating cluster groups.

Table 6.8 Standardised canonical coefficients for environmental variables. The strongest discriminating factor on each axis is highlighted in bold font

	TWINSPAN			Cluster		
	Axis 1	**Axis 2**	**Axis 3**	**Axis 1**	**Axis 2**	**Axis 3**
pH	0.1164	0.5543	**-1.4236**	0.237	-0.6737	-0.0321
Conductivity	-0.7574	0.8636	0.806	-0.2566	**1.77**	**-1.0219**
Alkalinity	0.8085	**-1.6797**	-0.2906	**1.4018**	-1.533	-0.0087
Colour	0.4034	-0.5568	0.3718	0.2528	0.0001	-0.482
Altitude	-0.1602	0.7279	0.1977	0.162	0.2733	-0.7029
Lake area	0.1159	0.1304	0.3849	-0.2981	-0.1124	-0.327
% Peat	**-0.9502**	0.6645	-0.8645	-0.4889	-0.8384	-0.8614

Results of the forward selection of environmental variables can be seen in Tables 6.9 and 6.10. Forward selection finds the eigenvalue associated with each environmental variable separately then the next most important variable after the effects of the strongest variable have been taken into account. The marginal effects (percentage variance explained by each variable alone) indicates that percentage peat was the best environmental variable for discriminating between TWINSPAN groups while alkalinity followed by pH and percentage peat were the most efficient at discriminating between groups generated in the cluster analysis. The conditional effects lists the variables in order of inclusion in the model, the additional variance explained and whether or not inclusion of this additional variable was significant. For the TWINSPAN groups, percentage peat was the only significant environmental variable and the inclusion of additional variables did not increase the amount of variance explained. For the cluster end groups, alkalinity, conductivity, and percentage peat were the significant variables that contributed to explaining the variance in the data. The results produced using automatic forward selection of the seven environmental variables suggested that alkalinity (or surrogates of alkalinity - conductivity, % peat and pH) were all significant variables and contributed most to explaining the variance in the data .

Table 6.9 Summary of automatic forward selection in CVA ordination for TWINSPAN end groups. Environmental variables were log (x+2) transformed except pH. Significant p values are highlighted in bold font.

Marginal effects		Conditional Effects			
Variable	Lambda -1	Variable	Lambda-A	p	F
% Peat	0.29	% Peat	0.29	**0.002**	5.17
pH	0.23	Colour	0.11	0.079	2.04
Alkalinity	0.22	pH	0.10	0.123	1.88
Conductivity	0.12	Altitude	0.05	0.411	1.01
Colour	0.10	Conductivity	0.05	0.488	0.85
Lake Area	0.09	Alkalinity	0.05	0.412	1.03
Altitude	0.08	Lake Area	0.04	0.677	0.6

Table 6.10 Summary of automatic forward selection in CVA ordination for cluster end groups. Environmental variables were log (x+2) transformed except pH. Significant p values are highlighted in bold font.

Marginal effects		Conditional Effects			
Variable	Lambda -1	Variable	Lambda-A	p	F
Alkalinity	0.54	Alkalinity	0.54	**0.001**	12.17
pH	0.48	Conductivity	0.27	**0.001**	6.92
% Peat	0.46	% Peat	0.17	**0.009**	4.4
Conductivity	0.42	Altitude	0.06	0.174	1.77
Lake area	0.11	Colour	0.03	0.517	0.79
Colour	0.09	Lake area	0.02	0.586	0.64
Altitude	0.02	pH	0.02	0.629	0.53

6.3.4 Ordination using Canonical Correspondence Analysis

Canonical Correspondence Analysis (CCA) was used to examine the underlying relationship between the environmental variables and the *community* data, as opposed to CVA which looked at the relationship between the environmental variables and defined *groups*. CCA is designed to detect patterns of variation in species data that can be explained by measured environmental variables (Jongman *et al*., 1995). The resulting ordination illustrates the major patterns of biological variation with respect to underlying environmental variables by representing the species-weighted average optima.

CCA was performed on the data sets analysed earlier - spring presence-absence data excluding rare species - and combined season square root transformed species data

using seven environmental variables as before - altitude, lake area, alkalinity, pH, conductivity, colour and percentage peat. The environmental variables, excluding pH, were log (x+2) transformed prior to analysis. They were chosen using automatic forward selection and their significance tested using 999 randomised unrestricted Monte Carlo permutations. Figures 6.10 and 6.11 shows the resulting ordination plots. Each sample has been coded according to group membership from TWINSPAN or cluster classification. Results indicated that axes 1 and 2 of the CCA ordination plot represented 44.7% of the variation present in the spring data set and 55.6% of the variation seen in the combined data set data (Tables 6.11 and 6.12). Monte Carlo permutation tests indicated six of the seven abiotic factors were significant ($p < 0.013$). Colour was the only environmental variable found to be not significant.

Table 6.11 Summary statistics of CCA ordination on spring presence-absence data, excluding rare species, using automatic forward selection of 7 environmental variables.

		Axes 1	Axes 2	Axes 3	Axes 4
Eigenvalues	:	0.158	0.098	0.084	0.075
Species-environment correlations	:	0.869	0.820	0.770	0.775
Cumulative percentage variance					
of species data	:	4.3	7.0	9.4	11.4
of species environment relation	:	27.6	44.7	59.5	72.7

Table 6.12 Summary statistics of CCA ordination on combined season species data using automatic forward selection of 7 environmental variables.

		Axes 1	Axes 2	Axes 3	Axes 4
Eigenvalues	:	0.158	0.123	0.059	0.055
Species-environment correlations	:	0.881	0.786	0.771	0.720
Cumulative percentage variance					
of species data	:	7.6	13.6	16.4	19.1
of species environment relation	:	31.3	55.6	67.3	78.1

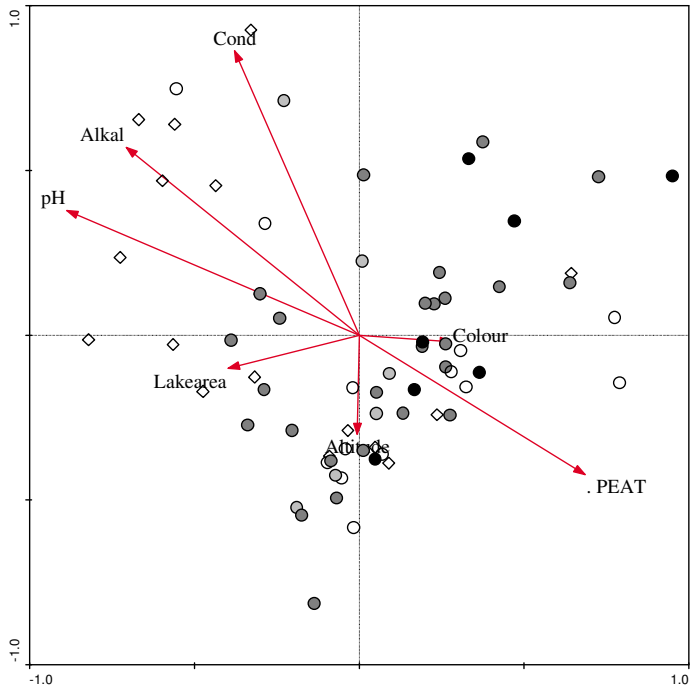


Figure 6.10. Axes 1 and 2 of CCA ordination using seven environmental variables and Spring presence-absence data excluding rare species. Sites are coded according to groups identified using TWINSPAN analysis. Group 1●, 2●, 3●, 4○, 5◇.

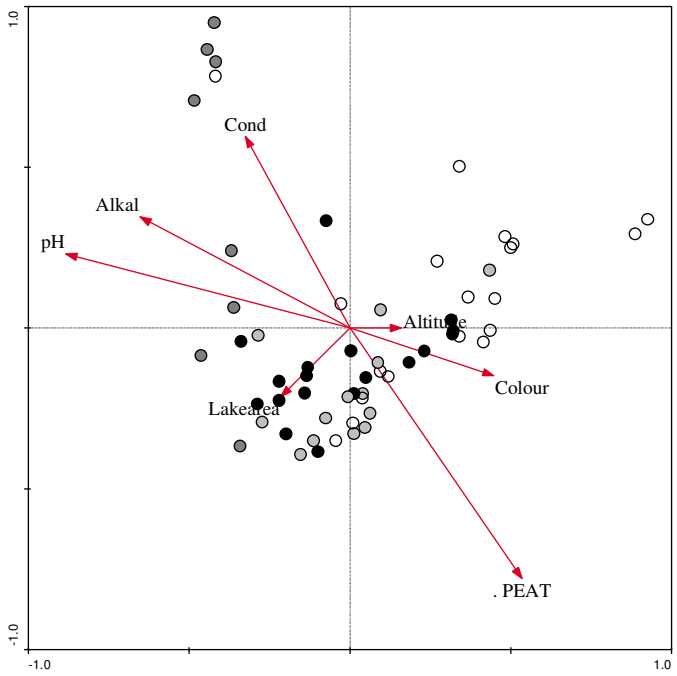


Figure 6.11. Axes 1 and 2 of CCA ordination using seven environmental variables and combined season square root transformed species data. Sites are coded according to groups identified using cluster analysis. Group 1●, 2●, 3●, 4○.

The length and direction of the arrows on the CCA ordination and the standardised canonical coefficients (Table 6.13) give an indication of the relative importance of each of the environmental factors in explaining the variation in the biotic community. pH, conductivity and alkalinity were the most important underlying variables along axes 1, 2 and 3, respectively, for both data sets.

Table 6.13. Standardised canonical coefficients obtained for the environmental variables.

	Spring Data			Combined Data		
	Axis 1	Axis 2	Axis 3	Axis 1	Axis 2	Axis 3
pH	-1.2041	-0.3061	-0.2612	-0.9755	-0.1994	-0.6036
Conductivity	0.6953	1.2623	0.7541	0.9243	0.9615	0.0251
Alkalinity	-0.0395	0.0512	-1.3521	-0.6106	-0.7773	1.2566
Colour	-0.1269	0.0557	0.0207	0.0456	0.1424	-0.2112
Altitude	-0.0212	0.0099	0.6906	0.2853	0.2125	-0.7318
Lake Area	0.0181	-0.4062	0.3756	0.2753	-0.1538	-0.755
% Peat	0.3057	0.0946	-1.1015	0.064	-0.9377	0.6443

Tables 6.14 and 6.15 show the results from the forward selection of environmental variables. The marginal effects (percentage variance explained by each variable alone) indicates that pH, alkalinity, percentage peat and conductivity were the variables that explained the maximum amount of variation in both data sets, while lake area, altitude and colour explained the least. The conditional effects lists the variables in order of inclusion in the model, the additional variance explained after the effects of the stronger variables have been taken into account and whether or not inclusion of this additional variable was significant. pH, conductivity and percentage peat were the only significant variables explaining variation in the Spring data set while all the variables, apart from colour were significant in the combined season data.

Table 6.14. Summary of automatic forward selection in CCA ordination using Spring presence-absence data excluding rare species and seven environmental variables.

Marginal effects		Conditional Effects			
Variable	Lambda -1	Variable	Lambda-A	p	F
pH	0.14	pH	0.14	**0.001**	2.74
Alkalinity	0.12	Conductivity	0.11	**0.001**	2.03
% Peat	0.12	% Peat	0.07	**0.013**	1.51
Conductivity	0.10	Lake area	0.07	0.067	1.33
Lake area	0.08	Altitude	0.07	0.021	1.37
Altitude	0.07	Alkalinity	0.06	0.107	1.24
Colour	0.06	Colour	0.05	0.504	0.98

Table 6.15. Summary of automatic forward selection in CCA ordination using combined species data with seven environmental variables.

Marginal effects		Conditional Effects			
Variable	Lambda -1	Variable	Lambda-A	p	F
pH	0.14	pH	0.14	**0.010**	3.99
% Peat	0.12	% Peat	0.10	**0.010**	2.94
Alkalinity	0.11	Conductivity	0.07	**0.010**	2.23
Conductivity	0.09	Lake area	0.05	**0.013**	1.69
Colour	0.06	Altitude	0.06	**0.004**	1.74
Lake area	0.06	Alkalinity	0.05	**0.013**	1.55
Altitude	0.05	Colour	0.04	0.128	1.23

The CCA ordination plotted samples in multidimensional space but used the underlying environmental variables to constrain the results such that the maximum amount of community structure that could be explained by the environmental variables was examined. All of these variables, apart from lake area, are naturally autocorrelate; pH, alkalinity and conductivity are positively correlated with each other and are negatively correlated with percentage peat and altitude, thus there is only one major environmental gradient. Several groups of lakes identified using the biological classifications failed to separate out clearly on the CCA ordinations. Groups generated using TWINSPAN show considerable overlap with all other groups. Some of the group 5 sites, which included most of the high alkalinity lakes, are located on the left hand side of the ordination along the alkalinity gradient. However, as this was a relatively large group (n = 17), with a wide alkalinity range (2 – 209 mg l^{-1} $CaCO_3$), it was inevitable that the low alkalinity sites fell out on the opposite side of the

ordination (Figure 6.10). Groups generated using cluster analysis separated out a little better, although again there was overlap among some groups. Group 2 which contained high alkalinity lakes, fell out along the alkalinity gradient and lie to the left in the ordination. Group 4 lakes, primarily the small acid lakes were at the opposite end of this gradient and generally in the right hand side of the ordination (Figure 6.11). Although the CCA axes explain approximately half of the variation in biotic data, compatibility between the location of the sites in the ordinations and the groupings developed using biological classifications based upon species assemblages was poor.

6.4 Discussion on typology investigations

Multivariate statistical tools were used to determine if distinct types of lakes were evident on the basis of their littoral macroinvertebrate communities. TWINSPAN and cluster analysis are classification techniques that group samples into discrete classes on the basis of species composition using a set of quantitative criteria. Non-metric multi-dimensional scaling (MDS) is an ordination method which summarises and displays community data graphically in order to enable patterns to be extracted. These methods have been widely recommended as robust techniques (Minchin, 1987) which have a tendency to produce consistently reliable results (Moss *et al*., 1999). However, although these multivariate statistical methods are viewed as objective, Jackson (1993) acknowledged that multiple, discordant and equally objective solutions can occur using the same data. Classification techniques will also produce groups even where none exist and subsequent interpretations may be more dependent on the methodologies employed than any inherent ecological relationship (Jackson, 1997).

The tendency of different classification techniques to produce different solutions using the same data was evident in this study. TWINSPAN and cluster analysis produced different lake classifications when performed in parallel on 12 datasets. The classification procedures that produced the highest global R values in the subsequent ANOSIM test were further analysed to examine the robustness of the classifications.
TWINSPAN produced five endgroups using spring presence absence data, excluding rare species and returned the highest global R values. The majority of the high alkalinity sites fell out into group 5, though these lakes only made up half of the members of this group; the other half comprised low alkalinity lakes. The remaining

four endgroups were dominated by low alkalinity lakes although three of these contained some moderate to high alkalinity lakes. Consequently, the ranges of environmental characteristics for all of the end groups overlapped. This was confirmed by the post-hoc tests, which found few significant differences in the environmental variables between these groups. The high alkalinity Group 5 was different from groups 1, 2 and 4 in terms of pH and percentage peat and also from group 4 in terms of alkalinity. None of the other groups differed significantly from each other in terms of the environmental variables. However, although the environmental variables associated with group 5 were the most different from all the other groups, pairwise comparisons from the ANOSIM test revealed that the relatively low alkalinity lakes in groups 2 and 3 and in groups 1 and 3 supported invertebrate communities that were more different from each other than they were from the relatively high alkalinity lakes in group 5. Additionally, group 5 returned the lowest pairwise R value in the ANOSIM test with group 4 (R = 0.25) indicating little separation in terms of the invertebrate communities they contained despite being the most different in terms pH, alkalinity and percentage peat.

The cluster analysis on the combined transformed species data resulted in the formation of four end groups and returned the second highest Global R value. The high alkalinity groups clustered out together with one moderate alkalinity lake included in this group. The other three groups consisted primarily of low alkalinity lakes, again with one or two moderate or high alkalinity lakes per group. Among the low alkalinity lakes, the small acid lakes generally fell out into group 4. Post-hoc tests showed that pH, alkalinity, conductivity and percentage peat of the high alkalinity lakes were significantly different from all other groups. Global R values from the ANOSIM test were relatively high (> 0.65) for comparisons with groups 1 and 3, respectively, while R values for comparing high alkalinity group 2 lakes with small acid group 4 lakes was slightly lower (R = 0.51).

The classification techniques also produced different results when the data were subjected to different transformations or different levels of taxonomic resolution. Different results were also obtained when rare species were excluded. The level of taxonomic resolution and the inclusion of rare species are both contentious issues. Many argue that species level identifications are necessary for freshwater monitoring

as they increase sensitivity and allow for the detection of subtle changes (e.g. Resh and Unzicker, 1975; Guerold, 2000; Lenant and Resh, 2001; Schmidt-Kloiber and Nijboer, 2004). Alternative views have been put forward by, *inter alia*, Furse *et al*. (1984), Bowman and Bailey (1997), Bailey *et al*. (2001), Reynoldson *et al*. (2001) and Waite *et al*. (2004), who argued that higher levels of taxonomic resolution provide the same information as species level identifications. Rare species are, routinely, deleted from data sets with little or no biological or statistical justification in the belief that the information they provide does not compensate for the noise in the data they create (e.g. Faith and Norris, 1989; Marchant, 1999). Others argue that the abundance of ubiquitous species represents most of the redundant information and emphasise the value of rare species in detecting ecological change, as they are more sensitive to disturbance than abundant species (Cao and Williams, 1999; Nijboer and Schmidt-Kloiber, 2004)

Using single season (Spring) or two season (combined Spring and Summer) data, at species level, genus level, species level excluding rare species, and on quantitative square root transformed data or qualitative presence-absence data, resulted in multiple, objective solutions. A comparison of several different classification techniques should provide evidence of the robust nature of the results and similar patterns using a variety of techniques, should be representative. Non-metric multidimensional scaling (MDS), performed in order to further explore the patterns generated using the TWINSPAN and cluster classifications, revealed stress values of the ordination plots that were high enough to undermine reliable interpretation. Malmqvist and Hoffsten (2000) reported similar results from 120 stream sites in Sweden. The location of their sites indicated a strong gradient in pH and the authors concluded that while there was a clear (geographical) trend in community structure, transitions were smooth and there were no obvious categories into which sites could be readily assigned.

The four end groups produced in our study using cluster analysis reflected the subjective nature of Ward's linkage methods and Manhattan distances. Commonly, clustering methodology uses Unweighted Pair-Group Averages linkage and Euclidean distances. When these methods were employed on our data, no clear endgroups were identifiable, irrespective of taxonomic resolution or transformation. The resulting

dendrogram was highly chained. This was avoided by using Ward's linkage method in conjunction with Manhattan distances. However, when used together these two methods are deemed to violate one of the rules for updating the underlying similarity matrix. MDS failed to extract a pattern from the data and TWINSPAN produced five endgroups. That TWINSPAN classification produces clear end groups has been cited as one of its main attractions, (McCune and Grace, 2002), but the tendency of TWINSPAN to produce tidy endgroups is not necessarily indicative of a more powerful method but might reflect the necessity of the program to form divisive endgroups irrespective of whether they exist or not.

Canonical variates analysis was used on the biological classifications in order to examine which linear combination of environmental variables could best discriminate between the *a priori* groups and revealed that 78% and 88% of the variation could be explained in, respectively, the TWINSPAN and cluster endgroups. Percentage peat was the only significant variable in discriminating among the TWINSPAN groups while alkalinity, conductivity and percentage peat were significant in explaining the variation among the cluster groups. Altitude, colour, lake area and pH were not significant and their inclusion in the model did not increase the amount of variation explained for either classification. The CVA ordination did not separate the groups of lakes clearly in multi-dimensional space. The CVA ordination of the cluster groups, with the predominantly small acid group 4 lakes and high alkalinity group 2 separating out relatively well, provided a better separation of these groups than for the TWINSPAN groups. CVA uses environmental variables to discriminate between *a priori* identified groups, and although groups tended to be dominated by either high alkalinity or low alkalinity lakes, essentially the range of environmental variables in each group was similar.

Examining the community data using canonical correspondence analysis revealed that 45% of the variation present in the spring data set and 56% of the variation in the combined season data could be explained by axes one and two of the ordination. pH, conductivity and percentage peat explained significantly the variation in the spring data, while these three variables along with alkalinity, altitude and lake area were correlated significantly with the axes using the combined data. Imposing the biological groups on the CCA ordination revealed a similar pattern to that obtained

with the CVA. There was considerable overlap between some groups while the smaller acid lakes and higher alkalinity ones tended to separate out better than the rest, albeit still with some overlap. The samples in the CCA ordination are located in space constrained by the underlying measured environmental variables. The major underlying gradient was alkalinity, which explained up to 56% of the variation present in the invertebrate community.

The underlying assumption of the Water Framework Directive is that the abiotic physical and chemical factors of a lake determine the the structure and composition of its biological communities. Although there are six physical descriptors which must be considered when typing lakes, - latitude, longitude, altitude, mean depth, surface area and underlying geology, only three of these were considered in this work. Latitude and longitude were not used because of the small size of the island. Mean depth was not considered a useful factor for littoral invertebrates. Alkalinity was chosen as a surrogate for geology. Up to 56% of the variation found in the invertebrate community could be explained by the measured environmental variables using canonical correspondence analysis, implying that other factors were responsible for the remaining 44%. These factors could include*, inter alia*, additional localised abiotic variables such as lake shape, water level fluctuations, substrate size and percentage silt. The role of biotic factors such as inter and intra species competition for food and space, vertebrate and invertebrate predation, food availability, immigration and emigration abilities and parasitism are also likely to affect the structure of littoral invertebrate communities. Wiley *et al.* (1997) demonstrated that aquatic systems are both physically and biotically structured and while landscape analysis suggests that aquatic communities can be easily predicted by large scale geographical features, communities were often driven by predation, competition and disease. Hawkins *et al*. (2000) found that while landscape classification accounted for more variation than would have occurred randomly and was responsible for partitioning some of the biotic variance, more precise methods were needed to account for the influence of local environmental features on the biota. This view has been supported by further investigations including those by Allan and Johnson (1997), Johnson *et al*. (2004) and Malmqvist and Hoffsten (2000), concluding that regional differences in climate and landuse were strong determinants of large scale patterns, but at a smaller scale,

species distribution was determined fundamentally by physical and biological variables prevailing at the local or habitat scale.

6.4.1 Conclusions on typology investigations

Multivariate statistics are useful exploratory tools and despite some inconsistency among the various methods employed, this study shows a clear tendency for littoral invertebrate communities to be dependent on an underlying alkalinity gradient. Apart from this distinction into hard and soft water lakes, the littoral invertebrate community in Ireland is quite homogenous. In general, the alkalinity gradient is responsible for over 50% of the variation observed in the communities. Local factors, not included in the broad landscape typology approach, appear to be responsible for the rest of the variation. From the point of view of establishing reference communities therefore, a divide is proposed between low and high alkalinity – following the alkalinity bands proposed for macrophytes - as the main determinant of the expected species in littoral samples for any given lake. After that, it is not easy to specify landscape scale factors that control the reference communities. Thus, definition of reference communities is relatively straightforward - a broad division between low and high alkalinity lakes is required. Beyond that, the data from the candidate reference lakes suggests that the littoral invertebrate communities are relatively homogenous and a similar reference community will be applicable to large or small, shallow or deep, high and low altitude lakes once the alkalinity category is established. There is, however, a need to investigate further, the habitat requirements of macroinvertebrates and the importance of spatial and temporal scales for robust monitoring.

It was not possible to define typing boundaries using the invertebrate community and three alkalinity bands were chosen (< 20, 20 – 100, > 100 mg l^{-1} $CaCO_3$) using expert judgement and taking into account boundaries suggested by the phytoplankton and macrophyte communities (Chapters 4 and 5, respectively).

6.5 Reference communities in different lake types

Visual inspection of the littoral community did not reveal any large shifts in the major invertebrate orders in terms of relative abundance across an alkalinity gradient (Figure 6.12). Two samples, each containing over 1000 individuals of the tiny Hemipteran *Micronecta poweri*, were responsible for the increased Hemiptera levels in the moderate alkalinity lakes.

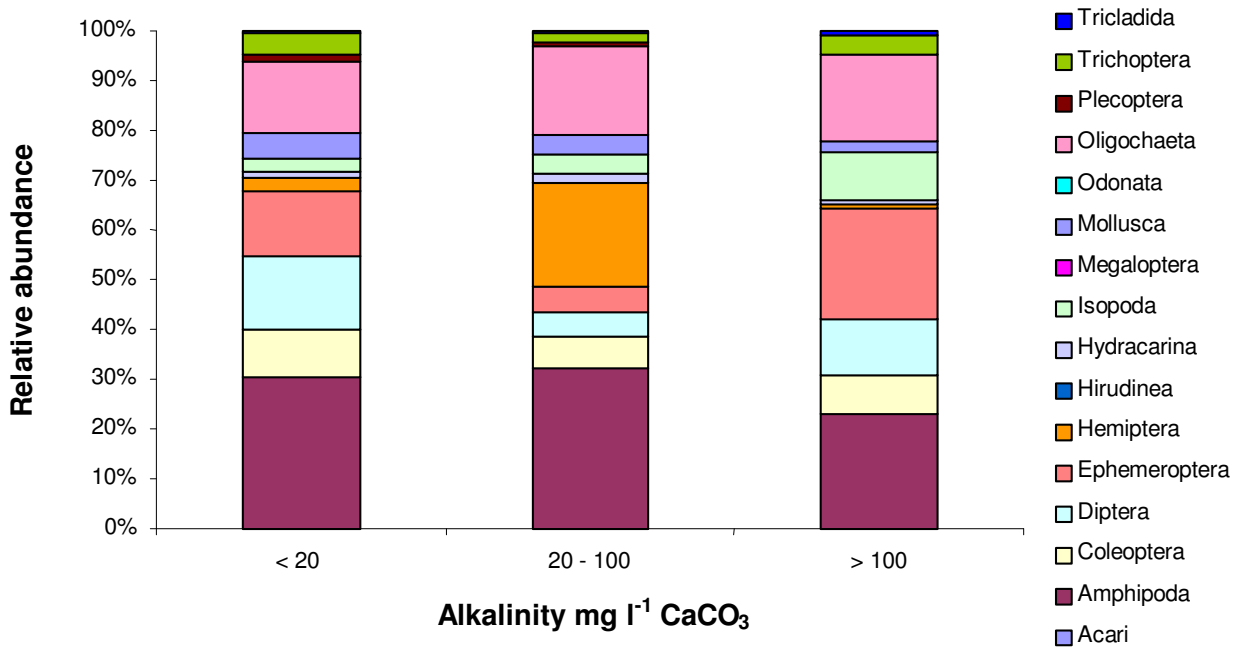


Figure 6.12 Bar chat illustrating relative abundance of major invertebrate orders across an alkalinity gradient. Sample size; n = 41, 7 and 10 for alkalinity < 20, 20 – 100 and > 100 mg l^{-1} $CaCO_3$, respectively.

Spearman rank correlation coefficients revealed that 27 of the 197 taxa in the combined log (x+1) transformed abundance data set were significantly ($p < 0.05$) correlated with alkalinity (Table 6.16). Four of these were negatively correlated and the remaining 23 were positively correlated. When binary data was examined an additional four taxa were correlated with alkalinity, one negatively so and the other three positively.

Table 6.16 Spearman rank correlation coefficients for 31 taxa from the combined data set that were significantly correlated with alkalinity ($p < 0.05$). n.s.=not significant.

Taxa	Log (x+1) abundance	Presence/Absence
Ancylus fluviatilis	n.s.	-0.28
Asellus aquaticus	0.74	0.62
Athripsodes arterrimus	0.54	0.46
Bithynia tentaculata	0.58	0.51
Caenis horaria	n.s	0.29
Caenis luctousa	0.36	n.s.
Centroptilum luteolum	0.28	0.37
Chloroperla torrentium	-0.32	-0.35
Crangonyx pseudogracilis	0.35	0.35
Dugesia tigrinia	0.44	0.44
Dugesia lugubris/polychroa	0.75	0.67
Ephemera danica	0.38	0.37
Glossosoma boltoni	n.s.	0.26
Helobdella stagnalis	0.61	0.64
Hydroptila sp.	-0.29	-0.39
Hydroporinae	0.29	n.s.
Lepidostoma hirtum	-0.34	-0.46
Leptophlebia vespertina	-0.33	n.s.
Lymnaea palustris	0.26	0.26
Mesophylax impunctatus	0.31	0.31
Mystacides longicornis	0.51	0.43
Ochthebius nanus	0.31	0.31
Oligochaetes	0.27	n.s.
Orthetrum coerulescens	0.32	0.32
Phryganea bipunctata	0.44	0.44
Pisidium/Sphaerium	n.s.	0.30
Planorbis planorbis	0.35	0.35
Planoria torva	0.50	0.54
Sialis lutera	0.46	0.39
Theodoxus fluviatilis	0.40	0.43
Tinodes spp.	0.45	0.48

Indicator species analysis (Dufrene and Legendre, 1997) was used to identify significant indicator species from different lake types. Initially, the analysis was performed using species level data but the resulting indicator values were relatively low (data not shown) and genus level data - *Asellus*, were not reduced to species level - was subsequently used. Where genera were represented by a single species, the species label was retained to preserve as much information as possible. It was not possible to define typing boundaries using the invertebrate community and three alkalinity bands were chosen (< 20, 20 – 100, > 100 mg l^{-1} $CaCO_3$) using expert judgement and taking into account boundaries suggested by the phytoplankton and

macrophyte communities (Chapters 4 and 5, respectively). These alkalinity bands also reflect the natural pattern present in the data. Irish lakes tend to be either soft or hard due to the disproportionate influence of even a small amount of calcareous rock in the catchment. The moderate alkalinity band was represented by only seven lakes.

Indicator species analysis identified 11 statistically significant indicator species from the three lake types (Table 6.17). Types were numbered 1 – 3 for low, moderate and high alkalinity lakes, respectively. The relative frequency and abundance values reflect the strength of association of a particular species with the group for which it is an indicator e.g. 100% of the samples in group 3 contained *Asellus aquaticus* but only 65% of the *A. aquaticus* found were found in group 3. In contrast, all specimens of the trichopteran *Metalype fragilis* encountered were in group 3, but only 20% of the sites in this group contained this species. A species would have an indicator value of 100% if all the individuals of a species were found at all sites of only one group.

Table 6.17 Significant indicator taxa using combined data on *a priori* defined groups (< 20, 20 – 100 and > 100 mg l^{-1} $CaCO_3$), numbered as groups 1, 2 and 3, respectively. 11 of 126 (8.7%) species were significant ($p < 0.05$).

Species	Group	Relative Frequency	Relative Abundance	Indicator Value	p - value
Asellus aquaticus	3	100	65	66	0.005
Bithynia spp.	3	60	77	66	0.005
Caenis spp.	3	100	71	71	0.008
Dugesia spp.	3	70	73	51	0.001
Glossosoma boltoni	3	20	100	20	0.039
Hydracarina	2	100	59	58	0.026
Leptophlebia vespertina	1	63	89	56	0.024
Metalype fragilis	3	20	100	20	0.037
Micronecta poweri	2	71	93	66	0.012
Sialis lutaria	3	30	92	27	0.023
Valvata spp.	2	43	97	41	0.007

Seven significant indicator taxa were identified for group 3, the high alkalinity lakes. The emphemeropterans, *Caenis* spp., had the highest indicator value at 71% followed by the isopod *Asellus aquaticus* and the gastropod *Bithynia* spp., both of which had indicator values of 66%. The flatworm *Dugesia* spp. returned the next highest indicator value (51%). The remaining three significant indicator taxa for the group 3 lakes, the megalopteran *Sialis lutaria* and the trichopterans *Glossosoma boltoni* and *Metalype fragilis* had relatively low indicator values of 26, 20 and 20%, respectively. These three species all had high relative abundance values but because they were not commonly encountered, their relative frequency was low, and consequently, their usefulness as indicator species was reduced. Most of the species identified for group 3 lakes were positively correlated with alkalinity (Table 6.16).

Three significant indicator taxa were identified for the moderate alkalinity group 2 lakes. These were the hemipteran, *Micronecta poweri* (66%), the water mites, Hydracarina (58%), and the gastropod, *Valvata* spp. (41%). *M. poweri* had a high relative abundance value (93%) which contributed to its relatively high indicator value. This large relative abundance value is a result of two sites within this group which each had high numbers (> 1000) of this hemipteran. None of the taxa identified for the group two moderate alkalinity lakes were found to be significantly correlated with alkalinity (Table 6.16). However, there were only seven moderate alkalinity lakes in the dataset.

Only one significant indicator taxa was identified for the low alkalinity group 1 lakes. The ephemeropteran, *Leptophlebia vespertina*, was found in 63% of all low alkalinity lakes and 89% of all individuals encountered were in group 1 lakes, resulting in an indicator value of 56%. This species was negatively correlated with alkalinity (Table 6.16) as were the trichopterans *Lepidostoma hirtum* and *Hydroptilla* spp. and the plecopteran *Chloroperla torrentium*; however these were not identified as significant indicator species.

6.6 Metrics and multimetric indices

6.6.1 Assessment of ecological quality using littoral invertebrates

There is a long tradition of using benthic invertebrates to monitor water quality in lotic systems (e.g. Woodiwiss, 1964; Chandler, 1970; Wright *et al*., 1984; Rosenberg and Resh, 1993) but the use of benthic invertebrate fauna in lentic systems has been confined largely to the profundal zone (e.g. Saether, 1980; Bazzanti and Seminara, 1995; Johnson and Weiderholm, 1989; Lang, 1990) where the habitat is less complex and the communities less diverse. It is only in recent years that attention has turned to the invertebrate communities of lake shores and their potential use in ecological monitoring has been investigated (Raddum *et al*., 1988; Brodersen, *et al*., 1998; Johnson and Sandin, 2001; Tolonen *et al*., 2001; White and Irvine, 2003).

In Ireland, a monitoring programme on acid-sensitive lakes has been in operation since the 1980s. This programme uses a biological index to assess the acid status of surface waters based upon the sensitivity of specific macroinvertebrate fauna to reduced pH (Bowman, 1991; McGarrigle *et al*., 2002). Lakes subjected to eutrophication pressure are currently monitored using a modified version of the OECD scheme which defines trophic categories based upon maximum chlorophyll a levels (McGarrigle *et al*., 2002). There are no biological metrics currently in use in Ireland for lakes subjected to eutrophication pressure, hence there is a need to develop metrics to assess and classify ecological status.

A metric is simply a numerical measure of some aspect of the community. A multimetric is a combination of three or more metrics that allows for the integration of biological information at the individual, population and community level. These are used widely in the United States to assess ecological status. The first multimetric index, the Index of Biotic Integrity, was developed in the 1980s and used fish based metrics to assess the ecology of streams (Karr, 1981). This approach has since been widely adopted and multimetric indices using benthic invertebrates to monitor streams in the United States (e.g. Kerans and Karr, 1994; DeShon, 1995; Barbour *et al*., 1996; Royer *et al*., 2001), New Zealand (Stark, 1993), and The Netherlands (Vlek *et al*., 2004) are a few examples of how successfully this method has been adapted in different regions of the world.

6.6.2 Calculation of metrics

Thirty-five candidate invertebrate metrics were selected from the literature and could be separated into five metric types – taxa richness, diversity, composition, pollution tolerances and functional feeding guilds (Table 6.18). Nine metrics based on taxa richness were calculated including total number of taxa, log (x+1) abundance and a number of other metrics using subsets of the total taxa richness based upon the use of key indicator groups of organisms. Composition metrics using the relative contribution of taxa or groups of taxa to the overall community is based on the premise that stable, healthy communities are relatively consistent in their proportional composition (Barbour *et al*., 1999). Richness and composition metrics are both measures of community structure and have a long tradition for detecting impacts on aquatic systems (Rosenberg and Resh, 1993). Stressed assemblages are considered to contain fewer species and undergo a shift in proportional abundances (Hellawell, 1986).

Nine pollution tolerant metrics, reflecting taxa sensitivity to eutrophication pressures were chosen from the literature. Intolerant species, defined as those which score 10 in the Biological Monitoring Working Party (BMWP) system (Armitage *et al*., 1983), indicate a high sensitivity to eutrophication pressure and their presence indicates good biological condition (Karr and Chu, 1999). The relative abundance of tolerant taxa, viz. oligochaetes and chironomids (the lowest scoring BMWP taxa), is a reliable metric, although the presence of such taxa may reveal little about ecological conditions because of their ability to occur across the trophic gradient. As conditions deteriorate their relative abundance increases (Karr and Chu, 1999).

The BMWP score and associated Average Score Per Taxon (ASPT) are both clean water indices indicating the presence of sensitive and tolerant groups. The ASPT is simply the BMWP score divided by the number of scoring taxa and eliminates effects of disproportionate sampling effort (Armitage *et al*., 1993). Hilsenhoff's Biotic Index (HBI) (Hilsenhoff, 1977), its improved Modified Biotic Index (MBI) using re-evaluated tolerance values (Hilsenhoff, 1987), and the associated Family Biotic Index (FBI) (Hilsenhoff, 1988) were also calculated, as was the Danish Fauna Index (DFI) (SEPA, 2000). All of these metrics have been designed for running water sites and not for invertebrate communities inhabiting the littoral zone of lakes. The Danish Fauna

Index, originally developed for assessing biological condition of running water sites in Denmark (Skriver *et al*., 2000), has been adopted by the Swedish EPA for assessing lakes and watercourses (SEPA, 2000).

Four diversity metrics (Shannon, Simpson, Margalef and Pielou's evenness index) were calculated. Diversity indices exploit species richness and equitability to evaluate impact and have been widely used in Europe despite receiving several critical reviews regarding their usefulness (Washington, 1984; Norris and Georges, 1993). Taxa were also assigned to one of four functional feeding guilds after Tachet *et al*., (2000) and expressed as relative abundance. Feeding guilds act as surrogates of complex biological processes and such metrics have been incorporated in biomonitoring approaches such as the Benthic Index of Biotic Integrity (Kerans and Karr, 1994) and the Florida Stream Condition Index (Barbour *et al*., 1996). More recently though, the use of these metrics has been questioned as their ability to reliably detect change has not been well demonstrated (Barbour *et al*., 1999). They have been found to respond differently across different stream types (Karr, 1999), and fundamental questions remain regarding the assignment of taxa to functional feeding groups (Rawer-Jost *et al*., 2000). Lenant and Resh (2001) stated that species are often assigned to functional feeding guilds based on knowledge that is derived at the genus or family level.

Table 6.18. Thirty five candidate metrics examined and their expected response to eutrophication pressure. ↓ = decrease, ↑ = increase.

Metric	Response	Metric	Response
RICHNESS METRICS		**FUNCTIONAL FEEDING GUILDS**	
Species richness	↓	% Predator	variable
Species abundance	↓	% Shredder	↓
No. EPT species	↓	% Scraper	variable
No. Ephemeroptera species	↓	% Filterer	variable
No. Plecoptera species	↓		
No. Trichoptera species	↓	**TOLERANCE METRICS**	
No. Coleoptera species	↓	No. of intolerant species	↓
No. Trichoptera & Coleoptera	↓	% Oligochaetes	↑
No. Crustacea & Mollusca	↑	% Oligochaetes & Chironomids	↑
		BMWP score	↓
COMPOSITION METRICS		ASPT	↓
% Dominant	↑	Danish Fauna Index	↓
% EPT taxa	↓	Family Biotic Index	↑
% Ephemeroptera	↓	Modified Biotic Index	↑
% Plecoptera	↓	Hilsenhoff Biotic Index	↑
% Trichoptera	↓		
% Coleoptera	↓	**DIVERSITY METRICS**	
% Dipteran	↑	Shannon's diversity	↓
% Crustacea	↑	Simpson's diversity	↓
Gammarus: Asellus	↓	Margalef diversity	↓
		Pielou's evenness	↓

6.6.3 The use of metrics in ecological assessment

Thirty-five metrics (Table 6.18) were calculated using data collected from 149 lakes across the trophic gradient sampled in Spring 2001 and 2002. Samples which contained less than 100 individuals were omitted from the analysis as were samples from three lakes which recorded negative alkalinities. Spearman rank correlation co-efficients (r_S) were calculated to identify metrics which had a linear or log linear response to TP in different alkalinity bands.

In low alkalinity lakes, alkalinity < 10 or < 20 mg l^{-1} $CaCO_3$ (n = 55 and 65 respectively), TP was not significantly correlated with any of the 35 metrics. This was not unexpected as the majority of these lakes had relatively low TP and consequently no trophic gradient existed. Of the 55 lakes with alkalinity values < 10 mg l^{-1} $CaCO_3$, 47 had TP values of 10 µg l^{-1} or less while the remaining lakes had values of between 11 – 20 µg l^{-1}, with only one lake having a TP value > 20 µg l^{-1}.

In lakes with alkalinities >10 and >20 mg l^{-1} $CaCO_3$ (n = 94 and 84 respectively) 18 and 16 of the 35 candidate metrics respectively were significantly associated with TP ($p < 0.05$). Sixteen of these metrics were common to both alkalinity bands and predominantly related to the number or proportion of specific insect taxa (number EPT, number of Plecoptera, number of Trichoptera, number of Trichoptera and Coleoptera, %EPT, %Plecoptera, %Trichoptera, %Coleoptera, %Diptera), the presence of intolerant taxa, the abundance of tolerant taxa and the BMWP, ASPT and DFI scores. In addition, the number of Coleoptera and the FBI metric was significant in lakes with alkalinity >10 mg l^{-1} $CaCO_3$ but not significant in lakes with alkalinity > 20 mg l^{-1} $CaCO_3$.

In lakes with alkalinity values between 20 and 100 mg l^{-1} $CaCO_3$, (n = 48) 11 of the 35 candidate metrics were significant with ASPT returning the highest negative correlation against log TP (r_S = -0.48). In lakes with alkalinity > 100 mg l^{-1} $CaCO_3$, (n = 36) only 7 of the 35 candidate metrics were significant, with ASPT again recording the strongest negative correlation against log TP (r_S = -0.52). The DFI was also strongly negatively correlated with log TP (r_S = -0.47) in this group of lakes. For lakes with alkalinities < 100 mg l^{-1} $CaCO_3$, (n = 113), 18 of the 36 metrics were significant, with ASPT returning the strongest coefficient (r_S = -0.50).

When all of the lakes were included in the analysis (n=149), 20 of the candidate metrics were correlated significantly with log TP. The number of significant metrics increases with increasing number of samples in all of the alkalinity bands examined apart from the low alkalinity lakes where there were no significant correlations because of a lack of a trophic gradient in this group. The metric that consistently performed the best was ASPT and this recorded the highest coefficient value (r_S = -0.50) using the full data set, followed by the number of intolerant taxa (r_S = -0.48). Other significant metrics, related again, to presence or proportion of insects (number of EPT, Plecoptera, Trichoptera, Coleoptera, Trichoptera and Coleoptera, and the percent composition of EPT, Plecoptera, Trichoptera, Oligochaetes, Oligochaetes and chironomids). The HBI, MBI and FBI were also significantly correlated with TP. These three metrics were not consistently correlated across the different alkalinity bands examined, they were not significant in any of the other alkalinity bands examined apart from the FBI which was significant in alkalinity bands > 10 mg l^{-1} $CaCO_3$, and 20 – 100 mg l^{-1} $CaCO_3$. None of the diversity or evenness indices were correlated significantly with log TP in any of the alkalinity bands, nor were taxa richness, percentage scrapers or filterers. Of the remaining functional feeding guilds metrics percentage shredders had a weak negative correlation with TP (r_S = -0.18, $p < 0.05$), using all 149 lakes, while percentage predators was positively correlated with TP but only when lakes with alkalinity > 100 mg l^{-1} $CaCO_3$ were considered. Total abundance, relative abundance or the number of species of Ephemeroptera were also not significant. The significance of the EPT metric seems to be largely the result of the presence of Plecoptera and Trichoptera, which, separately, were significantly and negatively correlated with TP. Ephemeroptera alone were not significantly correlated.

Spearman rank correlation coefficients were generated between the 20 significant metrics to check for redundancy, which was relatively high. In a pair of correlating metrics, the one with the lower correlation coefficient with log TP was eliminated. One pair of metrics, the ASPT and the number of intolerant taxa, were strongly correlated with each other (r_S = 0.80, $p < 0.05$), but were not eliminated. This is the level of correlation above which one metric should be excluded owing to redundancy (CEN, 2004). However, both of these metrics were included as they had the strongest co-efficients with log TP. After eliminating correlated metrics from the 20 significant metrics, 13 remained - the numbers of Plecoptera, Coleoptera, Crustacea and

Mollusca and the number of intolerant species, the percentage Trichoptera, Shredder, Oligochaetes; the biotic indices BMWP, ASPT, DFI, MBI and HBI. Table 6.19 shows the correlations among the 13 remaining metrics. Figure 6.13 shows the relationship between some selected metrics and transformed TP (Log (x+1)).

Table 6.19 Spearman rank correlation coefficients for selected metrics and TP (Log (x+1)). n = 149. Highlighted coefficients are significant at p < 0.05.

	TP (Log (x+1))	No. Plecoptera species	No. Coleoptera species	No. Crustacea + Mollusca	% Trichoptera	% Shredder	No. intolerant taxa	% Oligochaete	BMWP	APST	DFI	MBI
Log (x+1) TP	1.00											
No. Plecoptera	**-0.40**	1.00										
No. Coleoptera	**-0.18**	**0.31**	1.00									
No. Crustacea	**0.32**	**-0.18**	**0.18**	1.00								
% Trichoptera	**-0.29**	**0.21**	0.16	-0.07	1.00							
% Shredder	**-0.18**	**0.18**	**0.25**	0.14	0.13	1.00						
No. intolerant	**-0.48**	**0.64**	**0.40**	-0.03	**0.55**	**0.29**	1.00					
% Oligochaete	**0.23**	-0.20	-0.08	0.02	**-0.23**	**-0.57**	**-0.34**	1.00				
BMWP	**-0.23**	**0.50**	**0.59**	**0.38**	**0.53**	**0.36**	**0.78**	**-0.24**	1.00			
ASPT	**-0.50**	**0.63**	**0.32**	**-0.24**	**0.56**	**0.24**	**0.80**	**-0.34**	**0.64**	1.00		
DFI	**-0.44**	**0.62**	**0.36**	**-0.19**	**0.39**	**0.31**	**0.78**	**-0.36**	**0.60**	**0.72**	1.00	
MBI	**0.32**	**-0.44**	-0.10	**0.28**	**-0.21**	0.00	**-0.39**	**0.54**	**-0.18**	**-0.50**	**-0.41**	1.00
HBI	**0.38**	-0.41	-0.03	**0.37**	**-0.19**	**0.25**	**-0.30**	0.09	-0.06	**-0.45**	**0.32**	**0.77**

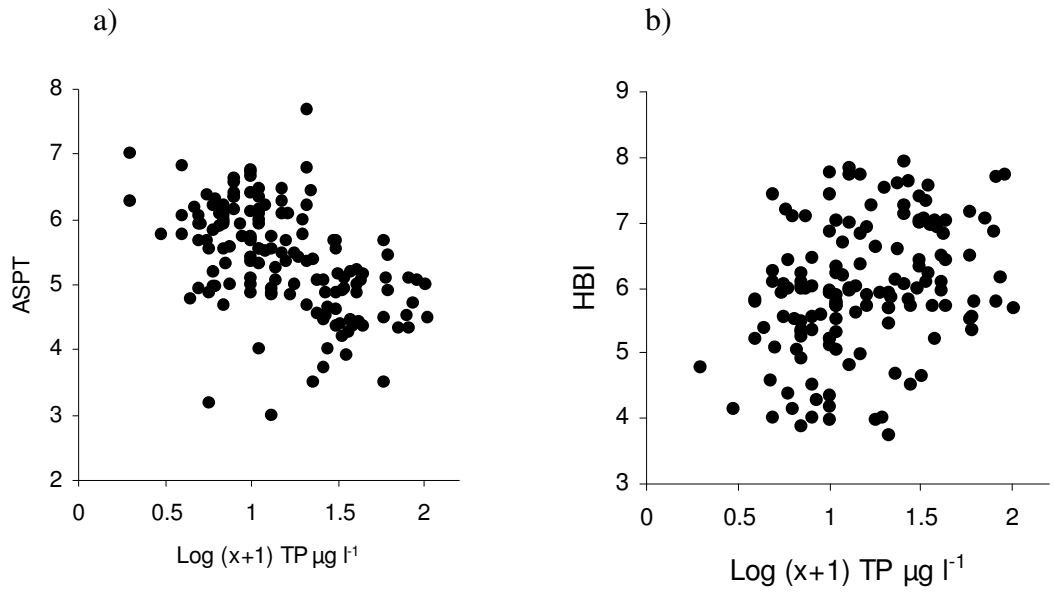


Figure 6.13 Linear relationships found between two selected macroinvertebrate metrics (a) Average Score Per Taxon (ASPT) and (b) Hilsenhoff's Biotic Index (HBI) with (Log (x+1)) transformed TP µg l^{-1} (n = 149).

In addition to the 13 selected metrics, a macroinvertebrate trophic score was developed using data from 190 samples collected in Spring 2001 and 2002. The lakes were divided into six TP bands (<10, 10 – 20, 20 – 30, 30 – 40, 40 – 60, >60 µg l^{-1} TP) and for each invertebrate taxon the average TP value was calculated within each of those bands. To allow for the different sampling intensity among the various TP bands, a weighted average TP concentration for each taxon was calculated based on the percentage of sites where the taxon was present in each TP band. Weighted averages were calculated only for taxa found in five or more samples. Of the 223 taxa encountered, 106 taxa were found in less than five samples and were excluded, leaving 117 taxa remaining. Immature specimens from families or genera currently existing in the dataset were also excluded, resulting in 101 taxa used. Table 6.20 lists the average weighted TP values calculated for these 101 taxa. For each lake the weighted average taxon scores were averaged to give a macroinvertebrate trophic score. The relationship between this macroinvertebrate trophic score and (Log (x+1)) TP for all lakes and for lakes with TP ≤ 15 µg $^{-1}$ and > 15 µg l^{-1} can be seen in Figure 6.14.

Table 6.20: Weighted Spring TP (µg l^{-1}) for 101 taxa from 191 samples. n = taxon frequency of occurrence. Total n = 190.

Taxa	Weighted TP	n	Taxa	Weighted TP	n
Ceraclea nigra	8	23	*Sigara distincta*	34	10
Diura bicaudata	11	10	*Agraylea multipuctuata*	34	12
Haliplus flavicollis	12	6	*Polycentropus kingi*	35	27
Holocentropus dubius	13	6	*Athripsodes cinerus*	35	60
Heptagenia sulphurea	13	32	*Tinodes waeneri*	35	123
Haliplus obliquua	14	9	*Ephemera danica*	35	28
Lype phaeopa	15	5	*Centroptilum luteolum*	36	77
Metalype fragilis	15	8	*Lymnaea peregra*	36	57
Leuctra hippopus	15	37	Oligochaetes	36	188
Nemoura cinera	15	8	*Polycelis nigra/tenius*	36	92
Neureclipsis bimaculata	15	11	*Hydraena gracilis*	36	13
Lymnaea palustris	16	5	*Gammarus* spp.	37	161
Ephemera ignita	16	26	Chirnomids	37	185
Valvata macrostoma	17	8	*Polycentropus flavomaculatus*	37	63
Limnius volckmari	17	59	*Ecnomus tenellus*	37	15
Esolus parallelepipedus	17	62	*Sigara scotti*	37	15
Dina lineata	17	9	*Limnephilus marmoratus*	38	7
Haliplus confinis	19	6	*Caenis luctousa*	38	152
Elmis aenea	19	6	*Hydroptilla* spp.	38	38
Tipulidae larvae	19	31	*Bithynia tentaculata*	38	34
Lepidostoma hirtum	20	57	Sphaeriidae	39	102
Sigara dorsalis	20	10	*Helobdella stagnalis*	39	49
Holocentropus stagnalis	20	7	Hydroacarina	40	137
Chloroperla torrentium	21	55	Ceratopogonidae larvae	40	103
Goera pilosa	21	6	*Limnephilus lunatus*	40	26
Sigara fallenoidea	21	15	Diptera	40	131
Athripsodes arterrimus	21	6	*Valvata cristata*	40	11
Asellus meridanus	21	26	*Potamopyrgus jenkinsi*	41	54
Ceraclea senelis	21	5	*Planorbis crista*	42	6
Caenis rivulorum	22	7	*Planorbis planorbis*	42	13
Mystacides azurea	22	29	*Asellus aquaticus*	43	96
Sericostoma personatum	23	45	*Heptagenia fuscogrisea*	43	9
Electrogenia lateralis	23	27	*Ancylus fluviatilils*	43	32
Mystacides longicornis	26	6	*Arctocorisa germari*	44	5
Limnephilus vittatus	24	6	*Anabola nervosa*	45	7
Segmentina complanata	24	13	*Dugesia lugubris/polychroa*	45	42
Procloeon bifidium	24	6	*Sialis lutaria*	47	11
Leptophlebia vespertina	25	69	*Erpobdella octoculata*	49	26
Cloeon simile	25	11	*Nebrioporus depressus*	49	29
Orectochilus larvae	27	36	*Helophorus brevipalpus*	51	6
Planorbis alba	27	27	*Haliplus ruficollis* group	52	5
Plectrocnemia conspersa	28	34	*Dendrocoelum lacteum*	52	24
Theromyzon tessulatum	28	6	*Caenis horaria*	52	43
Oribatei sp.	30	9	*Velia* spp.	53	5
Oulimnius sp.	30	154	*Cyrinus trimaculatus*	58	15
Micronecta sp.	33	80	*Hygrotus quinquelineatus*	59	6
Baetis sp.	33	9	*Physa fontanalis*	60	10
Glossoma complanata	33	34	*Crangonyx pseudogracilis*	61	11
Theodoxus fluviatilis	33	11	*Sigara falleni*	66	13
Ceraclea fulva	34	6	*Callicorixa praeusta*	70	12
Glossiphonia heteroclita	34	6			

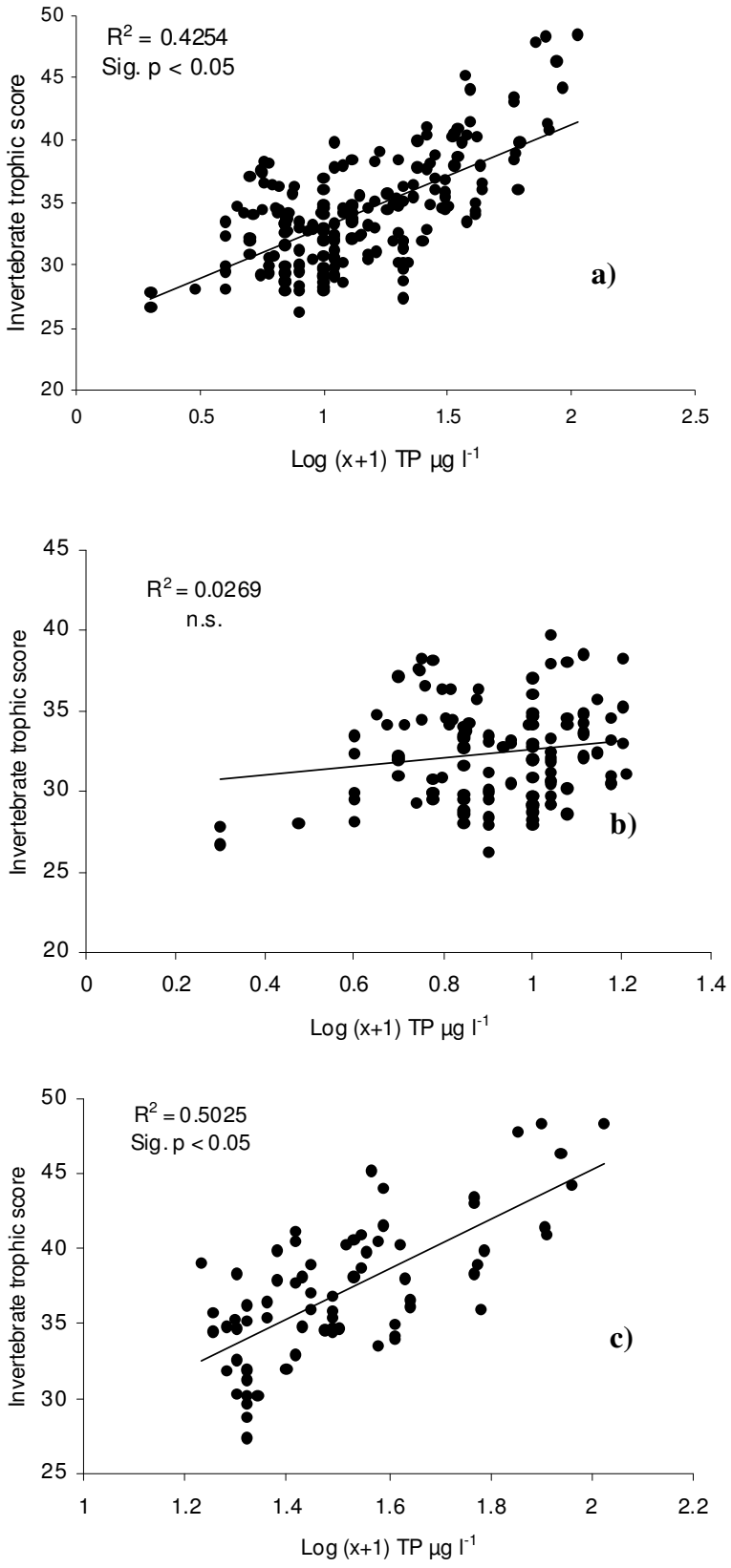


Figure 6.14. Relationship between invertebrate trophic score and transformed TP (Log (x+1)) for: (a) all lakes (n = 191), (b) lakes with TP ≤ 15 µg l^{-1} (n = 119) and (c) lakes with TP > 15 µg l^{-1} (n = 72).

Using all lakes, the invertebrate trophic score was significantly correlated with TP (Log (x+1)) (r^2= 0.43, $p < 0.05$). In lakes with TP values less than 15 µg l^{-1}, there was no significant correlation between the invertebrate trophic score and TP (r^2 = 0.03). Above a TP level of 15 µg l^{-1}, there is a significant correlation between the score and the TP levels in the lake (r^2 = 0.50, $p < 0.05$), suggesting that the invertebrate communities are not influenced by relatively low TP levels. However, once a certain threshold is reached, the community starts to respond and taxa that are more tolerant of the higher nutrient levels begin to increase (Figure 6.14).

The selected metrics and the macroinvertebrate trophic score were scaled and inverted, if necessary, into deciles ranging from 1 (perceived high status) to 0.1 (perceived bad status) in order to place them on a comparable scale (Table 6.21). Several of the metrics were not amenable to scaling due to their short range and, therefore, were not included. These comprised the number of Plecoptera, number of Coleoptera, number of Crustacea and Mollusca and the DFI. The scaled metrics were then averaged to give a macroinvertebrate index. The relationship between this invertebrate index and TP (Figure 6.15) was poor with a r^2 value = 0.22, though the reference lakes did tend to have a higher value of the index. Combining and averaging the metrics did not increase their ability to explain a relationship with total phosphorus. Regression coefficients for each of the individual metrics scaled decile values revealed a weak underlying relationship with the total phosphorus gradient. Correlation coefficients ranged from 0.35 (for the invertebrate trophic index) to 0.01 (% Oligochaeta). Calculating the macroinvertebrate index using different combinations of metrics did not increase the r^2 value above that achieved using the invertebrate trophic index alone. The inclusion of any additional metric to this served only to decrease its r^2 value.

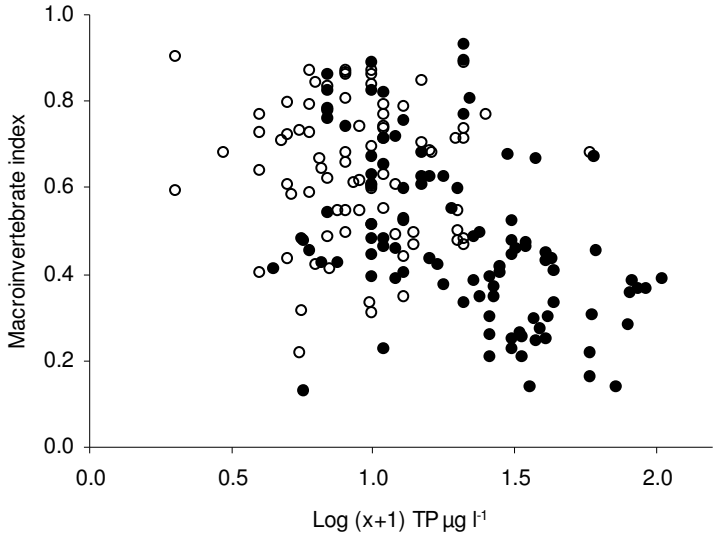


Figure 6.15. Relationship between the macroinvertebrate index and transformed TP (Log (x+1)). ○ = reference samples (n = 80). ● = non reference samples (n = 100)

Table 6.21 Scaled deciles for 10 metrics that had a log-linear response to spring TP.

Scaled Decile	Invertebrate trophic score	ASPT	No. intolerant taxa	% EPT	% Trichoptera	BMWP	HBI	MBI	% Oligochaetes	% Shredder
1.0	< 29.1	< 6.41	≥ 8	> 50	> 30.4	> 123	< 4.22	< 5.33	<1.6	< 81.9
0.9	29.1 – 30.2	6.17 – 6.41	7	45.8 – 50	27.3 – 30.4	114 –123	4.22 - 5.09	5.33 – 5.70	1.6 - 5.3	72.7 – 81.6
0.8	30.2 – 31.6	6.00 – 6.17	6	42.9 – 45.8	24.1 – 27.3	102 – 114	5.09 - 5.42	5.70 – 5.94	5.3 - 8.5	68.7 – 72.7
0.7	31.6 – 32.6	5.69 – 6.00	5	40 – 42.9	22.2 – 24.1	92 – 102	5.42 - 5.70	5.94 – 6.05	8.5 - 10.9	61.9 – 68.7
0.6	32.6 – 34.0	5.50 – 5.69	4	36.4 – 40.0	20.0 – 22.2	83 – 92	5.70 - 5.82	6.05 – 6.21	10.9 - 15.0	53.8 – 61.9
0.5	34.0 – 34.6	5.21 – 5.50	3	33.3 – 36.4	17.6 – 20.0	78 – 83	5.82 - 6.00	6.21 – 6.46	15.0 - 19.3	44.5 – 53.8
0.4	34.6 – 35.9	5.00 – 5.21	2	29.4 – 33.3	14.3 – 17.6	68 – 78	6.00 - 6.23	6.46 – 6.66	19.3 - 24.3	36.5 – 44.5
0.3	35.9 – 37.9	4.86 – 5.00	1	26.1 – 29.4	10.7 – 14.3	58 – 68	6.23 - 6.91	6.66 – 6.92	24.3 – 34.4	25.9 – 36.5
0.2	37.9 – 39.9	4.44 – 4.86	0	20.8 – 26.1	6.3 – 10.7	45 – 58	6.91 - 7.27	6.92 – 7.23	34.4 – 44.5	15.4 – 25.9
0.1	> 39.9	< 4.44	0	< 20.8	< 6.3	< 58	> 7.27	> 7.23	> 44.5	< 15.4

6.6.4 Discussion on metrics and multimetrics

Preliminary investigations into using invertebrate metrics to assess ecological status met with limited but encouraging results. Thirty - five existing metrics were selected from the literature, none of which, apart from the Danish Fauna Index, were designed for use in lentic ecosystems, nor were any designed for use in Ireland. The metric, Average Score Per Taxon (ASPT), consistently performed best. Sandin and Hering (2004) also reported that ASPT was clearly the metric that correlated well with an organic pollution gradient in streams across four European countries (Austria, the Czech Republic, Portugal and Sweden). The ASPT is a modification of the Biological Monitoring Water Party score (BMWP). The BMWP is a simple metric derived using qualitative family level data from lotic sites in the UK (Armitage *et al*., 1983). Families perceived to be pollution intolerant are given a high score and those considered to be pollution tolerant are given a low score. The site score is subsequently divided by the number of scoring families, to suppress the effect of disproportionate sampling effort, to produce the ASPT. If the scores assigned to each family were refined, the sensitivity of the index could be improved to increase its ability to detect disturbance. The refined scores should preferably be generated at genus or species level, as individual species have distinct pollution tolerances, e.g. Resh and Unzicker (1975) demonstrated that in 89 genera for which pollution tolerance values had been established for more than one species, the component species had different tolerance levels. The availability of the recently developed AQEM software (AQEM Version 2.3.4a, 2004) which generates values over 100 different metrics, should also allow for the selection of multi-metric indices to assess the impact of anthropogenic pressures (AQEM, 2002).

The invertebrate trophic score, developed using species weighted averaging TP concentrations, returned an r^2 value = 0.50 from lakes with TP concentrations > 15 µg l^{-1}; significant at $p < 0.05$, yet indicated no significant relationship at concentrations below this level (r^2 = 0.02). This suggests a certain threshold below which the community remains unimpacted but above which, taxa more tolerant of higher nutrient levels, begin to increase. The reliability of this metric needs to be tested using independent samples and the inclusion of additional samples would improve the values of each taxa's trophic ranking score.

The WFD requires the separation of lakes into certain types to ensure appropriate comparisons are made when assessing the effect of pressures. The low alkalinity lakes in this study had relatively low TP concentrations and thus no trophic gradient existed, consequently, it was not possible to examine type specific metrics for this lake type. Additionally, there were relatively few moderate alkalinity lakes in reference condition. Future work, to fill in these gaps, should allow for the development of a robust system of metrics for each lake type in order to reliably assess ecological status.

7. Profundal Macroinvertebrates

7.1 Introduction

The (WFD) Water Framework Directive (CEC, 2000) requires that lakes be differentiated by typology using physical and chemical factors. For each of the lake types identified, reference conditions must be determined for the biological communities, in this instance, the profundal macroinvertebrates. There are two approaches that can be taken to define typologies for lakes. Firstly, System A of the WFD can be adopted where types are, in a sense, arbitrarily defined categories of preset hydro-morphological factors and the community composition of these is then assessed. Alternatively, System B, which is based on both obligatory and optional factors to partition lakes with recognisably similar faunal communities, may be used.

7.1.1 The influence of eutrophication on profundal invertebrates

Eutrophication or nutrient enrichment is caused by inputs of phosphates and to a lesser extent nitrates resulting in increased primary production. With progressive eutrophication, oxygen may become depleted or absent (anoxia) at the sediment interface and often for a distance above in the water column. This is caused by the decomposition of the algae. The influence of eutrophication on profundal invertebrates has been well studied but has mainly focussed on either of two components, the oligochaetes (worms) or the chironomids (midge larvae). The two components are believed to respond at a different pace to changes in trophic status. The oligochaete community maintains its structure for a longer period i.e. it is slower to respond.

With progressive eutrophication, oligotrophic oligochaete taxa decline and eventually disappear and eutrophic tolerant taxa increase in abundance often resulting in monocultures. This is because different taxa have different oxygen requirements. Oligochaetes, in particular *Limnodrilus* spp. and *Tubifex* spp., are reasonably tolerant of anoxic conditions and can survive for long periods. However, their growth usually declines and maturation and reproduction is slowed or ceases. The chironomid community responds in a similar manner. Eutrophic tolerant taxa such as *Chironomus* spp. increase in abundance and may dominate. Oligotrophic taxa such as *Protanypus* spp. decline and eventually disappear. Again, the response is linked to oxygen requirements.

However, the effects of eutrophication on the profundal community may be compounded by thermal stratification. A thermally stratified lake exhibits differences in temperature between the upper and lower water column, resulting in layers of water differentiated by temperature and density. The layer of water above the sediments (hypolimnion) with the lowest temperature and consequently the greatest density is cut off from the upper layer (epilimnion) by the intervening layer (metalimnion) where the greatest change in temperature occurs (thermocline). With prolonged stable stratification, the hypolimnion can become anoxic because there is no exchange of oxygen either with the atmosphere or with the other lake layers. This is usually a summer phenomenon and varies in duration. Not all lakes stratify because its occurrence is dependent on factors such as lake morphology (depth, shape), climatic conditions, presence of inflows and outflows and geographical location. The extent of deoxygenation is dependent on productivity, lake shape, dissolved organic content and hypolimnetic temperature (Nürnberg, 1995). Most Irish lakes do not stratify in the classical sense i.e. for more than 2 months. However, many lakes undergo short periods of stratification which may influence community composition.

7.1.2 Assessment of ecological quality using profundal macroinvertebrates

Four approaches were adopted in developing an ecological assessment system for eutrophication. Firstly, the oligochaete and chironomid components were classified separately to identify trophic status related assemblages and to assist in the development of appropriate metrics. It has been shown in the literature for both the oligochaete (Milbrink *et al.*, 2002; Särkka & Aho, 1980) and chironomid components (Saether, 1979; Lang, 1984) that assemblages exist representing different trophic states. Reference lakes would be expected to form separate groups from eutrophic lakes based on their profundal invertebrate communities.

For the second approach, a number of indices in the literature were selected for investigation and applied to the data collected in this study. Trophic status indices have been developed for lakes for both components (Milbrink, 1980; Wiederholm, 1980; Howmiller and Scott, 1977). None have been applied beyond the studies within which they were developed with perhaps the exception of the Swedish Environmental Agency who utilise the biological quality index based on chironomids and the o/c index (oligochaetes/chironomids), developed by Wiederholm (1980). These indices were originally developed from Swedish lakes.

The third approach taken involved developing a multimetric system based on the draft CEN (2004) standard as per Chapter 5 and the alternative approach using multiple regression. Lastly, an index of Biological Integrity was developed based on Karr and Chu (1999). This method does not depend on linear metrics nor does it recommend over-reliance on statistical tests of significance. The emphasis is placed on recognising patterns of change. At points of change, scores are assigned which represent minimally (5) moderately (3) and disturbed (1) conditions. A multi-metric score is the summation of these scores for the selected indices. In the case of reference sites, this should result in their clear separation from disturbed conditions.

7.2 Methods

7.2.1 Sampling method

The methods used for sampling profundal invertebrates are outlined in Chapter 3.

7.2.2 Statistical analysis

Specimen counts were averaged for each lake and expressed as numbers per m^2 with taxonomic identification standardised. Only taxa with an occurrence in 5 or more lakes were included in the analyses. Data from candidate reference lakes (n=63) was classified using TWINSPAN. Oligochaeta (n=166) and chironomid data (n=93, only lakes with an alkalinity >20 mg l^{-1} $CaCO_3$ had an adequate trophic - TP - gradient) were classified separately. Five pseudospecies cut levels (Appendix 6) were selected to reflect abundance distributions. The analysis was terminated after 6 division levels. The resulting TWINSPAN groups at each division level were validated using a multi-response permutation procedure (MRPP or ANOSIM) with rank transformed Sorensen (Bray-Curtis) distance measure. Data was log (x+1) transformed. Pair-wise comparisons and comparison of endgroups with the same parent group were made as far as was practicable. Indicator species analysis was used to confirm TWINSPAN selected indicator species and identify additional indicator species for the selected end groups.

The environmental dataset consisted of 19 variables (Appendix 6). These were appropriately transformed (log, log (x+2) or square root). Land-use data was not normally distributed even when transformed and was not available for one lake (Ballynakill, Gorumna) which was omitted from analysis that involved those variables. A second dataset consisting of 10 variables was also used omitting landuse data, chl *a*, TP and Secchi depth. The relationship

between the environmental variables (both datasets) and the profundal community was investigated using Canonical Correspondence Analysis (CCA). The significance of the first axis for all CCA was tested using Monte Carlo permutation tests. Canonical Variates Analysis (CVA) was used to identify the environmental variables that best discriminated between TWINSPAN endgroups and these were subsequently tested for statistical significance using Mann-Whitney tests.

A similar analysis procedure was adopted for investigating the relationships between TP and chironomid community structure and TP and oligochaete community structure. A summary of the datasets used is given in Appendices 5 and 6.

An additional three variables: compactness coefficient of catchment (CCC) (Gravelius cited in Wisler and Brater, 1959), crenulation of catchment perimeter (CCP) (Miller, 1953, cited in Mitchell and McDonald, 1995) and index of lake basin shape (ILBS) following Nürnberg (1995) were used to investigate the relationship between chironomid community structure and TP. Mean depth (y) was estimated from station depth (X) using the equation 7.1 derived from Irvine *et al*. (2001):

$$y=0.314X \qquad \textbf{Equation 7.1}$$

Compactness of coefficient of catchment (CCC), CCP and ILBS were calculated according to Equations 7.2, 7.3 and 7.4 respectively:

$$CCC = \frac{0.28(\text{catchment perimeter (km)})}{(\text{Catchment area (km}^2))^{0.5}} \qquad \textbf{Equation 7.2}$$

$$CCP = \frac{\text{catchment perimeter (km)}}{\text{Catchment area (km}^2)} \qquad \textbf{Equation7.3}$$

$$ILBS = \text{mean depth (m):lake area (km}^2) \qquad \textbf{Equation 7.4}$$

Because stratification would be expected to influence chironomid community structure, a measure (PS) of the potential number of times a lake would be expected to stratify over the summer was predicted from data in the ecological assessment of lakes report (Irvine *et al.*,

2001). The estimate was based on a prediction of the total number of times stratification would have been encountered while sampling in the months of April, June, July and September in 1996 and 1997. The prediction is based on the easting co-ordinate and transformed (log (x+1)) mean depth (m) /Square root of lake area (km^2) (r^2 = 0.73, n = 28). The numbers of sampling occasions predicted to co-incide with stratification are interpreted as follows:

> 5 = Strong stable stratification over the summer months (e.g. Dan and Bray)

5-3 = Stratified for one or more months of the summer

3-1 = Likely to stratify occasionally

<1 = Unlikely to stratify for long periods

7.2.3 Calculation of metrics

The following oligochaete indices were selected for investigation:

- Environmental Index (EI) based on relative abundance of oligochaetes developed by Howmiller and Scott (1977);
- a modified version of the Environmental Index (mEI) (Milbrink, 1980) and
- the Benthic Quality Index using oligochaetes (Wiederholm, 1980).

The taxa used in the indices are presented in Table 7.1. The indices were calculated as follows:

Wiederholm (1980): $BQI=\frac{\sum n_i k_i}{N}$, **Equation 7.5**

where n_i is the number of individuals for the respective indicator species and N is the total abundance of indicator species and k_i is a constant given to each species (Table 7.1).

Howmiller & Scott (1977): Index Value = $\frac{\sum n_1+2\sum n_2}{\sum n_0+\sum n_1+\sum n_2}$ **Equation 7.6**

Milbrink (1980): Index Value = $c\,\frac{0.5\sum n_0+\sum n_1+2\sum n_2+3\sum n_3}{\sum n_0+\sum n_1+\sum n_2+\sum n_3}$ **Equation 7.7**

where c is a coefficient determined by the total abundance of oligochaetes (Table 7.2) and $\sum n_0 \ldots \sum n_3$ is the total number of oligochaetes belonging to Group 0….3 (Table 7.1) .

Table 7.1 A list of the oligochaete taxa encountered in this study with an indication of their perceived trophic tolerance and use in the respective indices from the literature.

Taxa used in indices or recorded in present study	Howmiller & Scott (1977) EI	Milbrink (1980) mEI	Wiederholm (1980) BQI ki values
Limnodrilus group	Group 2 - eutrophic	Group 3 - eutrophic	1
Aulodrilus pluriseta	Group 1 - mesotrophic	Group 2 - eutrophic	
Potamothrix hammoniensis		Group 2 - eutrophic	2
Ttubifex tubifex	Group 2 - eutrophic	Group 3 - eutrophic	
Spirosperma ferox	Group 1 - mesotrophic	Group 0 - oligotrophic	3
Stylodrilus heringinaus	Group 0 - oligotrophic	Group 0 - oligotrophic	4
Dero digitata	Group 1 - mesotrophic		
Slavina appendiculata	Group 1 - mesotrophic		
Scheme range	0 - 2	0 - 3	1 - 4
Olgiotrophic	near 0	</=0.6	4
Mesotrophic	no clear definition	0.6 - </=1	
Eutrophic	near 2	1- 3	1

Table 7.2 Coefficient c values determined by oligochaete abundance.

Value of Coefficient c	Total abundance of oligochaetes (m^{-2})
1	>3000
0.75	1200 <n<3000
0.5	400 <n<1200
0.25	160 <n<400
0	<n<160

Lumbriculus variegatus was frequently encountered in this study but it was not utilised by any of the aforementioned indices. Three indices; Index 3 to 5 were devised based on the mEI. Taxa were reassigned (Table 7.3) and coefficient values altered (Table 7.4).

Table 7.3 Reassignment of trophic preference of oligochaete taxa and inclusion of additional taxa for calculating Indices 3 to 5 (IV 3 to IV5) based on Milbrink (1980).

Taxa used in indices:	Assignment:	Preference
Limnodrilus group	Group 3	eutrophic
Aulodrilus pluriseta	Group 1	oligotrophic
Potamothrix-tubifex group	Group 2	eutrophic
Tubifex tubifex	Group 0	oligotrophic
P. bavaricus	Group 1	oligotrophic
Psammoryctides barbatus	Group 1	oligotrophic
Spirosperma ferox	Group 0	oligotrophic
Lumbriculus variegatus	Group 0	oligotrophic
Stylodrilus heringinaus	Group 0	oligotrophic
Dero digitata	Group 1	oligotrophic
Vejdovskyella comata	Group 1	oligotrophic
Naididae	Group 1	oligotrophic
Slavina appendiculata	Group 1	oligotrophic
Enchytraeidae	Group 1	oligotrophic

Table 7.4 Alterations to coefficient value for Index 3, 4 and 5.

Value of Coefficient c	Index 3	Index 4	Index 5
1	>3000	>1500	>750
0.75	1200 <n<3000	1500 <n<1000	750 <n<500
0.5	400 <n<1200	1000<n<500	500 <n<250
0.25	160 <n<400	500<n<250	250<n<50
0.1	n<160	n<250	n<50

Saether's key to chironomid associations of the profundal zone (Saether, 1979) and the BQI for chironomids (Wiederholm, 1980) were applied. The BQI and modifications thereof (Table 7.5) were calculated as follows:

Wiederholm (1980): $BQI=\frac{\sum n_i k_i}{N}$, **Equation 7.8**

n_i is the number of individuals for the respective indicator species and N is the total abundance of indicator species and k_i is a constant given to each species (Table 7.5). Index values range from 1 to 3. Values close to 1 are indicative of oligotrophic status and values close to 3 are indicative of eutrophication.

The Quirke Index (Irvine *et al.*, 2001) was also calculated:

Quirke Index 1

=Log abundance (*Limnodrilus* group + *Potamothrix-tubifex* group+0.1 *Chironomus* spp.)

Equation 7.9

Quirke Index 2

= Log abundance (Limnodrilus group + *Potamothrix-tubifex* group+0.1 *Chironomus* spp.+ *Procladius* spp.) **Equation 7.10**

A number of metrics for the multimetric index were calculated (Table 7.6). Metrics with log-linear relationship with TP were identified. These metrics were rescaled from 0 (poor status) to 1 (high status) and averaged to develop a profundal index. In the linear multiple regression method, the metric with the highest correlation was added to a linear regression model. Additional metrics were only included if an additional 5% of the variation was explained.

Table 7.5 The value constants (k_i) assigned to each taxa used in calculating the original Biological Quality Indices (BQI) based on chironomids and 4 further modifications.

	k_i				
taxa	BQI	BQI 1	BQI 2	BQI 3	BQI 4
Chironomus spp.	1	1	1	1	1
Cladotanytarsus sp.		5	5	4	5
Cryptochironomus sp.		1	2	1	1
Dicrotendipes spp.		4	4	1	4
Heterotanytarsus spp.				4	
Heterotrissocladius sp.	5	5	5	2	5
Micropsectra sp.	4	4	4	2	4
Microtendipes spp.		3	3	4	3
Monodiamesa sp.				3	
Pagastiella spp.		5	5	4	5
Paracladopelma sp.	4	4	4	2	4
Pentaneurini		5	5	2	5
Phaenopsectra sp.	3	3	3	4	3
Polypedilum spp.		5	4	4	5
Procladius spp.		3	2	4	3
Protanypus sp.				3	5
Sergentia spp.		5	5	3	5
Stictochironomus sp.	3	5	5	3	5
Tanytarsus spp.		3	2	4	3

Table 7.6 The metrics examined with some abbreviations and estimated response (R) to eutrophication. Metrics used in either the Profundal or the Karr method indices are in bold. Legend: l=*Limnodrilus* group, p=*Potamothrix-tubifex* group, s=*Spirosperma ferox*, v=*Lumbriculus variegatus*, C=*Chironomus* spp., P=*Procladius* spp., T=*Tanytarsus*, *Cryptochironomus* spp. =cC, *Microchironomus* spp. =mC, ch=chironomids and ol=oligochaetes. ↑ = increases with pressure, ↓ = decreases with pressure, ∩= unimodal response to pressure. H', H'(loge), H'(log10) = Shannon diversity, LdRich =log Species Richness (Margalef), J'= Pielou's eveness, LFsr=Log Fisher's statistic, 1-Lambda'= Simpson's index.

Metric	**R**	**Metric**	**R**
Relative abundance expressed as:		Richness:	
% of total abundance		*Number of:*	
Individual taxon	↑/↓/∩	chironomid taxa	↓
C +P+T	↑	oligochaete taxa	↓
C +P+T+L+P	↑	Ch taxa *excluding (*C+P+T+ cC).* **(no. of chironomids taxa *)**	↓
L+P **lp**	↑	littoral chironomid taxa	∩
Ephemeroptera+Trichoptera+Plecoptera	↓	*Diversity Measures:*	
ch-(*C +P+T+ cC)* **(Other chironomids**	↓	Ldrich	∩
s+v	↓	J'	∩
Others (excluding chironomids+olgiochaetes)	↓	LFsr	∩
C+L+P **(% Eutr sp.)**	↑	H'(loge)	∩
as a proportion of oligochaetes:		H'(log10)	∩
L+p	↑	1-Lambda'	∩
s+v	↓	**Abundance (m^2):**	
Aulodrilus spp.	↓	individual taxon	↓/↑/∩
as a proportion of chironomids:		*l+p*	↑
C	↑	*s+v*	↓
C+P+T	↑	*l+p+ C*	↑
C+P+T	↑	*l+p+ C+ P*	↑
Ch-(*C +P+T) (*ch-CPT)	↓	other chiron	↓
as a proportion of oligochaetes + chironomids:		**Depth adjusted ratios:**	
l	↑	(ol/D)/((0l/D)+c)	↑
p	↑	(lp/sv)/D	↑
Ch- (C +cC+mC+ P+T) **(ch-cPT)**	↑	*lp*/depth **(Dlp)**	↑
P	↑	(o/c)/D	↑
T	↑	(ol/D)/chir+1	↑
l+p+ C	↑	lp/D+c+p	↑
l+p+ C+cC+mC) **(Eutrophic taxa dominance**	↑	(lp/others)/D	↑
C	↑	(lp+c)/D	↑
Ratios:		(lp+c+P)/D	↑
(*l+p*)/ (*l+p*+chironominae) **(o/c)**	↑	(D(lp/D)/((lp/D)+sv)+(ch/ Ch taxa	↑
(*l+p*) /*C*			
(*l+p*) /(*C*+1)			
(*l+p*)/ (*s+v*) **(lp/sv)**			
(C +P+t+ cC)/(chir –same) **ratio ccrtp:others**			

*excluding (*C+P+T+ cC*)

7.3 Results

7.3.1 Evaluation of sampling method

Due to time constraints, the evaluation of the sampling methods did not include all the lakes samples but a random selection of lakes.

Accuracy of mean

It was evident that five replicate samples generally provided a good estimate of abundance (Figure 7.1) and an adequate representation of taxa richness with the exception of Cloongat, a shallow low alkalinity lake (Figure 7.2). A second order jack-knife estimate was calculated (Table 7.7). This method showed that the number of taxa found was close to the actual number estimated to be present with the exception of Cloongat. Cloongat required more samples but was exceptional in having such high taxa richness (Figure 7.3).

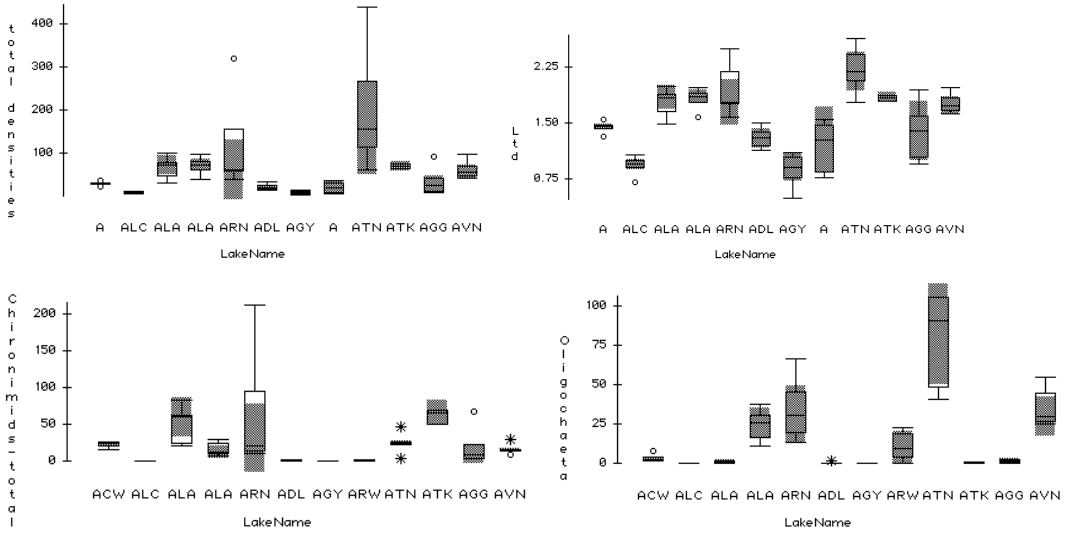


Figure 7.1 Boxplots of total abundance, log transformed total abundance (Ltd), chironomid (chironomids-total) and oligochaeta abundance (m^{-2}). Abbreviations in sequence: Acrow-ACW, Aleckmore-ALC, Alewinghta-ALA, Alua-ALA, Annaghmore Rn-ARN, Anure Dl-ADL, Ardderry Gy-AGY, Arrow-ARW, Ateduan-ATN, Atorick-ATK, Aughrusbeg-AGG and Avaghon-AVN.

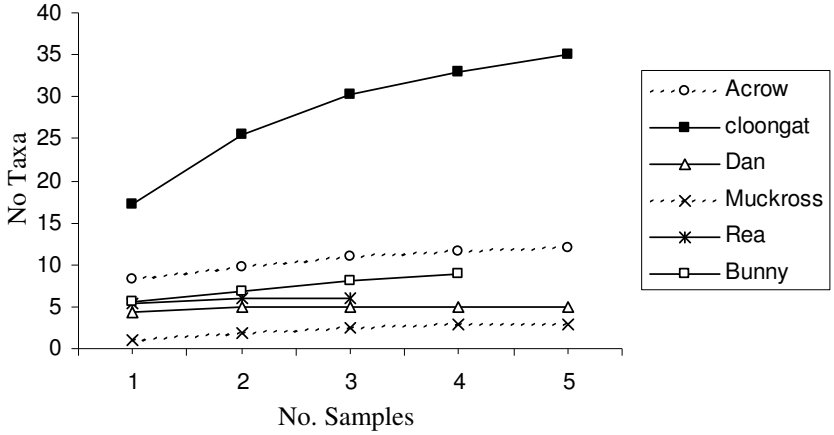


Figure 7.2 Species area curves for lakes representing a cross section of lakes.

Table 7.7 Jack-knife estimate of number of taxa in six lakes.

Lake	n species	1st order jack knife	2nd order jack knife
Acrow	12	13.6	13.4
Cloongat	35	43	45.4
Dan	5	5	5
Muckross	3	3.8	3.4
Rea	6	6	5.7
Bunny	9	12	13.7

7.3.2 General description of profundal macroinvertebrates for candidate reference lakes

Seventy-two taxa were identified from the 63 reference lakes investigated, comprising 31 chironomid taxa, predominantly identified to genus level, 10 oligochaete taxa (some identified to species level) and 31 additional taxa from other groups at various levels of identification. Fifty taxa occurred in ten or less lakes. Seven taxa occurred in thirty or more lakes. The number of taxa recorded per lake ranged from 1 to 35 with a median of 9 (Figure 7.3). The average number of taxa per lake was 10. Chironomids and oligochaetes were the most frequent (Figure 7.4) and abundant main groups (Figure 7.5). One lake, Ardderry, had no chironomids. Two lakes, Guitane and Nalughraman (DL) had no Chironominae. Oligochaeta were absent from seven lakes: Ardderry, Craghy, Dunglow, Guitane, Kindrum, Nalughraman and Waskel. Tubificidae were absent from Dan, Fad (west), Fee, Nakirka, Salt and Tay. With the exception of chironomid and oligochaete taxa, no taxa belonging to other groups were found in Barra, Currane, Muckross, Nalughraman and Rea.

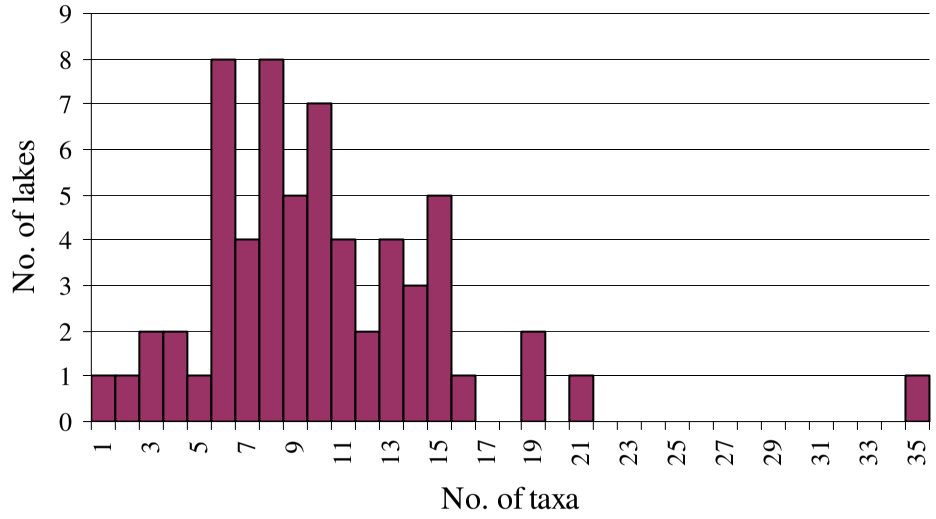


Figure 7.3 The number of lakes with a specified number of taxa.

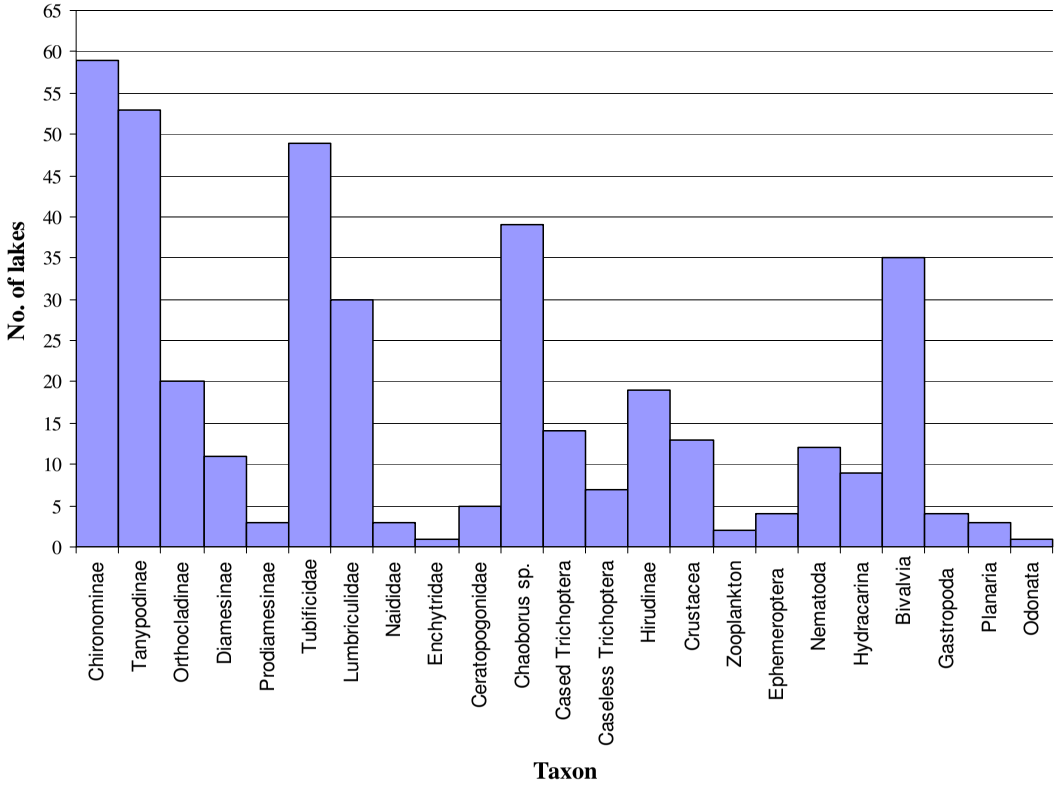


Figure 7.4 The frequency of occurrence for the taxa specified in 63 lakes.

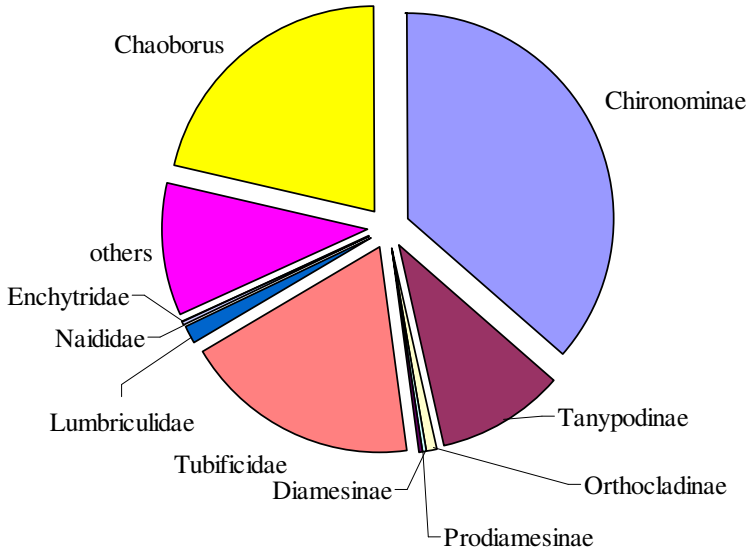


Figure 7.5 The relative abundance of taxa which contributed >5% to the total abundance.

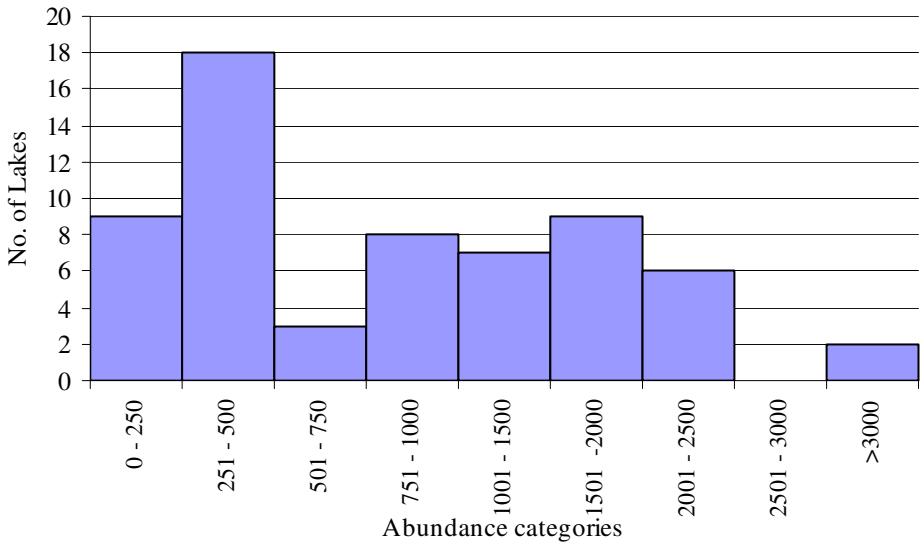


Figure 7.6 The number of lakes within a specified abundance category (m^{-2}).

Abundance of organisms ranged from 9 m^{-2} to 5644 m^{-2} with an average of 1051 m^{-2} and a median of 764 m^{-2}. Figure 7.6 gives an overall impression of the abundance levels encountered.

7.3.3 Typology based on classification of profundal macroinvertebrate data

The TWINSPAN classification resulted in the production of three endgroups at division level II (Figure 7.7). Multi-response permutation procedure (MRPP) showed that the 3 groups: Groups 1, 2 and 3, were significantly different (Table 7.8). Indicator Species Analysis identified 19 significant indicator taxa (Table 7.9). These included 9 of the 11 TWINSPAN indicator species selected at Division Levels I and II inclusive. Six indicator species were identified for Group 1 and 13 for Group 3. No indicator species were identified for Group 2. The DCA ordination supported the TWINSPAN classification (Figure 7.8). The 3 TWINSPAN groups were reasonably well separated. The length of the first DCA axis was 2.903. Therefore, CCA was appropriate to use to determine the underlying environmental variables influencing species composition (Figure 7.9).

Forward selection in CCA selected nine environmental variables from dataset 1 which explained most of the variance in the biological community - alkalinity, maximum depth, TP, peat, colour, catchment area, catchment perimeter, other catchment use and chlorophyll *a*. (Table 7.10). The resulting ordination showed a reasonable separation of the TWINSPAN groups (Figure 7.10) without a serious loss of information as indicated by the comparable eigenvalues (Table 7.11). The variables TP and chl *a* had short gradients indicating that they had little influence on the faunal data. This confirmed that the majority of the lakes were in reference condition. Alkalinity, maximum depth, colour and altitude were selected from dataset 2 as explaining most of the variance (Table 7.10).

Six variables from dataset 1: %Peat, maximum depth, %forest, lake area, TP and lake perimeter were identified by CVA as the variables important in discriminating between TWINSPAN endgroups (Table 7.12). The three groups were clearly separated (Figure 7.11) on the CVA ordination diagram. Six variables; conductivity, maximum depth, colour, lake perimeter, altitude and lake area were identified by CVA from dataset 2 as being significant. The findings of the CCA and CVA were also supported by Mann-Whitney tests (Table 7.13).

Indicator species identified by Indicator Species Analysis occupied the same environmental space as the groups they typified (Figure 7.12) e.g. the indicator taxa;

Chironomus sp., *Limnodrilus* group and *Potamothrix-Tubifex* group for Group 1, were located at the higher end of the alkalinity gradient. The indicator taxa; *Pagastiella* sp., Pentaneurini and *Polypedilum* sp. for Group 3 were located at the lower end of the depth gradient indicating their preference for shallow depth. Likewise, *Chaoborus* sp., the TWINSPAN indicator for Group 2 was located at the higher end of the depth gradient indicating its preference for deeper water. Significant differences in indicator species abundances were also found (Table 7.14).

The analysis of the profundal macroinvertebrates for 62 reference candidate lakes identified 3 distinct lake groups that could be biologically and environmentally characterised (Table 7.15). Group 1 consisted of clear, high alkalinity, deep lakes surrounded by pastures with a biological community predominantly consisting of *Chironomus* sp, *Limnodrilus* group, *Potamothrix-Tubifex* group, *Procladius* sp., *Tanytarsus* sp., *Microtendipes* sp. and cased Trichoptera. Group 2 consisted of coloured, low alkalinity, deep lakes surrounded by peatlands. No biological community could be described for this group. It had no significant indicator taxa with the exception of the TWINSPAN indicator *Chaoborus* sp. Group 3 consisted of coloured low alkalinity, shallow lakes surrounded by peatlands with a biological community predominantly consisting of littoral chironomids such as *Pagastiella* sp., Pentaneurini, *Polypedilum* sp., *Dicrotendipe*s spp. and chironomids characteristic of oligotrophic lakes e.g. *Demicryptochironomus* spp.

Alkalinity or a correlated variable (Table 7.16) and maximum depth were consistently selected throughout the analysis as the main variables responsible for the separation of the three TWINSPAN groups.

Table 7.8 Results of MRPP for Groups 1, 2 and 3. A = Chance-corrected within-group agreement.

	3 Groups		Pairwise Comparisons:		
		Average distance	Groups	p	A values
No. of groups:	3	Grp 1=0.123	1,2	<0.005	0.155
A	0.31	Grp 2 =0.476	1,3	<0.005	0.361
Significance	<0.005	Grp 3=0.306	2,3	<0.005	0.185

Table 7.9 Output from Indicator Species Analysis. Indicator species selected by TWINSPAN (T) up to and including Division level II are indicated with significant ($p<0.05$) indicator taxa highlighted in bold.

		Relative Abundance:			Relative Frequency (%):					Observed	
Group	Maxgrp	1	2	3	Maxgrp	1	2	3	Maxgrp	Indicator	
n		12	32	18						Value (IV)	p *
Chironomus spp.	1	92	5	2	1	100	34	33	**1, T**	**92.3**	**0.001**
Cladopelma spp.	3	37	16	47	3	8	9	11	3	5.2	0.844
Cladotanytarsus sp.	3	18	5	76	3	8	9	28	3	21.2	0.101
Demicryptochironomus spp.	3	0	0	100	3	0	0	28	**3**	**27.8**	**0.004**
Dicrotendipe s spp	3	0	4	96	3	0	6	56	**3**	**53.5**	**0.001**
Microtendipes spp.	1	93	3	5	1	33	6	11	**1**	**30.9**	**0.017**
Pagastiella spp.	3	1	1	98	3	17	16	89	**3**	**86.9**	**0.001**
Polypedilum spp	3	3	8	89	3	17	41	83	**3, T**	**74.1**	**0.001**
Pseudochirnomus spp.	3	0	0	100	3	0	0	33	**3**	**33.3**	**0.001**
Sergentia spp.	3	0	53	47	3	0	31	50	3	23.3	0.278
Stempellinella spp.	3	0	4	96	3	0	6	22	**3**	**21.3**	**0.019**
Tanytarsus spp.	3	22	9	69	3	67	66	83	**3, T**	**57.3**	**0.007**
Procladius spp.	1	46	10	44	1	100	69	89	**1, T**	**46.4**	**0.049**
Pentaneurini	3	9	15	75	3	8	19	67	**3, T**	**50.3**	**0.001**
Heterotanytarsus spp.	3	8	8	84	3	8	13	44	**3, T**	**37.4**	**0.01**
Protanypus spp.	3	0	49	51	3	0	16	33	3	17	0.194
Limnodrilus group	1	84	12	4	1	100	53	50	**1, T**	**83.6**	**0.001**
Aulodrilus pluriseta	1	23	5	72	1	25	9	17	3	12.1	0.593
Potamothrix-tubifex group	1	87	9	4	1	75	41	28	**1, T**	**65.5**	**0.001**
Spirosperma ferox	3	0	30	70	3	0	28	44	**3**	**30.9**	**0.027**
Lumbriculidae	3	7	38	55	3	25	47	72	**3**	**39.8**	**0.031**
Ceratopogonidae	3	0	5	95	3	0	3	28	**3**	**26.5**	**0.009**
Chaoborus sp.	1	57	41	2	2	75	78	28	1, T	42.9	0.085
Cased Trichoptera	1	81	3	16	1	50	6	39	**1, T**	**40.7**	**0.007**
Caseless Trichoptera	1	49	18	33	1	25	6	11	1	12.2	0.244
Hirudina	2	28	42	29	2	17	34	28	2	14.6	0.684
Asellus spp.	1	67	26	7	1	33	16	11	1	22.5	0.078
Gammarus spp.	3	0	0	100	3	0	0	11	3	11.1	0.106
Nematode	1	45	36	19	1	25	19	17	1	11.3	0.643
Hydracarina	3	20	4	76	3	17	3	33	**3**	**25.4**	**0.02**
Bivalves	3	15	29	56	2	25	66	61	3, T	34.4	0.247

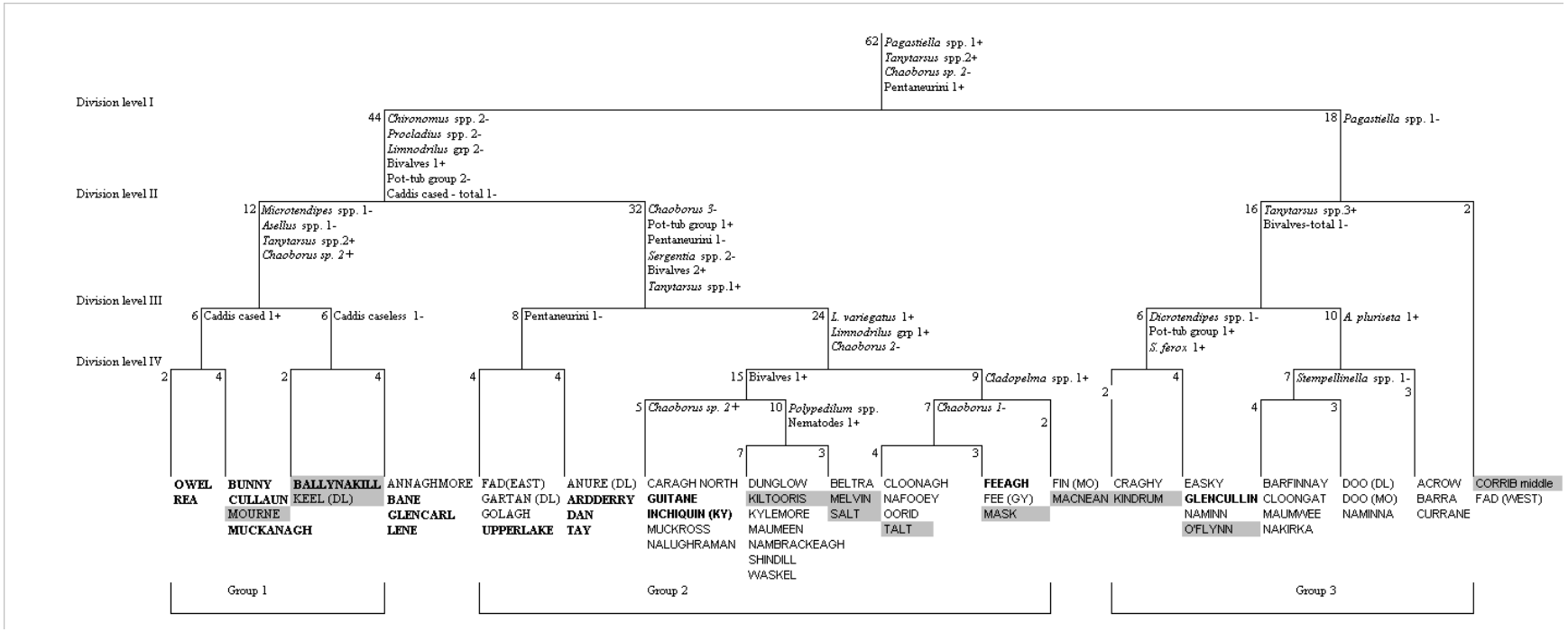


Figure 7.7 The TWINSPAN classification dendrogram of 62 potential reference lakes based on profundal macroinvertebrates. Indicator species selected by TWINSPAN, their pseudospecies cut levels and endgroup preferences are indicated. Lakes that stratified in the present study or in Irvine *et al.* (2001) are bolded. Lakes with atypical alkalinity for the TWINSPAN group to which they belong are shaded e.g. lakes with alkalinities <20 mg l^{-1} $CaCO_3$ in Group 1, typical alkalinity >100 mg l^{-1} $CaCO_3$.

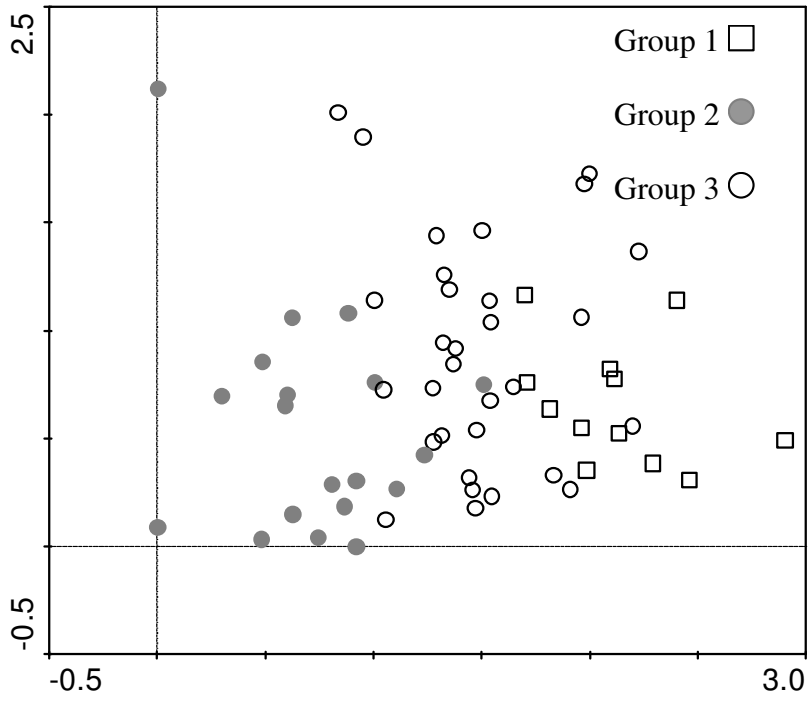


Figure 7.8 DCA ordination diagram with the lake groups indicated.

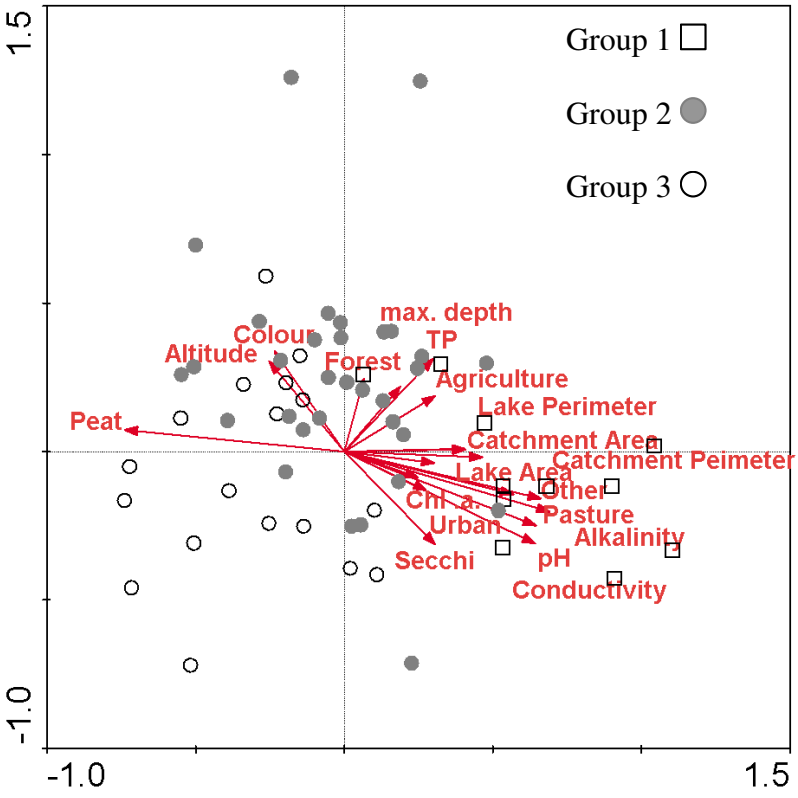


Figure 7.9 CCA ordination diagram of all samples and variables. TWINSPAN endgroups, Group 1, 2 and 3 are indicated.

Table 7.10 Results of the forward selection option for dataset 1 and 2. Variables accounting for a significant ($p < 0.05$) portion of the variation are presented. Lambda 1 = variance explained if the variable was the only one included. Lambda A = additional variance explained.

	Dataset 1			Dataset 2		
Variable	Lambda 1	Lambda A	p	Lambda 1	Lambda A	p
Alkalinity	0.15	**0.15**	**0.002**	0.14	**0.14**	**0.002**
max depth	0.08	**0.07**	**0.004**	0.08	**0.08**	**0.002**
Peat	0.15	**0.07**	**0.004**			
TP	0.07	**0.07**	**0.002**			
Catchment Area	0.09	**0.06**	**0.006**	0.08	0.03	0.266
Catchment Per	0.10	**0.06**	**0.008**	0.09	0.05	0.09
Colour	0.06	**0.05**	**0.004**	0.06	**0.06**	**0.01**
Other	0.11	**0.05**	**0.024**			
Chl *a*	0.05	**0.04**	**0.050**			
Altitude	0.07	0.03	0.24	0.07	**0.06**	**0.016**

Table 7.11 Summary of CCA outputs for Axis 1 using faunal data and full environmental dataset, forward selection (FS) and CVA selected variables, for dataset 1 and 2.

	Dataset 1			Dataset 2		
Axis 1	Full	FS	CVA	Full	FS	CVA
Eigenvalues :	0.205	0.188	0.188	0.171	0.151	0.157
Species-environment correlations :	0.866	0.84	0.838	0.882	0.797	0.792
Cumulative percentage variance						
of species data :	9.9	9.1	9.1	8.4	7.4	7.7
of species-environment relation:	23.4	30.2	38.7	30.5	44.7	42.5
Monte Carlo Test p-value	0.002	0.002	0.002	0.002	0.002	0.002

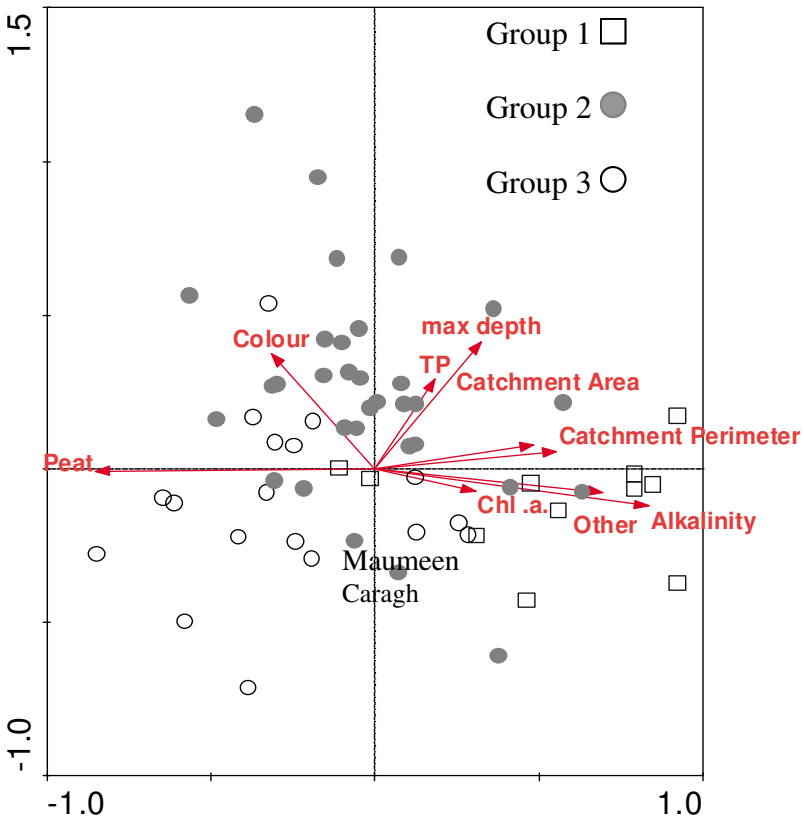


Figure 7.10 CCA ordination diagram of samples and forward selected variables (dataset 1). Group 1, 2, and 3 are indicated.

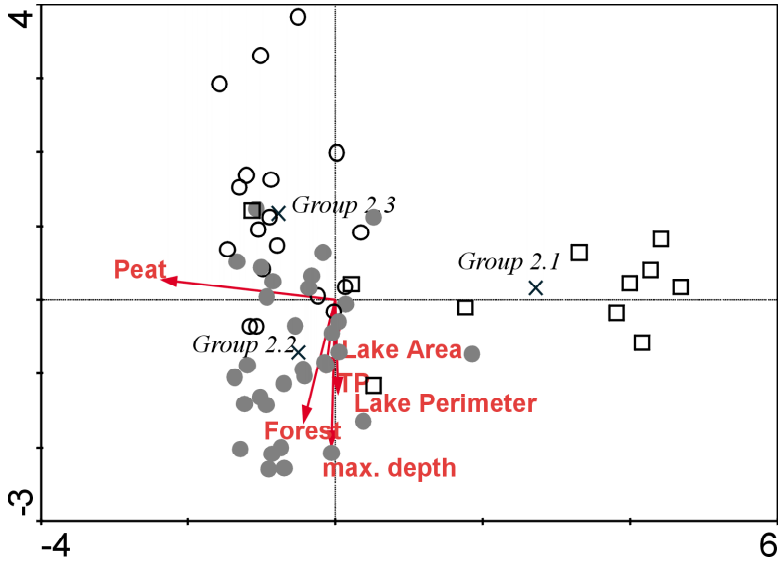


Figure 7.11 CVA ordination diagram of the lakes, group centres (as species data) and variables identified by CVA from dataset 1 as the significant gradients accounting for the separation of the TWINSPAN Groups 1 (2.1), 2 (2.2) and 3 (2.3).

Table 7.12 CVA selected variables, the conditional variation they accounted for (Lambda A) and their significance level.

		Dataset 1		Dataset 2	
	Variable	Lambda A	p	Lambda A	p
Division level I	max. depth	0.13	0.002	0.13	0.006
	Peat	0.1	0.01		
	Lake area	0.06	0.046		
	Lake perimeter	0.07	0.024	0.09	0.022
	Forest	0.04	0.04		
Division level II	Peat	0.53	0.002		
(Group 1 vs Group 2)	Lake perimeter	0.07	0.026		
	Conductivity	0.06	0.012	0.51	0.002
	Altitude	0.02	0.048	0.08	0.004
	Catchment perimeter	0.04	0.05		
	Lake area			0.05	0.008
3 Endgroups	Peat	0.55	0.002		
	max depth	0.16	0.002	0.15	0.002
	TP	0.12	0.014		
	L. Area	0.08	0.038		
	Lake perimeter	0.06	0.036	0.06	0.044
	Forest	0.06	0.046		
	Conductivity			0.39	0.002
	Colour			0.12	0.006
	Altitude			0.08	0.036
	Catchment perimeter			0.11	0.008

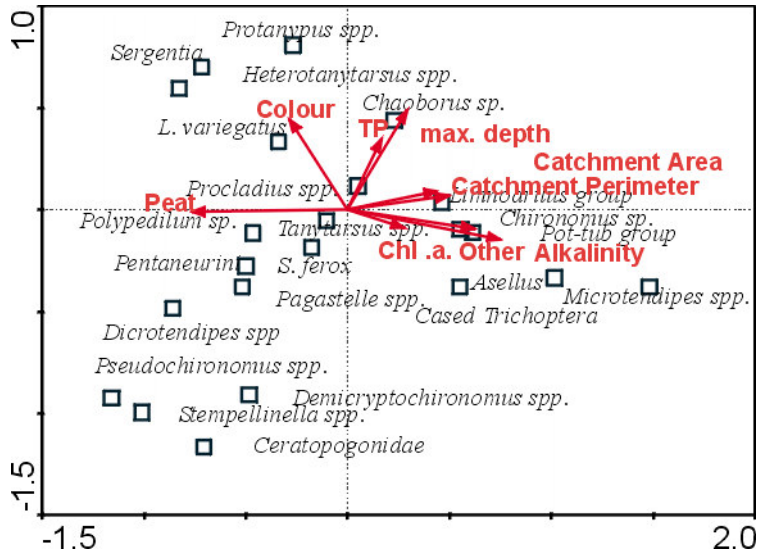


Figure 7.12 CCA ordination diagram of species and forward selected environmental variables (Dataset 1). The analysis focussed on interspecies distance scaling using Hills biplot. Thus species points are located at the optimum of their unimodal response.

Table 7.13 Results of pairwise comparisons (Mann-Whitney tests, $p<0.05$) of variables between TWINSPAN endgroups are presented with their median values. Only significant results are presented.

Variable	Grp 1	Grp 2	Grp 3	Mann-Whitney Results					
				Grps	p	Grps	p	Grps	p
Conductivity	275.50	80.05	78.60	1,2	0.0001	1,3	0.0007		
pH	8.35	6.74	6.39	1,2	0.0046	1,3	0.0046		
Alkalinity	115.42	6.04	3.79	1,2	0.0005	1,3	0.0019		
Colour	15.50	27.50	41.50	1,2	0.0016	1,3	0.0097		
Secchi	4.4	2.75	3.1	1,2	0.0338	1,3	0.0209		
Depth (at site)	16.6	19.65	8.55	1,3	0.0514	2,3	0.0040		
Lake Area	0.86	1	0.37	2,3	0.0171				
Lake perimeter	5.71	7.07	3.66	2,3	0.0138				
Catchment Area	11.14	19.53	5.02	2,3	0.0288				
Forest	0	4.64	0	1,2	0.0445	2,3	0.0043		
Pasture	72.15	0.32	0	1,2	0.0002	1,3	0.0003		
Peat	0.41	66.94	97.51	1,2	0.0003	1,3	0.0001	2,3	0.0258
other	2.97	0.01	0	1,2	0.0215	1,3	0.0018		
Agriculture	9.57	11.04	0.10	2,3	0.0139				
Max. depth	17.65	23.2	10.35	1,3	0.0753	2,3	0.0070		

Table 7.14 Significant results of pairwise comparisons by Mann-Whitney tests, ($p<0.05$) of indicator taxa abundance (m^{-2}) and total abundance (m^{-2}) between TWINSPAN endgroups are presented. Median and mean values of abundance for each taxon are included. Group indicator taxa are highlighted by bolded median values.

Taxa	Median values			Mean values			p	
	Group 1	Group 2	Group 3	Group 1	Group 2	Group 3		
Total abundance	1693	364	1018	1983	657	1100	1,2; p = 0.0003	2,3; p = 0.0083
Chironomus spp.	**285**	0	0	502	30	12	1,2; p= 0.0001	1,3; p = 0.0001
Demicryptochironomus spp.	0	0	**0**	0	0	5	1,3; p = 0.0502	2,3; p = 0.0019
*Dicrotendipe*s spp.	0	0	**9**	0	1	29	1,3; p = 0.0025	2,3; p = 0.0001
Microtendipes spp.	**0**	0	0	20	1	1	1,2; p = 0.0170	
Pagastiella spp.	0	0	**93**	2	2	179	1,3; p = 0.0001	2,3; p = 0.0001
Polypedilum spp.	0	0	**44**	3	9	99	1,3; p = 0.0003	2,3; p = 0.0002
Pseudochironomus spp.	0	0	**0**	0	0	9	1,3; p = 0.0292	2,3; p = 0.0006
Sergentia spp.	0	0	**7**	0	75	65	1,2; p = 0.0312	1,3; p = 0.0048
Stempellinella spp.	0	0	**0**	0	1	13	ns	
Tanytarsus spp.	14	8.89	**104**	71	28	216	1,3; p = 0.0267	2,3; p = 0.0003
Procladius spp.	**116**	18	98	164	34	156	1,2; p = 0.0001	2,3; p = 0.0001
Pentaneurini	0	0	**13**	3	5	25	1,3; p = 0.0037	2,3; p = 0.0007
Heterotanytarsus spp.	0	0	**0**	1	1	15	1,3; p = 0.0440	2,3; p = 0.0084
Protanypus spp.	0	0	**0**	0.00	4	5	1,3; p = 0.0288	
Limnodrilus group	**169**	13	4	492	70	26	1,2; p = 0.0001	1,3; p = 0.0001
Potamothrix-tubifex group	**27**	0	0	179	18	8	1,2; p = 0.0057	1,3; p = 0.0038
Spirosperma ferox	0	0	**0**	0	6	14	1,2; p = 0.0432	1,3; p = 0.0090
Lumbriculidae	0	0	**9**	3	15	23	1,3; p = 0.0074	
Ceratopogonidae	0	0	**0**	0		6	1,3; p = 0.0502	2,3; p = 0.0099
Chaoborus sp.	120	53	0	377	271	11	1,3; p = 0.0016	2,3; p = 0.0002
Cased Trichoptera	**4**	0	0	45	1	9	1,2; p = 0.0007	2,3; p = 0.0047
Hydracarina	0	0	**0**	3	1	11	2,3; p = 0.0032	

Table 7.15 Description of TWINSPAN groups.

TWINSPAN Group	Description
Group 1	Clear (colour median 15.5 mg l^{-1} PtCo, Secchi depth median 4.4 m) high alkalinity (median 115.42 mg l^{-1} $CaCO_3$) deep (median 17.65 m) lakes surrounded by pastures (median 72.15%). Alkalinity and Secchi depth were significantly higher than other groups. Significant indicator taxa were *Chironomus* spp. (IV=92.3), *Limnodrilus* group (IV=83.6), *Potamothrix-Tubifex* group (IV=65.5), *Procladius* spp. (IV=46.4), *Tanytarsus* spp. (IV=57.3), *Microtendipes* spp. (IV=30.9) and cased Trichoptera (IV=40.7) Median abundance values for the aforementioned taxa were significantly higher than other groups with the exception of *Tanytarsus* sp.
Group 2	Coloured (colour median 27.50 mg l^{-1} PtCo) low alkalinity (median 6.04 mg l^{-1} $CaCO_3$) deep (median 23.2 m) lakes surrounded by peatlands (median 66.94%). No significant indicator taxa.
Group 3	Coloured (colour median 41.50 mg l^{-1} PtCo) low alkalinity (median 3.79 mg l^{-1} $CaCO_3$) shallow (median 10.35 m) lakes surrounded by peatlands (median 97.51%). Depth and %peat cover was significantly higher than other groups. Significant indicator taxa were the littoral chironomid taxa, *Pagastiella* spp. (IV=89.9); Pentaneurini (IV=50.3), *Polypedilum* spp. (IV=74.1). *Dicrotendipe*s spp. (IV=53.5), *Pseudochironomus* spp. (IV=33.3), *Heterotanytarsus* spp. (IV=37.4) and one characteristic of oligotrophic lakes; *Demicryptochironomus* spp. (IV=27.8). Median abundance values for the aforementioned taxa were significantly higher compared to the other groups. Additional significant indicator taxa were *Spirosperma ferox*, Lumbriculidae, Ceratopogonidae and Hydracarina, *Sergentia* spp., *Stempellinella* spp. and *Protanypus* spp.

Table 7.16 Part of the correlation matrix from CCA output with Lambda 1, the variance explained if a variable was the only one included. All variables within each correlated set, accounted for a similar level of variation.

Lambda 1	Variable	Correlation Coefficient			Lambda 1	Variable	Correlation Coefficient		
							L. Area	Lake Per	C. Area
					0.07	Colour	**-0.80**		
		pH	Conduct	Alk	0.06		Secchi		
0.14	pH	1					Lake Per	C. Area	C. Per
0.14	Conduct	**0.81**	1		0.08	L. Area	1		
0.15	Alk	**0.93**	**0.89**	1	0.07	Lake Per	**0.93**	1	
.013	Pasture	**0.83**	**0.79**	**0.87**	0.09	C. Area	**0.83**	**0.85**	1
.0.15	Peat	**-0.74**	**-0.71**	**-0.76**	0.1	C. Per	**0.78**	**0.84**	**0.984**

7.3.4 Selection of lake type boundaries

The preceding analyses suggested a typology based on alkalinity and maximum depth, with possibly some influence from colour. The proposed boundaries for alkalinity are < 20 mg l^{-1} $CaCO_3$ (Group 2 and 3) and > 100 mg l^{-1} $CaCO_3$ (Group 1) (Figure 7.13). This was also suggested from the classification of the oligochaetes and chironomids (see relevant sections). A tentative third category would be 20 to 100 mg l^{-1} $CaCO_3$ alkalinity. However, few reference lakes were in this category and they did not form a separate TWINSPAN group; furthermore, such a category was not suggested by the classification of the oligochaetes and chironomids (see Section 7.3.7). This suggested type categories for depth were < 12 m (Group 3) and > 12 m (Group 1 and 2, Figure 7.13). The latter corresponded to the lower 25th percentile for these groups. The

distribution of the three littoral chironomid taxa, *Polypedilum* spp., *Pagastiella* spp. and Pentaneurini also indicated a depth boundary between 10 and 15 m (Figure 7.14). In most instances, sample depth coincided with maximum depth but not always. In all cases, sample depth was considered to be the maximum depth. There was no evidence from TWINSPAN of a shallow lake category for the moderate and high alkalinities. There were few lakes within these categories.

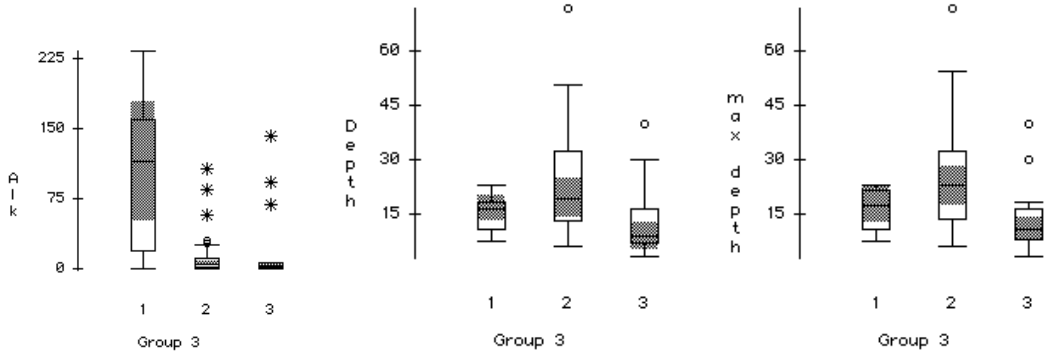


Figure 7.13 Boxplots of the variables, alkalinity, depth (sample depth) and max. depth for the TWINSPAN Groups 1, 2 and 3. Circles= extreme values. Starburst= very extreme data values.

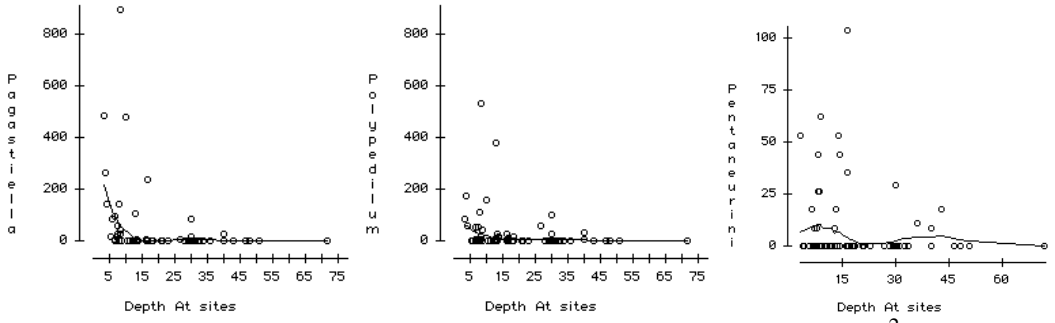


Figure 7.14 The distribution of selected chironomid taxa (nos. m^{-2}) by depth with lowess smoothing for all lakes.

7.3.5 Testing and describing the typology

The lakes were typed based on alkalinity and depth (Table 7.17).

Table 7.17 Reference lake membership in each type. Depth= sample depth

Types	Number of Lakes	Type Code
alkalinity <20 mg l^{-1} $CaCO_3$ depth < 12 m	17	11
alkalinity <20 mg l^{-1} $CaCO_3$ depth >12 m	26	12
alkalinity 20 mg l^{-1}-100 mg l^{-1} $CaCO_3$ depth < 12 m	2	21
alkalinity 20 mg l^{-1}-100 mg l^{-1} $CaCO_3$ depth >12m	6	22
alkalinity >100 mg l^{-1} $CaCO_3$ depth < 12 m	2	31
alkalinity >100 mg l^{-1} $CaCO_3$ depth >12 m	9	32

Distinct biological communities were verified for each type by MRPP (Table 7.18 and Table 7.19). The lake types, 21 and 31, had only two member lakes. This was too few to form valid comparisons.

Table 7.18 Results of MRPP for an alkalinity based typology and a depth based typology. Significant results are highlighted A = Chance-corrected within-group agreement. p = probability.

				Average distance		
		A	p	1	2	3
	n			*43*	*8*	*11*
Alkalinity		0.11	***<0.0001***	0.51	0.46	0.17
Groups	1,2	0.02	*0.1113*	0.50	0.45	
	1,3	0.11	***<0.0001***	0.51		0.17
	2,3	0.13	***0.0004***		0.61	0.31
Depth		A	*p*	1	2	
	n			21	41	
Groups	1,2	0.04	***<0.0001***	0.43	0.50	

Table 7.19 Results of MRPP on communities based on proposed typology. Significant results are highlighted in bold. A = Chance-corrected within-group agreement. p = probability (in italics). Average distance is also given for each group for the initial analysis.

		Groups		11	12	21	22	31	32
	A	p		17	26	2	6	2	9
Alk x depth	0.14	***<0.0001***		0.46	0.52	0.57	0.44	0.04	0.15
			A	Probability					
11: alkalinity <20 mg l^{-1} depth < 12m			11		***0.04***	*0.90*	***0.02***	*0.12*	***0.000***
12: alkalinity <20 mg l^{-1} depth >12m			12	0.03		*0.46*	*0.15*	*0.08*	***0.000***
21:alkalinity 20 mg l^{-1} –100 mg l^{-1} depth<12m			21	-0.05	0.00		*0.77*	*NA*	*0.086*
22: alkalinity 20 mg l^{-1} –100 mg l^{-1} depth			22	0.07	0.02	-0.05		*0.10*	***0.000***
31: alkalinity >100 mg l^{-1} depth < 12m			31	0.05	0.05	0.07			***0.042***
32: alkalinity >100 mg l^{-1} depth >12m			32	0.20	0.15	0.08	0.16	0.14	

Six significant indicator taxa were identified(Table 7.20): three for lake type 31 - *Chironomus* spp., *Cladotanytarsus* sp., and Hydracarina-, one for lake type 21 - *Pagastiella* sp.-, one for lake type 11 - *Heterotanytarsus* spp.- and one for lake type 32 – *Asellus* sp. Identification of indicator taxa for each type in reference condition was difficult for many reasons. Firstly, the typology increased the number of groups from 3 TWINSPAN groups to 6 types based on the same number of lakes with the same fauna. There were insufficient lake numbers to confirm three types, 21, 22 and 32 and to describe their fauna. Indicator taxa for the TWINSPAN groups were selected based on either alkalinity and/or depth preferences. The likelihood of a taxon being confined to a specific lake type was reduced by the increase in group numbers.

For instance, taxa with a preference for shallow depths may not be confined by alkalinity and will therefore occur in 3 lake types. Taxa with a preference for low or high alkalinity may occur in moderate alkalinity lakes also.

It was evident that *Pagastiella* spp. and *Polypedilum* spp. had a preference for shallow depth but their distributions were not confined by alkalinity. The littoral chironomid, *Dicrotendipes* spp. and *Pseudochironomus* spp. preferred shallow, low to moderate alkalinity lakes. Other taxa viz. *Cladotanytarsus* sp. and *Demicryptochironomus* spp. preferred shallow highly alkaline lakes. *Limnodrilus* group, *Aulodrilus pluriseta* and *Potamothrix-tubifex* group occurred across all lakes types, but were more frequent (always present) and abundant in high alkaline lakes. *Spirosperma ferox* and Lumbriculidae occurred across all lakes types but were more abundant and prevalent in the low to moderate alkaline lakes. *Stempellinella* spp., *Sergentia* spp. and *Heterotanytarsus* spp. preferred low alkalinity lakes.

Testing of the WFD System A typology was undertaken using MRPP pairwise comparisons which were limited to types with at least 3 lakes (Table 7.21). Testing of types in block was also carried out. Comparison of the MRPP results of the proposed typology and System A typology showed (Table 7.19, Table 7.22) a slight improvement in the A value using the proposed typology. Both MRPP results were poor compared to those for the TWINSPAN groups. This was because all lakes within the TWINSPAN groups had a similar fauna but not necessarily the same environmental conditions. There were lakes within each TWINSPAN group that did not share the same physical and/or chemical characteristics as their counterparts (Figure 7.9, Figure 7.10). This might also suggest that some lakes were not in reference condition. The TWINSPAN groups were inherently better because they were formed based on faunal composition whereas the typed lakes were based on similarity of environmental conditions. In the latter case, then, lakes with different faunal assemblages were grouped together resulting in a poorer global R. It should be noted, however, that values of A are commonly below 0.1 (McCune *et al*., 2002) in community ecology.

Table 7.20 Description of the proposed typology using indicator values. Taxa which have a preference for a lake type or types are indicated by Indicator values in bold.

	INDICATOR VALUES								
Types	11	12	21	22	31	32	Maxgrp	Indicator	
Numbers:	17	26	2	6	2	9		Value (IV)	p *
Chironomus sp.	1	1	0	0	72	20	**31**	**71.8**	**0.033**
Cladopelma spp.	2	1	14	0	0	4	21	14.4	0.456
Cladotanytarsus sp.	3	0	5	1	59	0	**31**	**58.9**	**0.017**
Demicryptochironomus spp.	8	0	0	2	21	0	31	20.9	0.264
Dicrotendipe s spp.	35	1	6	0	0	0	11	35.1	0.117
Microtendipes spp.	0	0	7	1	0	36	32	35.6	0.095
Pagastiella spp.	11	1	72	0	8	0	**21**	**71.6**	**0.016**
Polypedilum spp.	33	10	24	3	0	0	11	32.9	0.253
Pseudochironomus spp.	15	0	17	0	0	0	21	16.6	0.424
Sergentia spp.	23	18	0	0	0	0	11	23.3	0.407
Stempellinella spp.	12	4	0	0	0	0	11	12.1	0.524
Tanytarsus spp.	19	10	4	4	38	8	31	37.6	0.225
Procladius spp.	11	6	11	3	45	18	31	44.6	0.052
Pentaneurini	8	6	23	0	6	1	21	22.6	0.307
Heterotanytarsus spp.	48	1	0	0	0	0	**11**	**48.2**	**0.045**
Protanypus spp.	1	6	11	0	11	3	31	10.8	0.801
Limnodrilus group	1	2	10	2	43	28	31	42.9	0.179
Aulodrilus pluriseta	1	6	0	2	0	9	32	9.1	0.871
Potamothrix-tubifex group	1	0	2	2	34	44	32	43.7	0.124
Spirosperma ferox	7	16	0	1	8	1	12	15.8	0.642
Lumbriculidae	14	27	0	6	4	2	12	27.3	0.298
Ceratopogonidae	12	1	0	3	0	0	11	11.7	0.609
Chaoborus sp.	11	9	4	16	26	7	31	26.2	0.51
Cased Trichoptera	5	0	3	3	3	31	32	30.8	0.193
Caseless Trichoptera	13	2	0	4	0	0	11	13.2	0.564
Hirudinae	5	15	0	1	0	11	12	14.6	0.669
Asellus spp.	0	1	0	0	0	52	**32**	**51.7**	**0.023**
Gammarus spp.	0	1	0	14	0	0	22	13.5	0.205
Nematode	1	1	0	15	28	1	31	27.6	0.22
Hydracarina	2	0	15	1	51	1	**31**	**50.9**	**0.021**
Bivalves	5	4	28	9	16	1	21	27.8	0.371

Table 7.21 System A typology with the code used and the number of lakes in each type.

Type	Code	n
Calcareous < 3 m mean depth > 50 ha < 200 m	1121	1
Calcareous < 3 m mean depth > 100 ha < 200 m	1131	2
Calcareous 3 - 15 m mean depth < 50 ha < 200 m	1211	1
Calcareous 3 - 15 m mean depth > 50 ha < 200 m	1221	2
Calcareous 3 - 15 m mean depth > 100 ha < 200 m	1231	3
Calcareous 3 - 15 m mean depth > 1000 ha < 200 m	1241	2
Siliceous 3 - 15 m mean depth < 50 ha < 200 m	2211	2
Siliceous 3 - 15 m mean depth > 100 ha < 200 m	2231	1
Organic < 3 m mean depth < 50 ha < 200 m	3111	7
Organic < 3 m mean depth > 50 ha < 200 m	3121	4
Organic < 3 m mean depth > 100 ha < 200 m	3131	1
Organic 3 - 15 m mean depth < 50 ha < 200 m	3211	6
Organic 3 - 15 m mean depth < 50 ha > 200 m	3212	4
Organic 3 - 15 m mean depth > 50 ha < 200 m	3221	8
Organic 3 - 15 m mean depth > 100 ha < 200 m	3231	12
Organic 3 - 15 m mean depth > 100 ha > 200 m	3232	1
Organic 3 - 15 m mean depth > 1000 ha < 200 m	3241	1
Organic > 15 m mean depth < 50 ha > 200 m	3312	1
Organic > 15 m mean depth > 100 ha < 200 m	3331	1

Table 7.22 Results (A values) of selected pairwise comparisons and block testing for System A typology. Data was log x+1 transformed and the Bray-Curtis distance measure was used. Bolded values were significant ($p<0.05$). Shading indicates types with 2 or less members. Stippled shading indicates types included in block testing.

Code	n	1131	1221	3111	3121	3131	3211	3212	3221	3231	Block tests				
1121	1										**0.11**	-0.06	-0.13		
1131	2														
1211	1														
1221	2														
1231	3	-0.01	0.04	0.06	-0.17		**0.15**	**0.30**	**0.18**	**0.15**					
1241	2													**0.08**	
2211	2														
2231	1														
3111	7	0.06			-0.01		0.04	-0.03	**0.12**	**0.18**					**0.06**
3121	4	-0.17					-0.08	-0.01	0.00	-0.02					
3131	1														
3211	6	-0.01						0.05	0.00	0.01					
3212	4	0.19							0.00	0.10					
3221	8	0.02								0.01					
3231	12	0.01					0.01								

7.3.6 Stratification in study lakes

Temperature and oxygen profiles were measured for 168 lakes and an additional 6 lake basins on at least one occasion (Table 7.23). Forty-five lakes thermally stratified, 17 lakes had an anoxic hypolimnion and 24 had low concentrations of oxygen in the hypolimnion on at least one sampling occasion (Table 7.24). Duration of stratification or anoxia could not be determined. However, if a lake was stratified on first sampling in summer and subsequently found to have low oxygen levels or to have become anoxic, it could be assumed that it had remained stratified for the intervening period. Approximately 8 lakes would have stratified for the summer based on this premise. Additional information (Irvine *et al*., 2001) indicated that twelve additional lakes not noted in the present study as having stratified did stratify in 1996/1997 (Table 7.25). In summary, but with consideration to the sampling intensity, the majority of lakes that became anoxic or that experienced low oxygen concentration in their hypolimnion were thermally stratified but did not necessarily have high TP concentration. Not all stratifying lakes experienced oxygen deficits. Stratification and associated DO depletion had implications for describing reference condition because the majority of potential reference lakes in the high alkalinity deep type and a considerable number in the low alkalinity deep type were stratifying lakes (Table 7.24).

Table 7.23 The numbers of lakes (No.) and measured profiles (oxygen and temperature (Temp.)) in each lake type are presented including the number of lakes that stratified, and those that exhibited low oxygen (< 5 mg/l O_2) or anoxia in their hypolimnia.

Reference Lakes		No. of Profiles			other lakes			Lakes with no Profundal Samples			
Type		No.	Oxygen	Temp.	No.	Oxygen	Temp.		No.	Oxygen	Temp.
11	profile	18	39	39	7	8	10	profile	14	16	20
11	thermal stratification	1	2	2	1	1	1	thermal stratification		2	4
11	anoxia	0			0			anoxia			
11	5 mg	0			0			5 mg		2	2
12	profile	27	61	61	15	13	17	**Grand Total**			
12	thermal stratification	5	11	11	4	4	5	profile	168	280	299
12	anoxia	3	3	3	2	2	2	thermal stratification	45	56	62
12	5 mg	8	8	8	1	1	1	anoxia	17	19	19
21	profile	1	3	3	32	47	48	5 mg	26	27	27
21	thermal stratification	0			8	8	9				
21	anoxia	0			2	3	3	lakes basins not counted:			
21	5 mg	1	1	1	3	5	5	Type reference		12	1
22	profile	5	11	12	9	14	15	other		21	3
22	thermal stratification	1	1	1	6	8	8	other		22	2
22	anoxia	1	1	1	4	4	4				
22	5 mg	0			3	3	3				
31	profile	2	4	4	16	19	21				
31	thermal stratification	0			4	4	5				
31	anoxia	0			1	1	1				
31	5 mg	0			2	2	2				
32	profile	10	34	34	12	11	15				
32	thermal stratification	6	9	9	7	6	7				
32	anoxia	2	2	2	3	3	3				
32	5 mg	3	3	3	2	2	2				
total	profile	63	152	153	91	112	126				
total	thermal stratification	13	23	23	30	31	35				
total	anoxia	6	6	6	12	13	13				
total	5 mg	12	12	12	13	13	13				

Table 7.24 The list of lakes (subdivided by type) with the number of profiles and sampling occasions on which they were found to be thermally stratified (TS), anoxic (A) or had low oxygen levels (< 5mg l^{-1}, Low). Date of collection values for Chl *a* and TP, indicator of lake basin shape (ILBS), potential for stratification (PS) and maximum Chl *a* and TP values are also presented. Bolded numbers indicate that a lake was either anoxic or low in oxygen in the absence of stratification. * = anoxia only. The occurrence of stratification prior to anoxia and low oxygen conditions is also indicated.

No. of	Candidate	Reference	Occasions of:				Date of collection				max	
Profiles	type	Lake Name	TS	A	Low	Prior	chl a	TP	ILBS	PS	chl a	TP
3	11	Ardderry	2				2.02	20	4.40	1.62	4.03	20
2	12	Beltra		**1**			2.58	14	3.63	1.33	8.87	26
1		Cloonaghlin			**1**		5.20	2	8.16	2.18	5.20	5
4		Dan	2		1	N	2.40	13	10.46	5.17	8.10	18
1		Fad (east)dl			1		3.31	6	12.51	4.82	3.59	6
3		Glencullin	1				4.39	4	7.27	2.17	7.50	4
4		Guitane	2				3.63	5	5.95	2.10	3.60	8
3		Inchiquin (Kerry)	3		1	Y	4.43	7	10.99	2.82	4.43	7
2		Kylemore			**1**		1.09	6	7.38	2.13	3.79	6
2		Oorid		**1**			2.26	20	5.47	1.91	6.53	20
3		Shindilla	1		1	N	2.38	10	8.63	2.62	3.10	10
2		Tay	1		1	N	0.60	8	14.57	5.72	1.80	8
1		Upper	1	1	1	N	1.80	5	8.69	2.57	2.60	5
3	21	Kindrum			**1**		5.60	13	4.82	3.02	10.08	13
2	22	Ballynakil (gorumna)	1	1		N	9.43	12	11.22	2.94	25.44	15
4	32	Bane	2	1		Y	1.80	5	6.13	3.74	2.40	9
4		Bunny	1				2.80	3	4.30	2.02	2.80	5
4		Cullaun	1		1	N	3.20	9	8.93	3.05	3.60	9
2		Glencar	1				4.31	11	6.47	2.66	7.90	11
4		Lene	3	1	1	Y	1.80	11	3.33	2.84	4.20	17
5		Muckanagh	1		1		4.40	8	5.70	2.42	4.40	11
		Total No. of Lakes:	15	6	13							
	Other Lakes											
1	11	Loughanillaungy	1				1.13	5	3.74	1.51	2.02	5
2	12	Allua	2	1		Y	14.11	11	4.20	1.80	43.10	20
1		Ballinahinchgy	1	1		N	2.62	10	8.14	2.33	3.55	12
1		Bofingy	1				1.57	10	4.09	1.60	10.36	10
2		Glenbeg			**1**		16.93	7	11.67	2.83	16.93	7
1		Nacungupr	1				1.61	20	6.42	3.14	2.70	20
1	21	Carrigencor	1		1	N	5.60	10	4.83	2.66	7.70	14
1		Corry	1				5.56	29	2.00	1.63	17.10	37
2		Duin	1		1	N	12.90	48	3.99		12.90	48
2		Glasshouse	2	2		Y	13.30	37	4.23	2.94	26.60	49
1		Gowna north	1		1	N	10.90	58	1.51	1.67	56.50	63
1		Knappaghbeg	1	1		N	23.14	37	7.70	2.50	27.60	37
1		Meelagh			**1**		2.74	12	3.62	2.31	4.72	24
2		Rowan	1		1	N	4.80	10	5.08	2.99	6.90	23
1		St Johns	1				10.10	35	2.21	1.89	16.10	35
2	22	Drumlaheen	2	2		Y	4.80	25	6.90	3.45	16.50	56
1		Inniscarra west	1				29.02	26	1.91		29.02	26
1		Muckno Sth	1	1		N	35.50	38	3.97		35.50	38
2		Nadreegeel	2		2	Y	4.20	25	6.53	3.83	14.10	34
2		Skeagh	2	1	1	Y	9.70	43	5.35	3.64	9.70	43
2	31	Bridget	1	1	1	Y	26.20	27	4.49	2.27	26.20	30
2		Derravaragh	1				5.00	12	0.55	0.97	7.70	23
2		Finn	1		1	N	4.03	14	1.93	1.06	8.90	22
1		Sunderlin	1				4.40	28	3.82	2.71	10.90	28
1		Templehouse	1				3.91	80	1.30	0.83	8.20	80
1	32	Cavetown	1	1		N	3.75	19	8.60	3.51	10.52	21
2		Culluanyheeda	1		1	N	3.60	34	5.85	2.58	3.60	34
2		Ennell	1		1	N	1.80	14	2.06	2.12	9.30	21
1		Errit	1				6.29	3	4.68	2.31	9.43	16
2		Inchiquin (Clare)	1	1		N	4.40	32	8.01	2.83	6.00	32
1		Nablahy	1	1		N	7.38	15	7.12	3.35	14.43	15
1		Oakport	1				8.75	22	7.47	3.36	8.75	25
		Total No. of Lakes:	30	11	15							

Table 7.25 A list of lakes and the number of occasions they were thermally stratified (TS), anoxic (A) or had low oxygen levels (< 5 mg l^{-1}, Low) in 1996/1997 after Irvine *et al*. (2001). Anoxia and low oxygen levels were based on the lower 25 percentile of the oxygen profile.

Lake Name	TS	A	Low
Ballycullinan	5	3	
Ballyquirke	2		1
Bunny	1		
Caragh	3	1	1
Cullaun	4	1	
Dan	5		1
Dromore	4	3	1
Egish	3		
Feeagh	1		1
Gara S	1		
Gowna	5		1
Graney	1		
Inchiquin CL	7	3	3
Lene	3	1	2
Lettercrafoe	1		
Lickeen	1		1
Maumwee			
Muckno	6	3	3
Owel	3		
Pollaphuca	1		
Ramor	1		
Rea	1		
Talt	3		

7.3.7 Oligochaete and chironomid community structure and lake trophic status

Oligochaete community structure and lake trophic status

TWINSPAN classification of oligochaete data from 166 lakes across the trophic range resulted in 16 communities (Figure 7.15) validated by ANOSIM (Table 7.26). Divisions were based on changes in abundance and often dominance of the five taxa; *Potamothrix-tubifex* group, *Limnodrilu*s group, *Lumbriculus variegatus*, *Aulodrilus pluriseta* and *Spirosperma ferox*, (Figure 7.16). Four taxa - L*imnodrilus* group, *Potamothrix-tubifex* group, *S. ferox* and *Lumbriculus variegatus* - were consistently selected as indicator species at each division level of TWINSPAN (Table 7.27). However, because of the limited number of taxa involved, not every group was assigned an indicator species. It was evident from the Mann-Whitney tests (Table 7.28), CCA (Figure 7.17, Table 7.28) and CVA (Figure 7.18) that oligochaete community structure was primarily influenced by alkalinity or a correlated variable (pH, conductivity), trophic status (TP or chl *a*) and to a lesser extent depth (Figure 7.19).

Table 7.26 Results of ANOSIM for TWINSPAN division levels III to VI from Figure 7.15.

	Division III	Division IV	Division V	Division VI
Global R	0.558	0.538	0.692	0.744
p	0.001	0.001	0.001	0.001
Breakdown of R values		Pairwise comparisons in each category:		
>/=0.75		2	13	78
0.5 -0 .75		1	11	28
0.25 0-.5		3	4	6
<0.25				2*
Insignificant tests				**6

** 5 tests involved Group 8 (n=2) too small for a valid comparison. Groups 3 and 13 had an insignificant test which indicated that these were similar communities.
*The two tests were for pairwise comparison of Groups 5 and 6 and groups 4 and 5

Table 7.27 Output from indicator species analysis for TWINSPAN division levels III to VI from Figure 7.15. See Appendix 5 for taxon abbreviations. Indicator value = Val *Pot-tub* group =*Potamothrix-tubifex* group

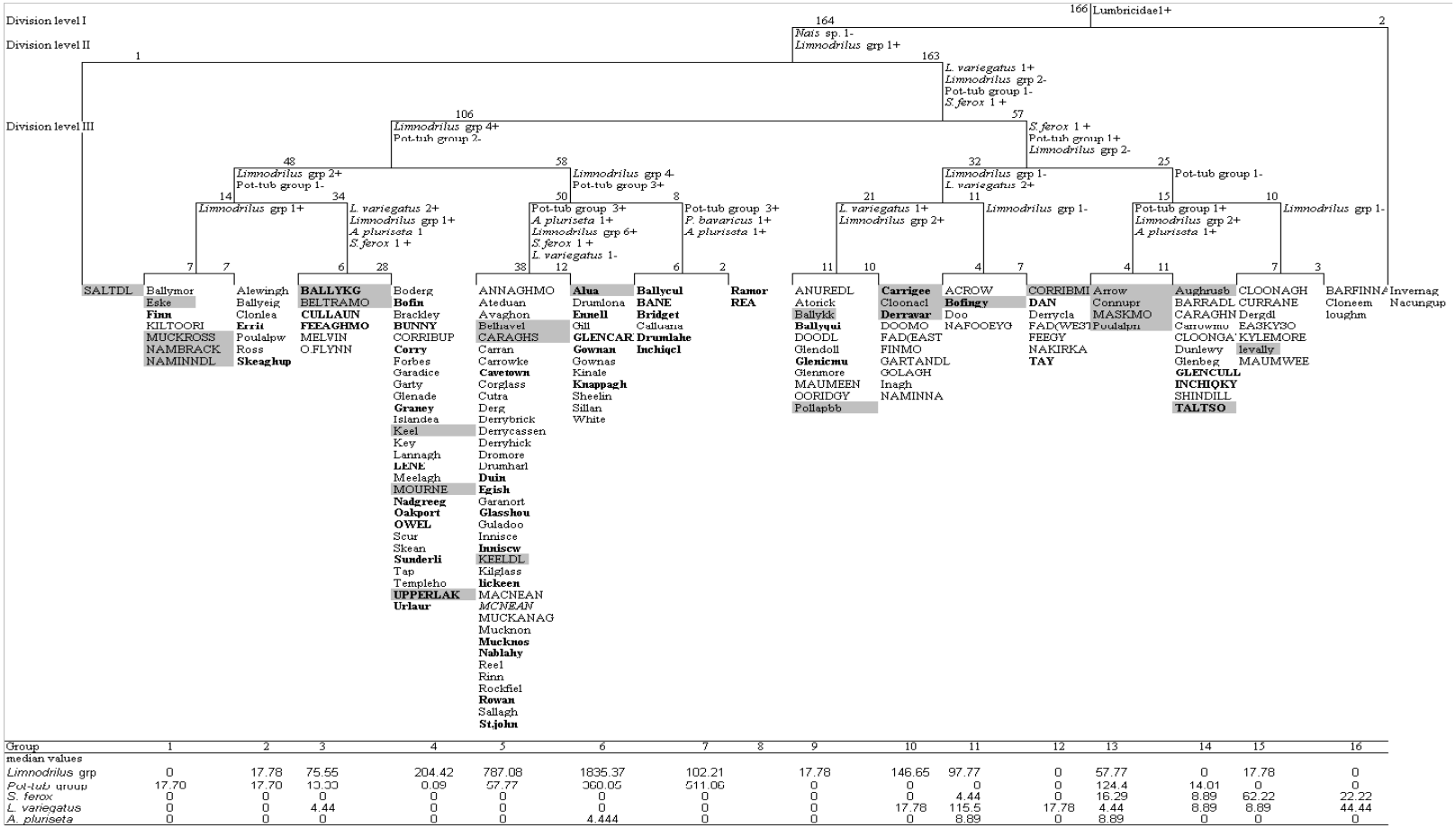
	Div III			Div IV			Div V			Div VI		
	Maxgr	Val	p *	Maxgrp	Val	p *	Maxgrp	Val	p *	Endgrp	Val	p *
Limnodrilus grp	1	87.1	0.001	2	84.9	0.001	3	75	0.001	6	62.9	0.001
Pot-tub group	1	74.8	0.001	2	75.8	0.001	4	83.4	0.001	8	68.5	0.003
Spirosperma ferox	2	44.4	0.001	4	77.7	0.001	8	80.9	0.001	16	65.1	0.001
Lumbriculus variegatus	2	62.4	0.001	3	42.7	0.001	6	57.9	0.001	11	44.7	0.004
Stylodrilus herringianus				4	8	0.023						
P. hammoniensis										8	50	0.016

In summary, 16 oligochaete communities were recognised for the 166 lakes which could be divided into two alkalinity types, >20 mg l^{-1} $CaCO_3$ – groups 2 to 8 – and <20 mg l^{-1} $CaCO_3$ – groups 1, 9 to 16 excluding group 13- (Figure 7.19). Group 13 was an exception having a higher alkalinity than the other groups (groups 9 to 16). Lakes with alkalinities between 20 and 100 mg l^{-1} $CaCO_3$ were not distinct from those with alkalinities greater than 100 mg l^{-1} $CaCO_3$. This led to the conclusion that for oligochaetes, there were only 2 alkalinity types. It was evident from the median TP values for Groups 2 to 8, that these communities represented a range in trophic status (Figure 7.19, Table 7.29). Groups 1 to 3 had low median TP. Groups 4 to 8 represented different degrees of eutrophication. In terms of community structure this was expressed as increased abundance in *Limnodrilus* group and *Potamothrix-Tubifex* group (Figure 7.16). The low alkalinity lakes (Groups 9 to 16, group 13 excluded because of its high alkalinity and similarity to group 3) had no clear trophic gradient

and were mostly oligotrophic. *Spirosperma ferox* and *L. variegatus* typified these groups by their presence but not necessarily dominance. *Limnodrilus* group and *Potamothrix-Tubifex* group were usually present. There was no suggestion that stratification influenced the oligochaete community but stratified lakes were concentrated among the moderate to high alkalinity lakes with increasing prevalence in groups 5 to 8 (Figure 7.15). A TP interaction with depth was evident (Table 7.29). For example, Endgroups 10 and 11 had similar median TP values – 10 and 10.5 μg l^{-1} respectively -but different median depth values –11.25 m and 26.25 m respectively- . *Limnodrilus* group dominated in the shallow lakes (group 10) but *Lumbriculus variegatus* dominated in the deep lakes (group 11).

Table 7.28 Results of significant Mann Whitney tests and CVA for each TWINSPAN division level are presented with the results of CCA using automatic forward selection (fs). At each division level, endgroups were numbered consecutively from left to right starting at 1 (see TWINSPAN diagram). Lamda A is the additional variation by a variable after selection of the variable that explained the most variation.

Division III - 2 groups						
	MW tests		CVA		CCA fs	
Variables	Groups	p	Lambda A	p	Lambda A	P
pH	1,2	0.0001				
Conductivity	1,2	0.0001				
Alkalinity	1,2	0.0001	0.41	0.002		
Secchi	1,2	0.0177				
Chl a	1,2	0.0001	0.02	0.012		
TP	1,2	0.0001				
Division IV - 4 groups						
pH			0.42	0.002		
Secchi	1,2	0.026				
Chl a	1,2	0.0006				
TP	1,2	0.0001	0.13	0.002		
Colour	3,4	0.0497	0.04	0.04		
Division V -8 groups						
pH			0.46	0.002		
Alkalinity	3,4	0.0304				
Conductivity	3,4	0.0447				
TP	5,6	0.0426	0.16	0.002		
Chl a	5,6	0.0214				
Conductivity	7,8	0.0304				
Depth	5,6	0.0236				
Colour			0.08	0.054		
Division VI - 16 groups						
pH			0.54	0.002		
Alkalinity	1,2	0.0331			0.19	0.002
Secchi	1,2	0.0177			0.06	0.002
Chl a	1,2	0.005			0.04	0.01
TP	3,4	0.067	0.2	0.002		
Depth	3,4	0.0548	0.16	0.02	0.04	0.012
Colour	5,6	0.0419	0.18	0.006		
Alkalinity	13,14	0.0059				



Group	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
median values																
Limnodrilus grp	0	17.78	75.55	204.42	787.08	1835.37	102.21		17.78	146.65	97.77	0	57.77	0	17.78	0
Pot-tub group	17.70	17.70	13.33	0.09	57.77	360.05	511.06		0	0	0	0	124.4	14.01	0	0
S. ferox	0	0	0	0	0	0	0		0	0	4.44	0	16.29	8.89	62.22	22.22
L. variegatus	0	0	4.44	0	0	0	0		0	17.78	115.5	17.78	4.44	8.89	8.89	44.44
A. pluriseta	0	0	0	0	0	4.444	0		0	0	8.89	0	8.89	0	0	0

Figure 7.15 The TWINSPAN classification dendrogram based on oilgochaete data with lakes that stratified highlighted in bold. Shaded lakes had atypical alkalinities for Division level 1 endgroups. Candidate reference lakes are in capital letters. A summary of the median abundance values for the oligochaete taxa is also presented.

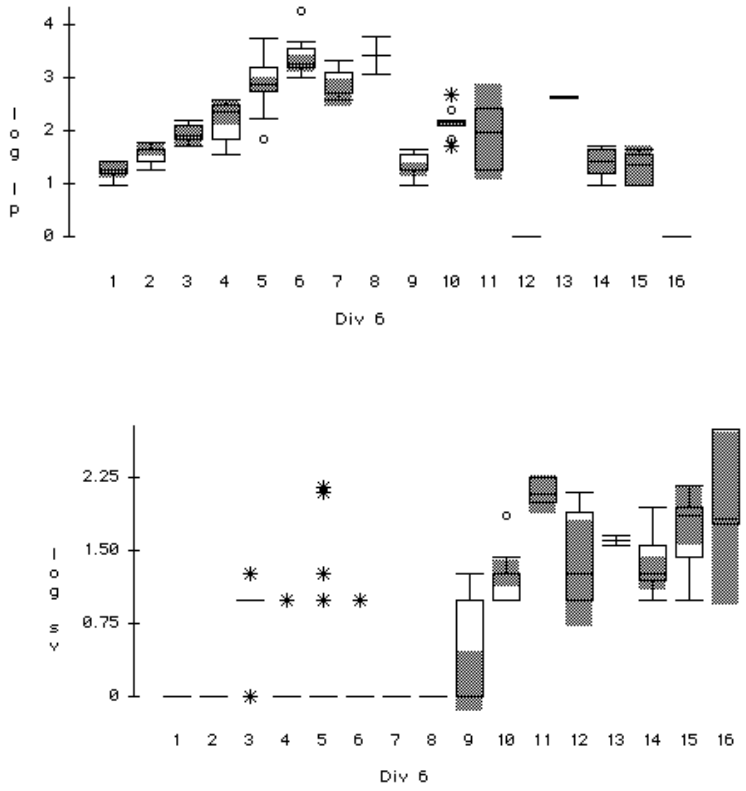


Figure 7.16 The log abundance distribution of *Limnodrilus* group plus *Potamothrix-tubifex* group (lp) and *L. variegatus* plus *S. ferox* (sv) across TWINSPAN endgroups.

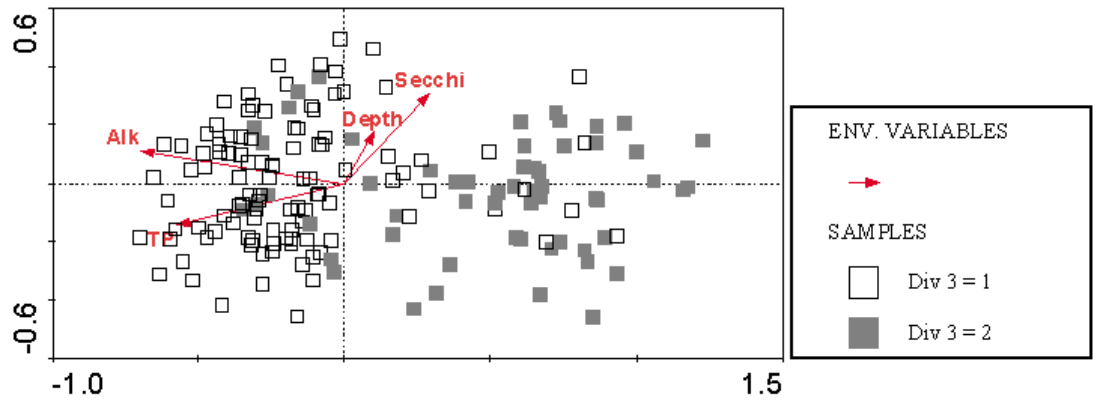


Figure 7.17 CCA ordination diagram with the variables identified by forward selection as explaining the most variance. TP was substituted for chl *a*. Eigenvalue for axis 1 did not change. TWINSPAN endgroups at division level 3 are indicated. (Div 3 = 1 consist of Groups 1 to 8 and Div3 = 2 consist of Groups 9 to 16 of the final groups) Inveragh, Nacung and Salt were omitted.

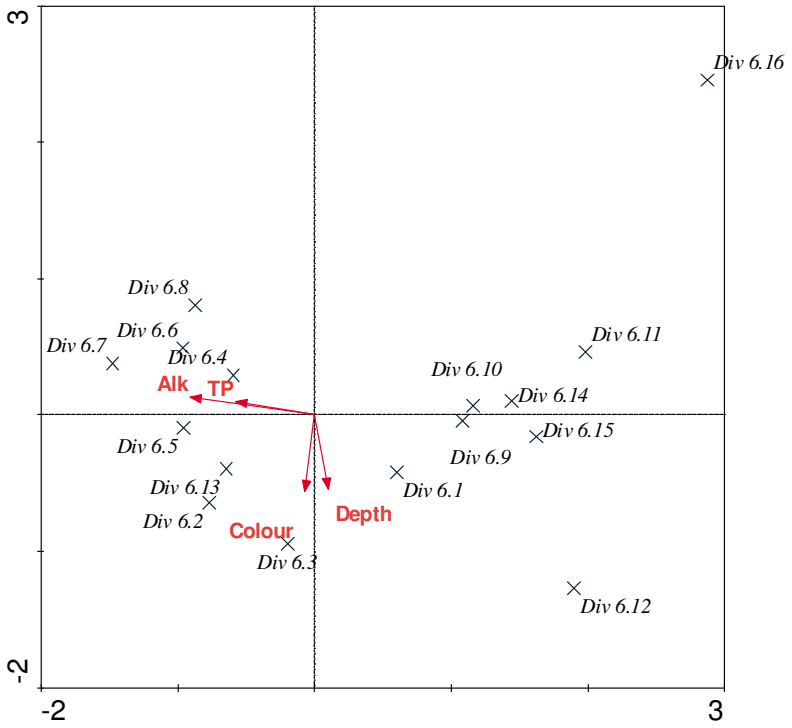


Figure 7.18 CVA ordination diagram of the endgroup centres (as species data) with the variables identified by CVA as the significant gradients accounting for the separation of the 16 groups (alkalinity (ALK) substituted for pH with no difference to the Axis 1 eigenvalue (0.569 compared to 0.567). These variables were correlated).

Table 7.29 Median values of abundance for *Limnodrilus* group, Pot-tub group *(Potamothrix-tubifex* group), *S. ferox* and *L. variegatus* and median values of the variables alkalinity, TP and depth across TWINSPAN end groups. Endgroup membership of reference lakes by type is also presented.

Group	N	*Limnodrilus* grp	Pot-tub group	*S. ferox*	*L. variegatus*	Alk	TP	depth	11	12	21	22	31	32	Grand Total
1	7	0	17.78	0	0	11.44	10	12.6	2	1		1			4
2	7	17.78	17.78	0	0	128.53	12	12.7							
3	6	75.55	13.33	0	4.444	39.04	13	16.45		2		2	1	1	6
4	28	204.42	8.89	0	0	78.98	23	8	1	1				3	5
5	38	787.08	57.77	0	0	77.08	33	10.3	1		1		1	1	4
6	12	1835.37	368.85	0	0	90.29	29	9.6						1	1
7	6	102.21	511.06	0	0	185.79	26	17.4						1	1
8	2	155.54	3436.7	0	0	96.8	34.5	12.95						1	1
9	11	17.78	0	0	0	8.06	17	9.6	2	2					4
10	10	146.65	0	0	17.78	6.22	10.5	11.25	2	4					6
11	4	97.77	0	4.44	115.54	5.72	10	26.25	1	1					2
12	7	0	0	0	17.78	3.06	8	30.4	1	4		1			6
13	4	57.77	124.43	16.29	4.44	97.04	14.5	14.05						1	1
14	11	0	14.81	8.89	8.89	4.61	9	15	2	4		1			7
15	7	17.78	0	62.22	8.89	4.04	9	27	2	3					5
16	3	0	0	22.22	44.44	3.55	4.14	2.5		1					1

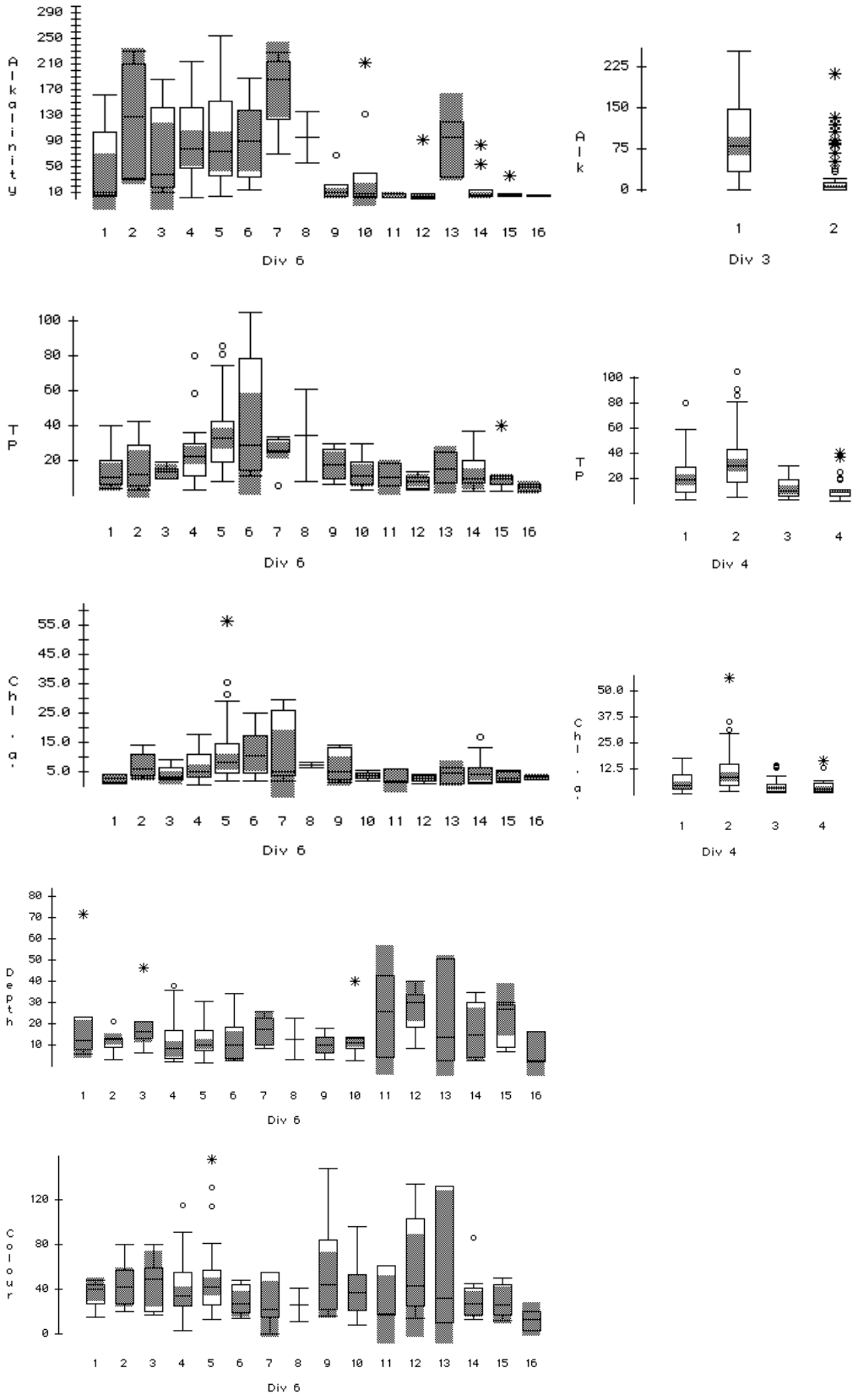


Figure 7.19 Trends in alkalinity, TP, chl a, depth and colour across TWINSPAN groups are shown by boxplots of the variables. Div indicates the TWINSPAN division level.

Chironomid Community Structure and Trophic Status

Chironomid community structure was examined using TWINSPAN for 93 lakes with alkalinities > 20mg l^{-1} $CaCO_3$. Lakes with a lower alkalinity were excluded because they lacked a TP pressure gradient. The TWINSPAN classification resulted in nine communities (Figure 7.20). Divisions were predominantly based on the presence / absence and changes in abundance of three taxa (*Procladius* spp., *Tanytarsus* spp. and *Chironomus* spp.) and also on the presence / absence of *Polypedilum* spp. and the oligotrophic species *Protanypus* spp. (Figure 7.21). ANOSIM validated all endgroups for division levels I to V except where group size was small (Table 7.30). Indicator species analyses confirmed TWINSPAN indicator species as significant indicators and identified additional indicator taxa (Table 7.31, Figure 7.20). The latter were usually rare in abundance and infrequent in occurrence.

The DCA ordination diagram showed that the endgroups 8 and 9 were distinct from the remaining groups (Figure 7.22). Endgroups 1 to 7 were not distinct because these groups were predominantly distinguished by the presence / absence of only three taxa (*Procladius* spp., *Tanytarsus* spp. and *Chironomus* spp.) and their abundance. There was a tendency for endgroups 1 and 5; endgroups 2, 3 and 4; and endgroups 7 and 8; to occupy the same species space. The length of the first DCA gradient was 3.746. Therefore, CCA could be used to detect the underlying environmental gradients. Lake morphology related variables (depth, ILBS), conductivity and trophic status (TP, Chl *a*) were identified as the primary variables influencing community structure (Table 7.32). This was supported by Mann-Whitney tests (Table 7.33). Chl a and other agriculture were identified by CVA as the variables that best explained the 9 endgroups (Table 7.34).

The CVA diagram suggested that the chironomid communities in Groups 1, 5, 8 and 9 were not a product of enrichment based on their position on the chl *a* gradient (Figure 7.23). Trends in the key variables across the TWINSPAN end groups are presented in Figure 7.24 and summarised in Table 7.35.

A summary description of the fauna and main environmental characteristics based on the detailed data in Table 7.36, is presented in Table 7.37. All endgroups consisted of a mix of moderate and high alkalinity lakes. Endgroups 1 to 3 consisted of stratifying

lakes (>50%) representing different trophic status as indicated by their median TP values (Table 7.36). Endgroup 1 lakes were of low nutrient status. Lake shape (ILBS) was quite distinct for this group and the potential for stratification was greatest. Endgroup 2 lakes were typically mesotrophic, resulting in increased *Chironomus* spp. numbers. Some lakes may have experienced periods of low oxygen levels as indicated by the reduced frequency of *Tanytarsus* spp. in this group (Table 7.36). Endgroup 3 lakes were more eutrophic. *Chironomus* spp. numbers were lower and *Tanytarsus* spp. was absent. This indicated that eutrophication may have led to low oxygen levels. Endgroups 4, 6 and 7 consisted mostly of shallow non-stratifying lakes representing different trophic status as indicated by their median TP values. Endgroup 8 consisted of deep non-stratifying lakes with low chlorophyll *a*. Endgroup 9 consisted of shallow non-stratifying lakes. Endgroup 8 or 9 were distinguished from the other endgroups, by the presence and abundance of *Protanypus* spp. and *Polypedilum* spp. respectively and the absence of *Chironomus* spp. Endgroup 5 was not clearly defined. It had a mix of non-stratifying and stratifying lakes with a depth more similar to Endgroups 1 to 3. The median TP value was relatively low. This suggested that a different combination of factors could result in the same fauna.

In summary, the factors considered responsible for the structure and composition of the chironomid communities of moderate to high alkalinity lakes were trophic status (TP) and lake morphology related (stratification) either separately or in tandem. Both factors impact on oxygen availability.

Table 7.30 Results of ANOSIM (lakes >20 mg l^{-1} $CaCO_3$ alkalinity) for division levels I to V, and the TWINSPAN endgroups from Figure 7.20.

	Division I	Division II	Division III	Division IV	Division V	End
Global R	0.434	0.461	0.528	0.58	0.581	0.428
Significance level	0.10%	0.10%	0.10%	0.10%	0.10%	0.10%
Breakdown of R values		Pairwise comparisons in each category:				
>/=0.75		1	9	20	25	6
.5 - .75		2	8	15	15	16
.25 -.5		3	5	8	11	11
<0.25			1	0	0	3
Insignificant tests			5*	22**	16***	2[E]

* all involved groups 5 and 8, n=1; **16 tests involved groups 3 and 8, n=1 the remaining tests involved, groups 2 or 11 (n=2), and groups 1, 2; ***11 tests involved groups 14 (n=1) 3 tests involved group 24 (n=2) the remaining tests were Groups 12, 13; Groups 12, 14; Groups 13, 14; Groups 23, 24. E tests involved Groups 6 and 9 and 7 and 9. Possibly influenced by sample size. Lake groups were numbered sequentially from left to right based on the TWINSPAN dendrogram starting at Division level 1 (1,2) as far as Division V, unless they did not divide further, in which case they were not assigned a new number but retained the number assigned from the previous division.

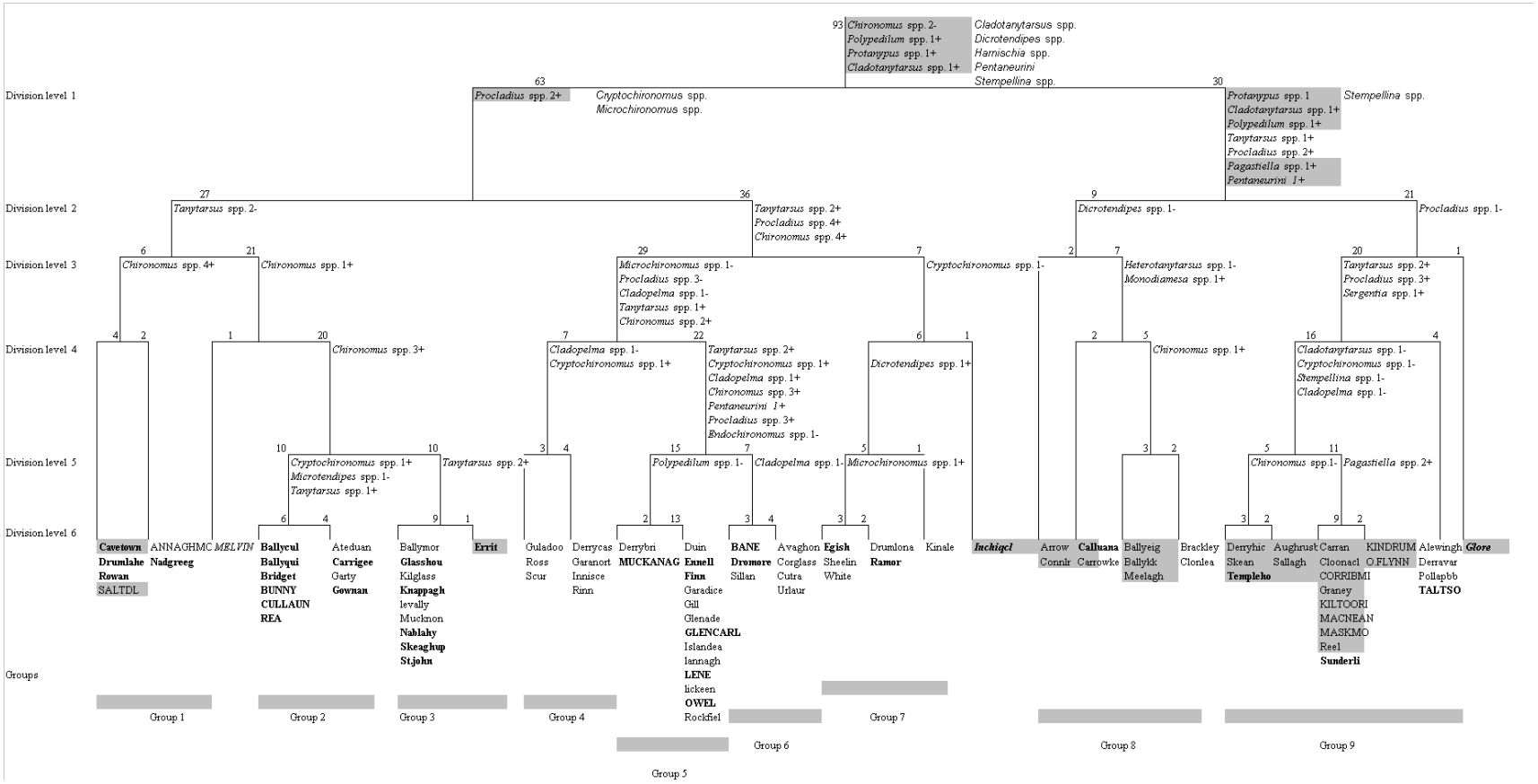


Figure 7.20 The dendrogram for the TWINSPAN classification of lakes with alkalinities > 20 mg l^{-1} $CaCO_3$. Lakes that stratified are highlighted in bold. Shaded lakes clustered with the <20mg l^{-1} $CaCO_3$ alkalinity lakes in the original TWINSPAN of chironomids and all lakes. Lakes with a chironomid assemblage atypical of the endgroup to which they belong are italicized. Indicator species, pseudospecies cut levels with group preferences are also presented. Shaded TWINSPAN indicator taxa were identified by indicator species analysis as being significant ($p<0.05$). Additional significant taxa are listed above their group preference.

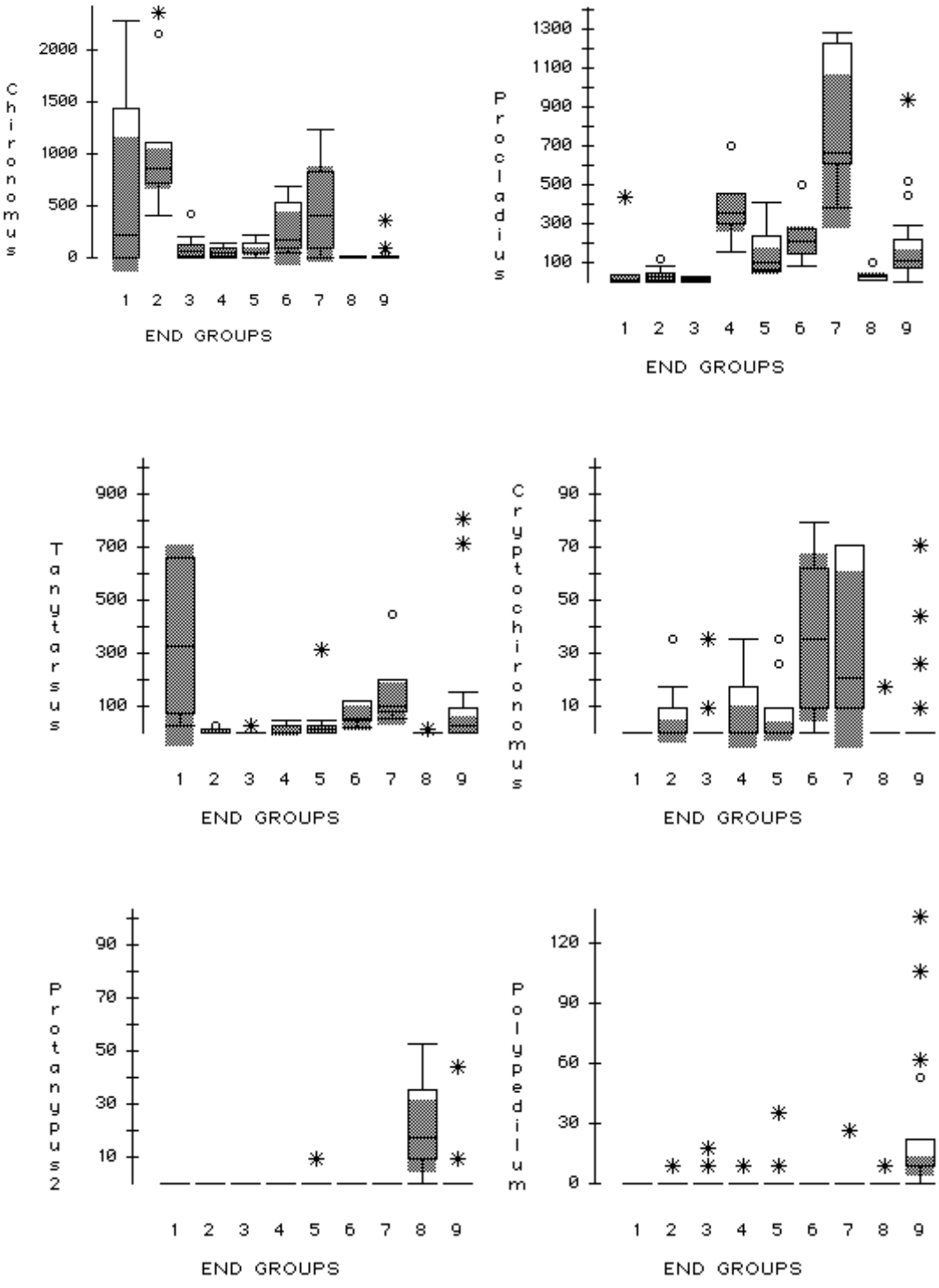


Figure 7.21 The abundance (m^{-2}) patterns of *Chironomus* spp., *Procladius* spp., *Tanytarsus* spp., *Cryptochironomus* spp., *Protanypus* spp. and *Polypedilum* spp. across the 9 endgroups.

Table 7.31 Results of Indicator species analysis for TWINSPAN division levels 1, 2, 3 (endgroups 1 to 4 at division level 3 only, from left to right on the TWINSPAN dendrogram), and endgroups. Significant ($p<0.05$) indicator taxa are highlighted in bold. IV=indicator value. See Appendix 7, for taxon abbreviations.

	Division 1			Division 2			Division 3 -Group 1-4			Endgroups		
Taxon	Maxgrp	IV	p	Maxgrp	IV	p	Maxgrp	IV	p	Maxgrp	IV	p
ChironSP	1	83.6	**0.001**	1	64.2	**0.001**	2	29.5	0.776	**2**	**37.4**	**0.005**
Cladop	2	21.9	**0.04**	4	20.4	0.099	3	14.6	0.301	6	13.3	0.417
Cladot	2	28	**0.003**	4	37.9	**0.002**	4	27.2	0.053	7	21.7	0.107
Crypto	1	27.4	0.152	2	34.1	**0.026**	4	68.6	**0.002**	**7**	**54.9**	**0.001**
Demicr	2	6.1	0.312	4	8	0.212	3	3.4	1	6	11.3	0.269
Dicrot	2	13	**0.04**	4	7.1	0.404	4	14.3	0.209	9	6.6	0.845
Endoch	1	6.3	0.298	2	11.1	0.101	4	11.8	0.242	7	11.8	0.235
Harnis	2	12.6	**0.032**	3	26.4	**0.005**	3	6.9	0.698	**8**	**22.7**	**0.028**
Microc	1	17.5	0.059	2	27.5	**0.02**	4	40.5	**0.017**	**7**	**41.6**	**0.007**
Microt	1	13.5	0.138	1	14.4	0.132	4	19.6	0.171	**2**	**26**	**0.028**
Pagast	2	19.8	**0.007**	4	28	**0.011**	1	14.6	0.086	9	28	0.075
Parach	2	3	0.777	4	4.3	0.49	3	3.4	1	4	5.2	0.816
Polype	2	50.9	**0.001**	4	63.6	**0.001**	4	7	0.8	**9**	**51.7**	**0.001**
Pseudo	2	6.7	0.106	4	9.5	0.154				9	10	0.511
Sergen	1	1.4	1	2	2.5	0.978	4	14.3	0.215	9	10	0.395
StempLI	2	3.3	0.333	4	4.8	0.316				9	5	1
StempeLA	2	9.8	**0.047**	4	14	**0.045**	3	6.9	0.689	9	13.7	0.241
Sticto	1	1.6	1	2	2.8	1	4	14.3	0.215			
Tanyta	1	39.1	0.53	1	24.8	0.849	1	80.3	**0.004**	**1**	**76.9**	**0.002**
Procla	1	52.5	0.379	2	56.1	**0.001**	4	68.5	**0.001**	**7**	**41**	**0.001**
Pentan	2	18	**0.014**	4	25.1	**0.017**	4	21.5	0.067	9	9.8	0.383
HeteroTA	2	8.9	0.072	3	30.8	**0.001**	2	4.8	0.536	**8**	**27.2**	**0.004**
HeteroTR	2	6.7	0.122	4	9.5	0.139				9	10	0.5
Psectr	1	1.6	1	2	2.8	1	3	3.4	1	5	6.7	0.766
Protan	2	32.3	**0.001**	3	67.5	**0.001**	3	6.9	0.684	**8**	**65.4**	**0.001**
Monodi	2	10.7	0.26	3	28.4	**0.005**	2	10.5	0.237	**8**	**22.9**	**0.057**

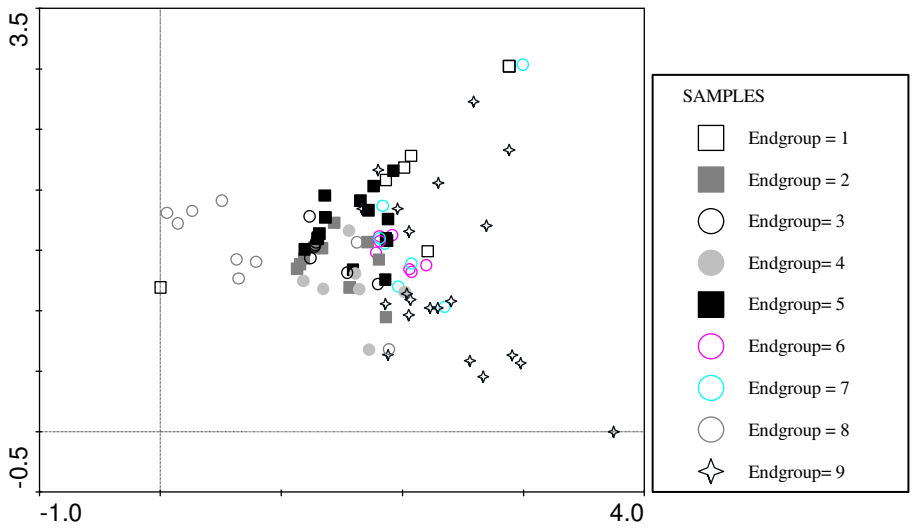


Figure 7.22 The DCA ordination diagram for lakes >20 mg l^{-1} $CaCO_3$ alkalinity with the 9 end groups indicated.

Table 7.32 Results of forward selection option in CCA (chironomid data, lakes > 20 mg l^{-1} $CaCO_3$).

Variable	Lambda 1	Lambda A	p
Dataset 1: 19 variables			
Conductivity	0.1	0.1	0.004
Depth	0.1	0.1	0.002
Lake Area	0.1	0.11	0.002
Altitude	0.09	0.08	0.034
TP	0.09	0.07	0.024
Dataset 2: 22 variables			
ILBS	0.11	0.11	0.004
Conductivity	0.1	0.1	0.002
Secchi	0.1	0.09	0.002
Altitude	0.09	0.08	0.044
Peat	0.07	0.07	0.014

Table 7.33 Results of comparisons (t-tests) of TWINSPAN endgroups for Division 1 and 2 (> 20 mg l^{-1} $CaCO_3$ alkalinity lakes). Only significant variables are shown. All 22 variables were tested.

TWINSPAN Endgroups	Variables	P
Division 1	Chl 'a'	0.002
t-tests	LAREA	0.020
	PEAT	0.004
Division 2		
1,2	TP	0.044
t-tests	DEPTH	0.025
	ILBS	0.013
	CCP	0.043
	CAREA	0.030
	CPERI	0.034
3,4	URBAN	0.027

Table 7.34 CVA results for TWINSPAN divisions 1 to 3 for lakes >20 mg l^{-1} $CaCO_3$. Lambda 1 is the variance explained if a variable was the only one included in the analyses and Lambda A is the additional variance explained.

	Dataset 1: 19 variables				Dataset 2: 22 variables			
	Variable	Lambda 1	Lambda A	p	Variable	Lambda 1	Lambda A	p
Division 1	Conduct	0.02	0.04	0.044	Chl .a.	0.11	0.11	0.004
	Chl. 'a'	0.11	0.11	0.004	Peat	0.1	0.06	0.01
	TP	0.01	0.03	0.044	LArea	0	0.05	0.01
	LArea	0.05	0.05	0.01	CCP	0	0.05	0.02
	CArea	0	0.04	0.022	TP	0.01	0.03	0.05
Division 2:								
Group 1, 2	LArea	0.04	0.07	0.028	ILBS	0.15	0.15	0.004
	Peat	0	0.07	0.04	TP	0.05	0.05	0.054
Group 3,4	Urban	0.14	0.14	0.048	Urban	0.14	0.048	0.14
Division 3								
Group 1, 2	CArea	0.2	0.2	0.022	CCP	0.31	0.31	0.004
	Altitude	0.15	0.11	0.05	CArea	0.2	0.15	0.01
Group 3,4	Peat	0.15	0.15	0.036	Peat	0.15	0.15	0.036
	Cperi	0.05	0.09	0.046	CCP	0.04	0.09	0.032
Endgroups					Chl .a.	0.25	0.25	0.004
					Oagric	0.17	0.17	0.05

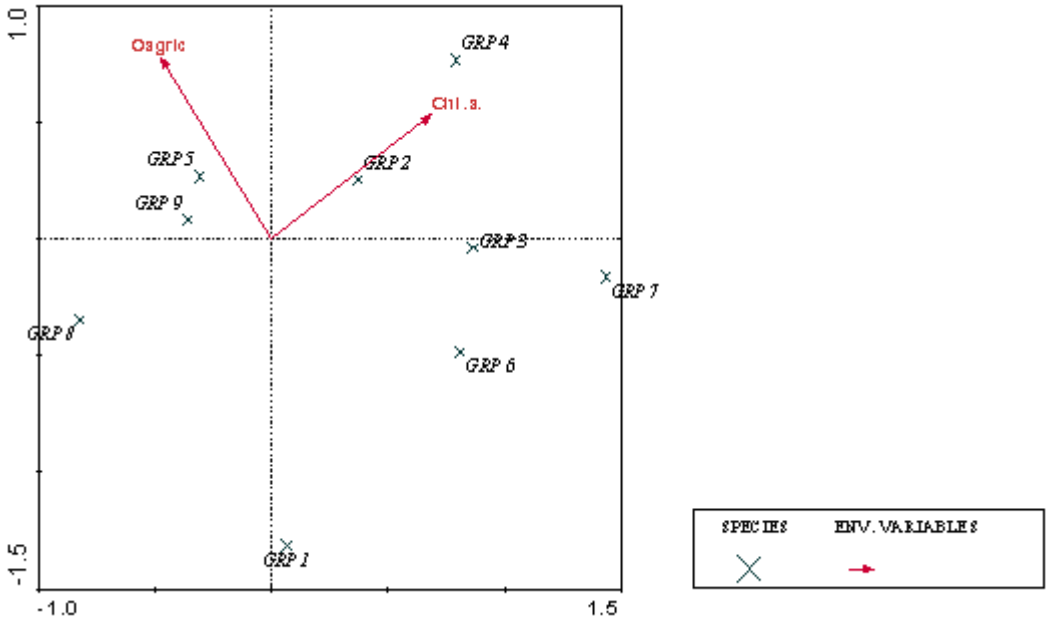


Figure 7.23 CVA ordination diagram of the endgroup centres (as species data) with the variables identified by CVA as the significant gradients accounting for the separation of the 9 groups.

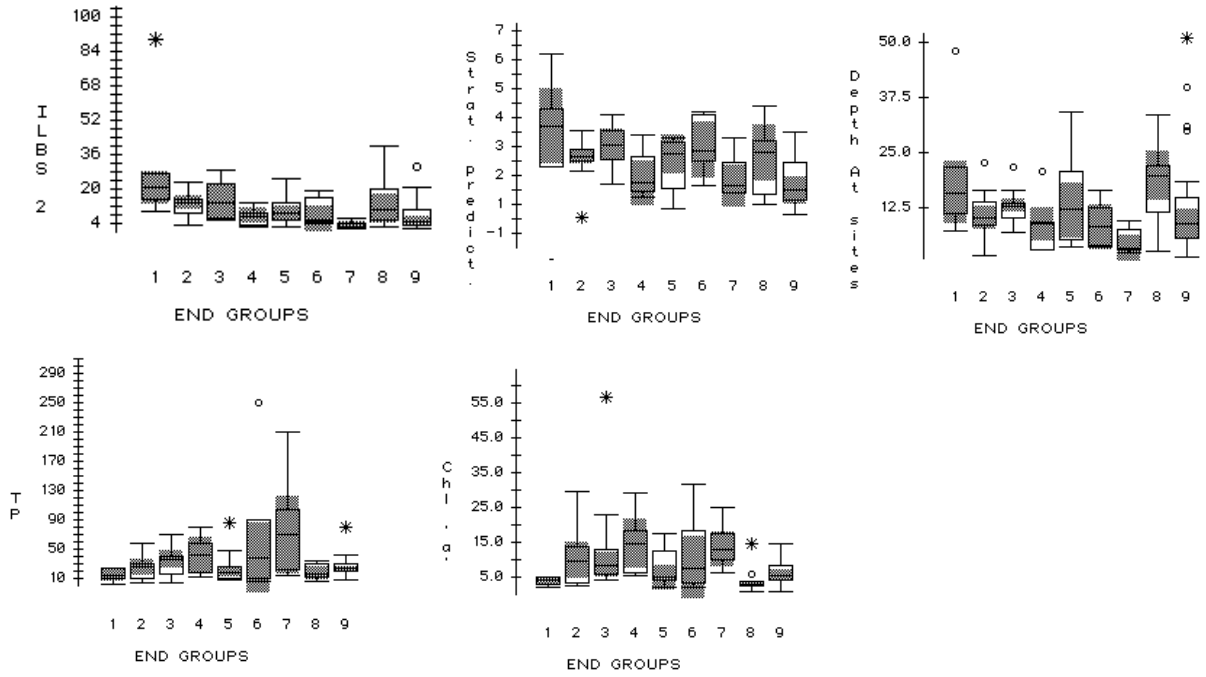


Figure 7.24 Trends in ILBS, prediction of stratification (strat. Predict.), depth, TP, and Chl *a*.

Table 7.35 The number of lakes in each endgroup (n) and in specified categories of alkalinity (1= 20 – 100 mg l^{-1} $CaCO_3$ 2 => 100 mg l^{-1} $CaCO_3$) and TP (1=<10 µg l^{-1}, 2= 10–20 µg l^{-1}, 3 =20-30 µg l^{-1}, 4=> 30 µg $^{-1}$) are presented. The number of stratifying lakes (Strat.) and the number of reference lakes by type (see Table 7.17 for codes) in each endgroup are also presented.

		Alkalinity Category		TP Category				Strat.	No. of Reference Lakes			
End	n	1	2	1	2	3	4		21	22	31	32
1	6	4	2	1	3	2		4		1	1	
	1	1						0				
2	10	4	6	3	1	2	4	8				3
3	10	6	4	1	2		7	6				
4	7	4	3		2		5	0				
5	15	6	9	2	6	4	3	8				4
6	7	4	3	2			5	2				1
7	6	4	2		1	1	4	3				
	1	1						1				
8	9	4	5	1	5		3	1				
9	20	11	9	1	4	9	6	2	2	3	1	1
	1	1						0				

Table 7.36 The frequency of occurrence of selected taxa and their abundance in each group together with the median values of selected environmental variables. + indicates that the median value was not calculated due to small sample size but the taxon were present.

Endgroup	1	2	3	4	5	6	7	8	9
n	6	10	10	7	15	7	6	9	20
Frequency									
Chironomus spp.	4	10	10	6	14	7	5	4	7
Tanytarsus spp.	6	4	1	2	8	7	6	1	13
Procladius spp.	3	8	9	7	15	7	6	9	20
Cryptochironomus spp.		3	2	3	4	6	6	1	5
Pagastiella spp.	1	1							6
Polypedilum spp.		2	2	1	2		1	1	16
Pentaneurini						2	2		5
Protanypus spp.					2			7	3
Abundance									
Chironomus spp.	225.16	862.14	75.55	53.33	62.22	177.76	413.29	+	+
Tanytarsus spp.	328.86	+	+	+	8.89	53.33	106.66	+	26.66
Procladius spp.	4.44	22.22	8.89	355.52	106.66	213.31	671.04	26.66	115.54
Cryptochironomus spp.	0	+	+	+	+	35.55	20.74	0	+
Pagastiella spp.	+	+	0	0	0	0	0	0	+
Polypedilum spp.	0	+	+	+	+	0	+	+	8.89
Pentaneurini	0	0	0	0	0	+	+	0	+
Protanypus spp.	0	0	0	0	+	0	0	17.78	+
Environmental Character									
No of lakes that stratified	4	8	6	0	8	2	3	1	2
ILBS	6.72	4.39	4.46	2.50	2.92	2.01	1.04	3.62	1.70
Chl *a*	3.97	9.68	8.54	14.90	4.80	7.70	13.10	2.74	5.60
TP	15.5	26	37	43	19	38	70	17	24
Depth	15.95	10.15	12.95	8.6	11.9	8	3.2	20	8.7

Table 7.37 Description of lakes and chironomid assemblages with a general guide to abundance and frequency for the 9 chironomid based endgroups.

End grp	Description
1	deep (median 15.5 m), stratifying, predominantly low TP lakes (median=15.5 µg l^{-1}). *Tanytarsus* spp. was present in all lakes, (>250 m^{-2}). *Procladius* spp. (0-500 m^{-2}) and *Chironomus* spp. (100-2000 m^{-2}) occurred in 75% of lakes.
2	shallow (median 10.15 m) stratifying lakes with medium (median=26 µg l^{-1}) TP levels. *Procladius* spp. (<100 m^{-2}) and *Chironomus* spp. (>400 m^{-2}) were present in all lakes. *Tanytarsus* spp. (<50 m^{-2}) was present in 25% of lakes.
3	shallow (median 12.95 m) stratifying lakes with high (median 37 µg l^{-1}) TP levels. *Tanytarsus* spp. was absent. *Procladius* spp. (<50 m^{-2}) and *Chironomus* spp. (<200 m^{-2}) were present in all lakes.
4	shallow (median 8.6 m) lakes with high (median 43 µg l^{-1}) TP levels. *Tanytarsus* spp. occurred in 2 lakes only. *Procladius* spp. (>300 m^{-2}) and *Chironomus* spp. (<100 m^{-2}) were present in all lakes. *Microchironomus* spp. (<50 m^{-2}) was also present.
5	shallow (median 11.9 m) lakes with low (median 19 µg l^{-1}) TP levels. *Tanytarsus* spp. (<50 m^{-2}) occurred in 50% of lakes. *Procladius* spp. (>50 m^{-2}) and C*hironomus* spp. (<200 m^{-2}) were present in all lakes.
6	shallow (median 8 m) lakes with high (median 38 µg l^{-1}) TP levels. *Tanytarsus* spp., *Procladius* spp. (>200 m^{-2}) *Chironomus* spp. (<200 m^{-2}) and C*ryptochironomus* spp. were present in all lakes.
7	shallow (median 3.2 m) lakes with high (median 70 µg l^{-1}) TP levels. *Tanytarsus* spp. (>50 m^{-2}), *Procladius* spp. (>300 m^{-2}), *Chironomus* spp. (>500 m^{-2}), and *Cryptochironomus* spp., were present in all lakes.
8	deep (median 20 m) lakes with low (median 17 µg l^{-1}) TP levels. *Procladius* spp. (<50m^{-2}) and the oligotrophic taxa; *Protanypus* spp. were present in all lakes. *Chironomus* spp. occurred in 25% of lakes in low abundance (<50 m^{-2}). *Tanytarsus* spp. absent.
9	shallow (median 8.7 m) lakes with medium (median 24 µg l^{-1}) TP levels. *Procladius* spp. (<250 m^{-2}) was present in all lakes. *Tanytarsus* spp. (<300 m^{-2}) was present in 75% of lakes. *Chironomus* spp. (<50 m^{-2}) occurred in 25% of lakes. *Polypedilium* spp. was present in 75% of lakes in low abundance (<50 m^{-2}). *Cladotanytarsus* spp. (<50 m^{-2}) was present in 50% of lakes.

7.3.8 Descriptions of reference condition and deviation from reference

Description of reference condition and deviation from reference for low and high alkalinity, deep (stratifying and non-stratifying) and shallow lakes, based on the development of the typology (reference lakes), classification of the oligochaete (all lakes) and chironomids (>20 mg l^{-1} $CaCO_3$ alkalinity lakes) components are presented in Table 7.38. The deviation from reference conditions described above was the expected sequence of events with progressive eutrophication and the changes in the chironomid and oligochaete communities would be expected to occur concurrently. However, the descriptions are incomplete because not all lakes were included for the classification of the chironomid components. Some lake types lacked a TP gradient and had either insufficiently low TP lakes or high TP lakes. It was not possible to ascertain the points or TP values at which each change - e.g. change in abundance, disappearance - in taxa would be triggered. A shallow, low alkalinity (<20 mg l^{-1} $CaCO_3$) reference lake might lose its oligotrophic taxa at different TP levels compared

to a lake with a higher alkalinity e.g. *Sergentia* sp. might disappear at 10 μg l^{-1} TP, where as *Polypedilum* spp., an indicator of reference condition for all types of shallow lakes, was present up to 30 μg l^{-1} TP in lakes with >20 mg l^{-1} $CaCO_3$. It is possible that *Chironomus* spp. begins to increase in numbers prior to the disappearance of the littoral or oligotrophic chironomids. Also, the oligochaete taxa richness may increase as lakes become mesotrophic, only to decline again. The alkalinity category >20 mg l^{-1} $CaCO_3$ was not subdivided because there was no evidence that a moderate and a high alkalinity type existed. A stratified type for deep lakes was suggested based on the evidence from the classification of the chironomid component.

A simple abundance - based system using abundance categories of *Chironomus* spp. and *Limnodrilus* group plus *Potamothrix-tubifex* group (Table 7.39) broadly distinguished oligochaete and chironomid TWINSPAN endgroups (Figure 7.25). Its relationship to log TP is also presented (Figure 7.26). This system is effectively the Quirke Index (Irvine *et al*., 2001) without the modification for *Chironomus* spp. abundance and excluding *Procladius* spp. abundance. It was statistically effective (Figure 7.26).

Table 7.38 Description of reference communities and expected deviation from reference for low and high alkalinity deep (stratifying and non-stratifying) and shallow lakes with progressive pressure (eutrophication) based on the classification of the oligochaete and chironomid components.

	Reference state	*Deviation from reference*
Shallow Lakes >20 mg l^{-1} $CaCO_3$	• *Chironomus* spp. absent or present in low in numbers (<44.44 m^{-2}). • *Tanytarsus* spp. present in low numbers. • *Procladius* spp. present. • *Polypedilum* spp. present and other littoral or oligotrophic chironomids maybe present. • Oligochaetes *(Limnodrilus* and *Pot-tub* group) present in low numbers (</=50 m^{-2}).	1. *Polypedilum* spp. and other littoral or oligotrophic chironomids decline in numbers becoming absent. 2. *Chironomus* spp. increase in numbers becoming the dominant chironomid taxon. 3. *Tanytarsus* spp. initially increases in abundance but as oxygen becomes limited it declines and disappears. 4. *Cryptochironomus* spp. appears. 5. Oligochaete: *S. ferox* and *L. variegatus* disappear. Oligochaete numbers increase i.e. *Limnodrilus* and *Pot-tub* group becoming dominant and as oxygen becomes limiting they are the dominant taxa.
Shallow Lakes <20 mg l^{-1} $CaCO_3$	• *Chironomus* spp. absent or low in numbers (<44.44 m^{-2}). • *Tanytarsus* spp. present. • *Procladius* spp. present. • *Polypedilum* spp., Pentaneurini, *Pagastiella* spp. present and other littoral or oligotrophic taxa such as *Sergentia* spp. H*eterotanytarsus* spp. present. • Oligochaetes (*Limnodrilus* group and Pot-tub group) low in numbers particularly (<50 m^{-2}) or absent. • *S. ferox* and /or *L. variegatus* present.	1. Littoral and oligotrophic chironomids decline in numbers becoming absent 2. *Chironomus* spp. increase in numbers becoming dominant. 3. *Tanytarsus* spp. initially increase in abundance but decline as oxygen becomes limiting eventually disappearing. 4. *Cryptochironomus* spp. appears. 5. Concurrently, the oligochaetes *S. ferox* and *L. variegatus* disappear. Oligochaete numbers increase i.e. *Limnodrilus* and Pot-tub group becoming dominant as oxygen becomes limiting they persist are the prevailing taxa.
Deep lakes >20 mg l^{-1} $CaCO_3$ and <20 mg l^{-1} $CaCO_3$ unstratified (or stratified but not oxygen limited)	• *Chironomus* spp. absent or present in low in numbers (<44.44 m^{-2}). • *Tanytarsus* spp. may be present in low numbers or absent. • *Procladius* spp. present. • *Protanypus* spp. present. • Other oligotrophic taxa with a preference for depth may be present. • Oligochaetes low in numbers particularly *Limnodrilus* group and Pot-tub group (<50 m^{-2}).	1. *Protanypus* spp. and other oligotrophic taxa decline in numbers and eventually disappear. 2. *Chironomus* spp. increase in numbers becoming dominant. 3. *Tanytarsus* spp. initially increases in abundance but when oxygen becomes limited they disappear. 4. Oligochaetes (*Limnodrilus* and Pot-tub group) numbers increase becoming dominant. 5. Oligochaetes only.
Deep lakes >20 mg l^{-1} $CaCO_3$ and <20 mg l^{-1} $CaCO_3$ stratified oxygen limited	• *Chironomus* spp. present in low numbers (</=44.44 m^{-2}). • T*anytarsus* spp. may be present in low numbers or absent. • *Procladius* spp. present. • Oligochaetes low in numbers (*Limnodrilus* group and Pot-tub group (<50 m^{-2}))	1. *Chironomus* spp. increase in numbers becoming dominant. 2. *Tanytarsus* spp. initially increases in abundance but when oxygen becomes limited they disappear. 3. Oligochaetes numbers increase becoming dominant.

Table 7.39 Suggested abundance categories representing different trophic status. Lp = *Limnodrilus* group plus *Potamothrix-tubifex* group. Clp = *Chironomus* spp. plus *Limnodrilus* group plus *Potamothrix-tubifex* group.

Chironomus spp.	lp	clp	Status	
0-50	0-50	0-100	high	5
50-250	50-250	100-250	good	4
250-500	250-500	250-500	mod	3
500-1000	500-750	500-1000	bad	2
>1000	>750	>1000	poor	1

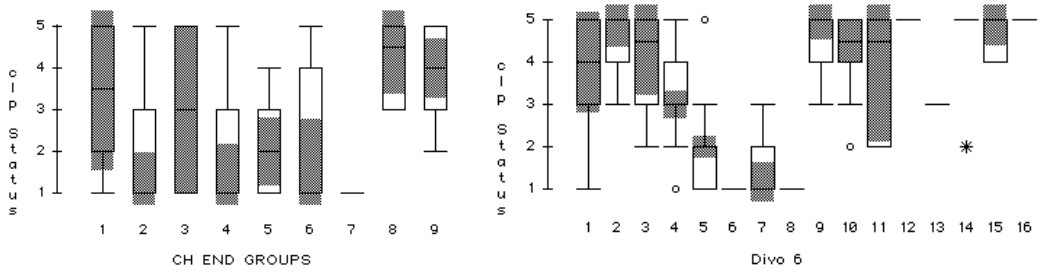
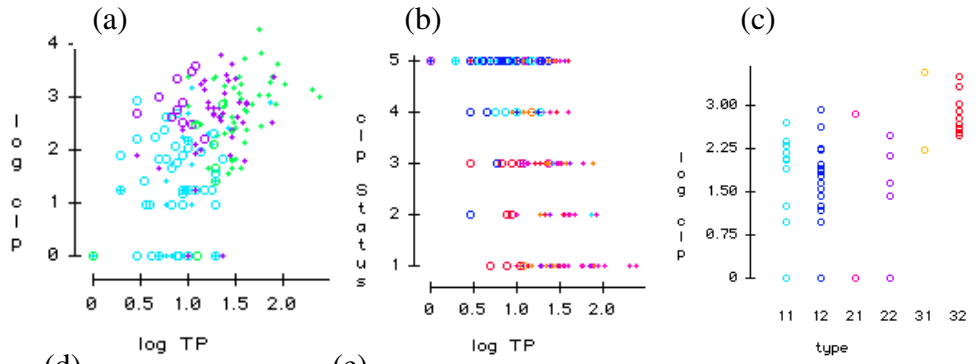


Figure 7.25 The distribution of clp values across chironomid (CH END GROUPS) and oligochaete (DIVO 6) TWINSPAN endgroups.



Regression r^2

	all lakes	shallow lakes
n	174	87
log clp	24.7	35.8
clp status	24.2	35.8
Pearson Product moment, r		
log clp	49.6	53.6
clp status	-50.1	-60.5

Figure 7.26 The relationship between (a) log clp and (b) clp status with log TP is presented. The distribution of (c) clp status values and (d) log clp across lakes types for reference lakes is also presented including some (e) statistics. Lake types are colour coded: 11=light blue, 12 = dark blue, 21=pink, 22=purple, 31=yellow and 32= red (see Table 7.17 for type codes).

7.3.9 Development of a metric based ecological assessment system

It was decided to concentrate on shallow lakes across all alkalinities in developing a multimetric index based on linear metrics. This was done to minimise any stratification related effects. However, there was the possibility that the trends detected were due to alkalinity and not TP because there was no segregation by alkalinity type. Metrics with a linear relationship with TP were identified. These metrics were rescaled from 0 (poor status) to 1 (high status) (Table 7.40) and averaged to develop the profundal index. However, the profundal index was no better than the metric Dlp alone in terms of TP correlation (Table 7.41). The component metrics were also highly correlated which is not desirable. Thus, the development of a system through linear multiple regression did not prove fruitful. Depth adjusted lp/sv had the highest correlation with TP (Table 7.42). With the exception of the number of littoral chironomids, the addition of further metrics did not explain a further 5% in the variation. However, this metric was not strictly linear. The metrics selected to develop a multimetric index following the approach of Karr and Chu (1999) included non-linear metrics and are presented in Table 7.43 with the range of values assigned: 1, 3, or 5. A number of metric combinations were explored. The metric combination with the highest correlation to log TP was selected for presentation (Table 7.43, Figure 7.27).

Of the mixed (chironomid and oligochaete) literature-based indices examined, Quirke 1 had the best correlation with TP (Table 7.44, Figure 7.28). The index Der BQI 2 had the highest correlation with TP of the chironomid-based indices. However, based on visual inspection, Der BQI 4 was considered a better discriminator of chironomid TWINSPAN groups. The index IV 5 had the highest correlation with TP of the oligochaete based indices. Plotting Der BQI 4 against IV 5 would give an overall impression of both profundal components (Figure 7.29). Both these indices were developed as generalised systems across all types, the effect of which was to generate different reference values for each type (Figure 7.30, Figure 7.31). Stable stratifying lakes were not distinguished. This resulted in a wide range in reference values particularly for deep lakes i.e. type 32. These lakes would be expected to have lower Der BQI 4 values in reference condition with possibly similar IV 5 values to their non-stratifying counterparts – however, IV 5 values did not suggest that this was the case - . Furthermore, both indices identified candidate reference lakes that were

possibly not in reference condition i.e. divergent lakes in types 21 (Macnean) and 31 (Annaghmore) and an outlier (Fin Mo) in type 12. Table 7.45 details the reference values for all metrics used in developing a multimetric system and for the indices presented.

Table 7.40 Scaling of transformed metrics that had a log linear relationship with TP. See Figure 7.27 for transformations.

Scale/Profundal Index	%Tubificidae	Ch-cPT	Dlp	o/c	%eutrophic sp.
1	<1	>0.9	<0.5	<0.1	<1
0.9	1-2	0.8-0.9	0.5 – 0.75	0.1-0.2	1-2
0.8	2-3	0.7-0.8	0.75 - 1	0.2-0.3	2-3
0.7	3-4	0.6-0.7	1- 1.25	0.3-0.4	3-4
0.6	4-5	0.5-0.6	1.25 - 1.5	0.4-0.5	4-5
0.5	5-6	0.4-0.5	1.5 - 1.75	0.5-0.6	5-6
0.4	6-7	0.3-0.4	1.75 - 2	0.6-0.7	6-7
0.3	7-8	0.2-0.3	2-2.25	0.7-0.8	7-8
0.2	8-9	0.1-0.2	2.25 - 3	0.8-0.9	8-9
0.1	>9	<0.1	>3	>0.9	>9

Table 7.41 Pearson Product-Moment Correlation for the Profundal Index and its components (transformed) for shallow lakes.

	Profundal Index	O/C	log Dlp	ch-cPT	%Tubificidae	%eutrophic sp	log TP
Profundal Index	1						
O/C	-0.914	1					
log Dlp	-0.899	0.77	1				
ch-cPT	0.836	-0.68	-0.66	1			
%Tubificidae	-0.938	0.873	0.829	-0.663	1		
%eutrophic sp	-0.948	0.809	0.822	-0.779	0.907	1	
log TP	-0.634	0.584	0.637	-0.521	0.577	0.582	1

Table 7.42 The relationship between the metric Dlp/sv and log TP for shallow moderate to high alkalinity lakes. Two lakes were omitted from the analysis – Egish and Dromore.

Dependent variable is:	log TP			
cases selected according to	shallow –Egish & Dromore			
174 total cases of which 91 are missing				
R squared = 46.0% R squared (adjusted) = 45.4% (31.3% for moderate-high alkalinities all depths)				
s = 0.2637 with 83 - 2 = 81 degrees of freedom				
Source	Sum of Squares	df	Mean Square	F-ratio
Regression	4.80459	1	4.80459	69.1
Residual	5.63116	81	0.069521	
Variable	Coefficient	s.e. of Coeff	t-ratio	prob
Constant	0.987609	0.04981	19.8	≤ 0.0001
Dlp/sv	0.269751	0.03245	8.31	≤ 0.0001

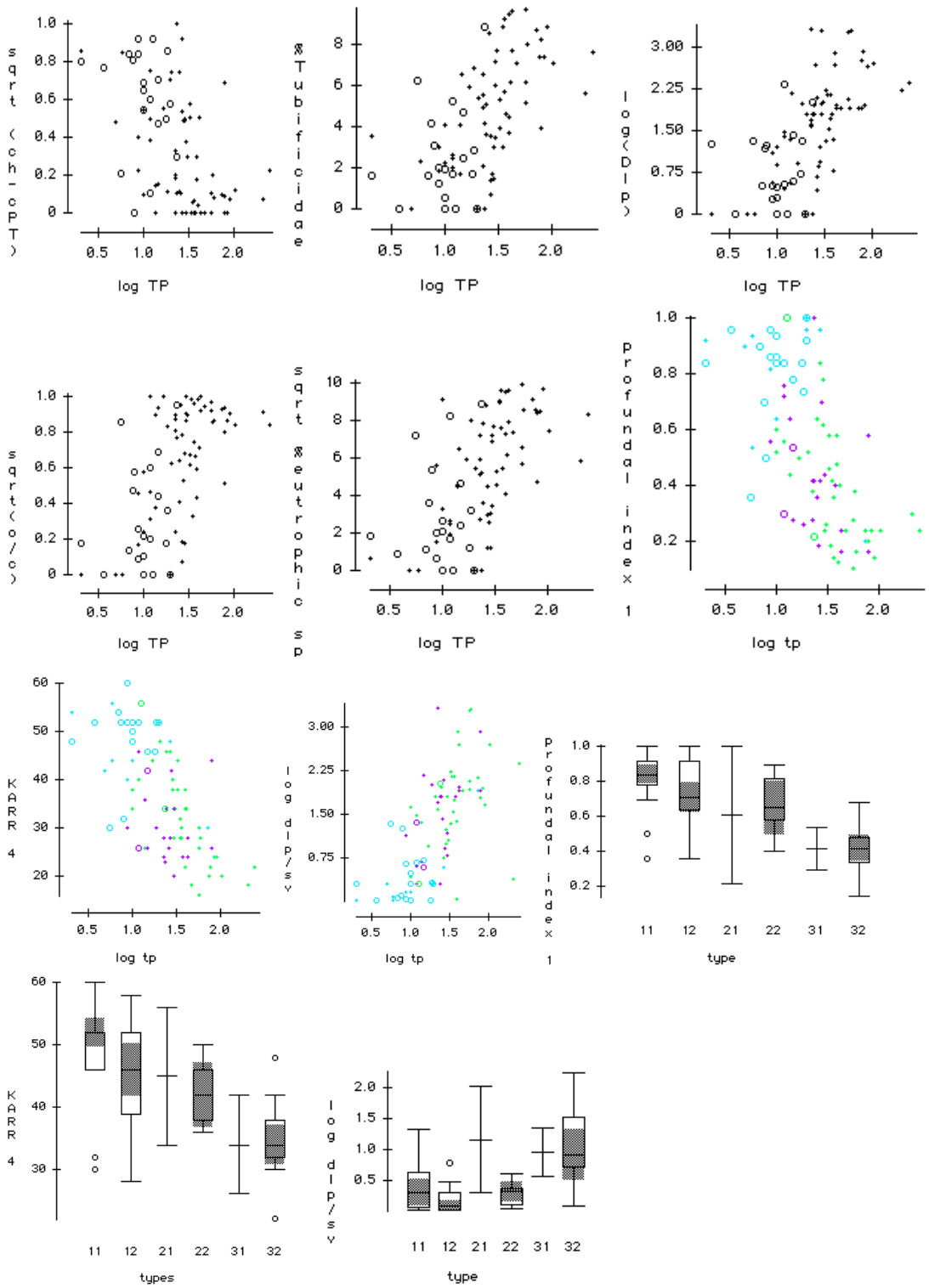


Figure 7.27 The relationship between Log TP and the metrics: ch-cPT (square root transformed) %Tubificidae (square root transformed), Dlp (log transformed) o/c (square root transformed) and %eutrophic sp. (square root transformed) used in constructing the profundal index and the index itself are presented, along with the Karr based index: Karr 4 and the metric with the best regression for all shallow lakes: Dlp/sv. Reference lakes are highlighted by o. The three alkalinity bands: low = blue, moderate = green and high = purple are highlighted for the relationship plots of the Profundal Index, Karr 4, dlp/sv with log TP.

Table 7.43 The list of metrics and combinations used to develop a metric system i.e. indices Karr 1 to Karr 10, following Karr and Chu (1999). The Pearson Product-Moment Correlation for the Karr based indices (shallow lakes only) and Log TP are also presented. Values of 1 (indicative of impact) 3 or 5 (relatively unimpacted) were assigned to the band of values for each metric as shown.

Metrics	Karr 1	Karr 2	Karr 3	Karr 4	Karr 5	Karr 6	Karr 7	Karr 8	Karr 9	Karr 10	1	3	5
no of chironomid taxa*	√	√	√	√	√						0	1-4	>4
Chironomus abundance	√	√	√	√	√				√	√	>500	1-500	=0
Cryptochironomus abundance	√	√	√	√	√					√	>0		0
Tanytarsus abundance	√	√	√	√	√						>100	>50-100	<50
Procladius abundance	√	√	√	√	√						>100	>50-100	<50
Lp abundance	√				√		√				<50	50-250	>250
Dlp		√	√	√		√		√	√	√	>40	10-40	<10
other chironomids abundance				√		√	√	√		√	0	1-100	>100
Eutrophic taxa dominance 2			√	√	√	√	√	√	√		>0.6	0.2 - 0.6	<0.2
ch -cpt				√					√		<0.3	0.3 - 0.6	>0.6
ratio limnodrilus/olgic+chironminae				√							>0.6	0.2 - 0.6	<0.2
o/c				√							>0.6	0.2 - 0.6	<0.2
ratio ccrtp:others				√				√			>100	3-100	<3
log TP	-0.511	-0.532	-0.587	**-0.647**	-0.582	-0.566	-0.535	-0.608	-0.612	-0.563			

Table 7.44 Pearson product moment correlations of literature based indices with log TP.

Oligochaete based		Chironomid based		Mixed	
IV 5, n=174	0.556	Chir BQI, n=168	-0.139	Quirke 1, n=174	0.528
IV 4, n=174	0.514	Der BQI 1, n=168	-0.279	Quirke 2, n=174	0.509
IV 3, n=174	0.519	Der BQI 2, n=168	-0.299		
BQI, n=148	-0.427	Der BQI 3, n=168	-0.128		
EI - HS, n=139	0.444	Der BQI 4, n=168	-0.28		
Mod EI M, n=148	0.477				

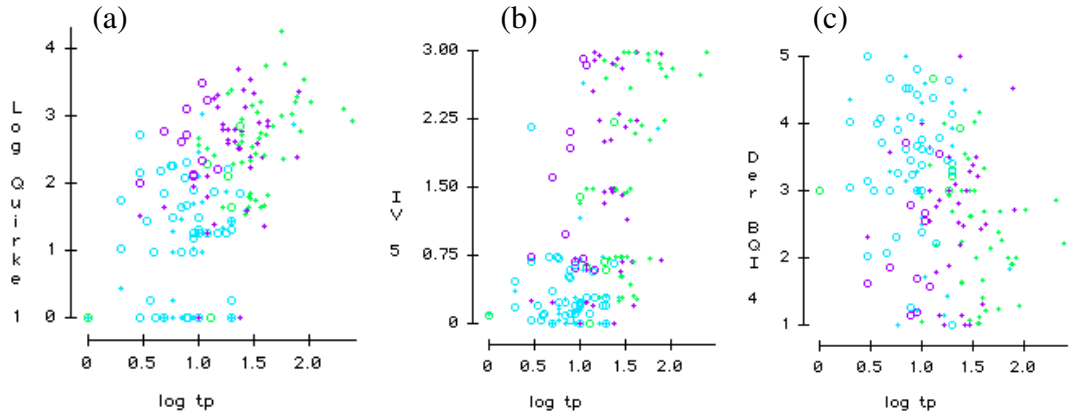


Figure 7.28 The relationship between log TP and the (a) Quirke 1 index (n=174), (b) IV 5 oligochaete index (n=174) and (c) the Der BQI 4 for chironomids (n=158). The three alkalinity bands: low = blue, moderate = green and high = purple and reference lakes (o) are also highlighted.

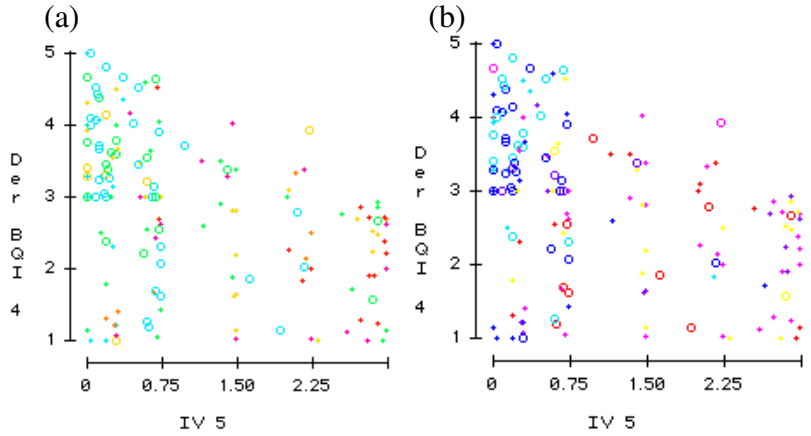


Figure 7.29 The relationship between the chironomid based index Der BQI 4 and the oligochaete based index IV 5 for reference lakes and all lakes (n=158) with (a) TP categories colour coded as light blue, green, yellow, pink, orange, red, in increments of 10 µg l^{-1} as far as 50 µg l^{-1}; the last category covers all values exceeding 60 µg l^{-1}; and (b) types are colour coded as 11=light blue, 12 = dark blue, 21=pink, 22=purple, 31=yellow and 32= red (see Table 7.17 for type codes). Reference lakes are highlighted by o.

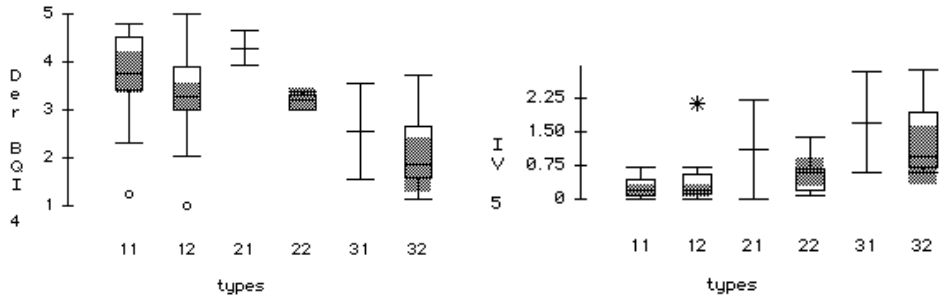


Figure 7.30 Boxplots of the relationship between the chironomid based index Der BQI 4 and the oligochaete based index IV 5 for reference lakes of types 11, 12, 21, 22, 31 and 32 (see table Table 7.17 for type codes). *=outlier.

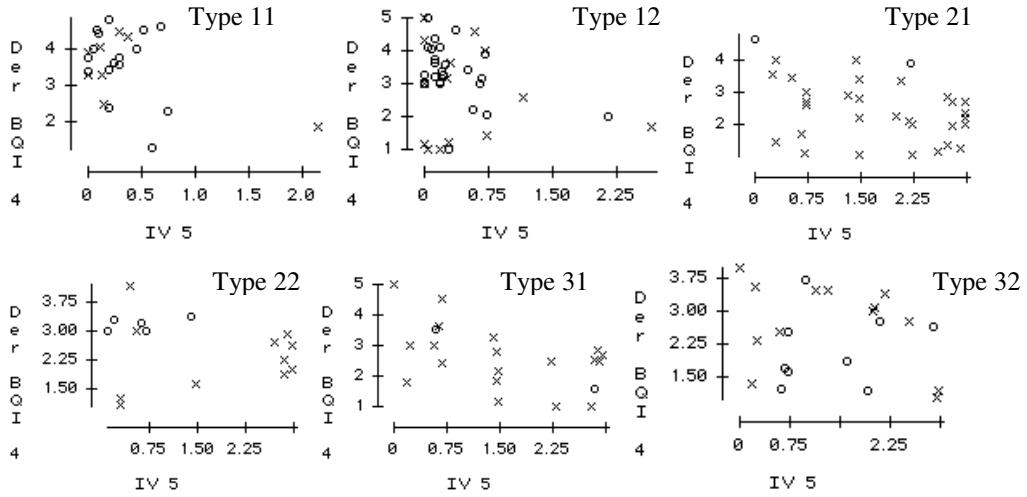


Figure 7.31 The relationship between the oligochaete based index IV 5 and the chironomid based index Der BQI 4 for types 11, 12, 21, 22, 31 and 32 with reference lakes highlighted by o (see Table 7.17 for type codes).

184 lakes were classified into 8 trophic categories and 2 tentative classifications (Table 7.46) using Saether's (1979) key for lake trophic classification based on chironomid composition. The majority of lakes were classified as eutrophic, including the majority of the reference lakes. This was because some key species were absent or rare (*Heterotrissocladius* sp.) and the lack of taxonomic resolution to species level e.g. *Chironomus* spp. limited the application of the classification system.

Table 7.45 The mean and standard deviations for metrics used in developing the Profundal Index and Karr 4 Index are presented along with the literature based indices: BQI 1, IV 5 and Quirke 1 for potential reference lakes.

Lake type		11	12	21	22	31	32
N		17	26	2	6	2	9
log Quirke Index 1	Mean	1.23	1.18	1.43	1.25	2.73	2.60
	StdDev	0.79	0.87	2.02	1.02	0.74	0.50
Karr 4	Mean	48.75	45.50	45.00	42.33	34.00	35.11
	StdDev	7.62	7.44	15.56	5.28	11.31	6.64
Der BQI 4	Mean	3.67	3.44	4.31	3.06	2.57	2.15
	StdDev	0.96	0.88	0.52	0.36	1.40	0.85
IV 5	Mean	0.34	0.41	2.22	0.38	1.72	1.37
	StdDev	0.23	0.45	•	0.29	1.59	0.81
Profundal Index 1	Mean	0.81	0.72	0.61	0.66	0.42	0.42
	StdDev	0.17	0.17	0.55	0.18	0.17	0.16
o/c	Mean	0.12	0.27	0.46	0.35	0.42	0.50
	StdDev	0.19	0.27	0.64	0.36	0.08	0.34
Dlp	Mean	6.27	2.97	53.59	4.14	122.15	35.84
	StdDev	7.70	6.99	75.79	5.11	136.25	54.67
chir -cPT	Mean	0.48	0.28	0.47	0.12	0.12	0.05
	StdDev	0.25	0.25	0.54	0.08	0.15	0.08
%Tubificidae	Mean	6.01	12.85	39.81	12.05	24.98	33.96
	StdDev	9.69	14.58	56.29	14.57	4.09	25.01
%eutrophic sp	Mean	8.14	12.49	39.81	12.49	45.30	58.12
	StdDev	13.58	15.81	56.29	13.68	32.82	22.69

Table 7.46 The classification of lakes using Saether (1980) and the mean and median TP values along with the average % composition of indicator taxa for each trophic category. Group 2 = the summed percentage composition of *Micropsect*ra spp., *Paracladopelm*a spp., *Heterotrissocladius* spp. and *Protanypus* spp.

	β- oligotrophic	γ- oligotrophic	ε- oligotrophic	ζ- oligotrophic ? *	ι - mesotrophic ?**	λ-eutrophic	μ-eutrophic	ν/ξ-eutrophic	o-eutrophic	χ-eutrophic
% Composition										
Sum group 2	2.5	21.7	19.82	2.83	3.70	0.04	0	0	0	1.49
Chironomus	0	0	17.82	0.00	59.26	15.39	33.54	54.93	0	2.05
tribe chironomini	0	19.8	27.70	21.92	59.26	34.91	54.18	54.93	0	10.77
Cryptochironomus	0	2.61	2.74	0.00	0.00	1.26	3.32	0	0	0.29
tribe chrinomini + tanytarsus+tanypodinae	12.5	64.5	69.15	57.75	96.30	79.56	99.95	100	0	77.22
Tanytarsus	7.5	12.8	13.45	19.46	7.41	14.05	17.01	0	0	20.44
Heterotrissocladius	0	1.6	2.38	0.00	0	0	0	0	0	0.28
TP statistics										
Mean TP	21.00	14.22	21.57	18.29	19.00	19.86	28.64	26.81	13.67	54.20
Median TP	21	12	19	18	19	12	20	20	20	15
No. of lakes	1	17	7	4	1	67	58	21	3	5
No. of reference lakes	0	6	0	1	0	30	20	4	1	3
No. of reference lakes per type:										
11		1				11	2	1	1	1
12		3		1		10	9	3		1
21		1				1				
22						4	3			
31						1				1
32		1				3	6			

7.4 Discussion

A typology has been developed using the profundal macroinvertebrates that is as statistically valid and has similar discriminatory powers as the System A typology of the Water Framework Directive. The devised typology has 4 types based on 2 alkalinity bands: moderate to high (> 20 mg l^{-1} $CaCO_3$) and low (< 20 mg l^{-1} $CaCO_3$) and 2 depth categories: deep (>12 m max. depth) and shallow (<12 m max. depth). A tentative third alkalinity band; 20 –100 mg l^{-1} $CaCO_3$ was implied, but no clear evidence was found. Depth has long been recognised as influencing profundal community composition (Heiri, 2004; Brodersen & Anderson, 2002; Johnson & Wiederholm, 1989). Verneaux and Aleya (1998) examined the bathymetric distribution of the chironomids. The controlling factors for the lakes with the greater depth-related change in taxa were thought to be length of anoxic conditions and type and source of food. Alkalinity has been highlighted as influencing chironomid community structure (Ruse, 2002; Real et al., 2000; Pinder and Morley, 1995), but has rarely been highlighted as influencing oligochaete community structure. It is possible that alkalinity is a surrogate for other factors such as substrate type or food availability / primary production. Other factors influencing oligochaete development, growth, reproduction, distribution and consequently community structure include temperature, dissolved oxygen regime, lake morphometry, substrate (in particular particle size and organic content), food availability, species interactions and predation by fish and the invertebrates; *Chironomus* a*nthracinus*, *Procladius* spp. and *Chaoborus* spp. (Reynoldson, 1987, 1990,; Lauritsen *et al.*, 1985; Chapman *et al.,* 1982; Poddubnaya, 1980; Särkkä, 1978; Jónasson and Thorhauge, 1976).

Classification of the individual profundal components provided useful information on their response to eutrophication and highlighted the synergistic influence of nutrient status, depth and stratification. It also aided the description of reference community and deviation from reference caused by eutrophication and aided the further development of indices.

The description of the reference community was possible for shallow low alkalinity lakes. There were insufficient lakes to describe the shallow moderate to high alkalinity type. Therefore their reference community could only be inferred from shallow low alkalinity lakes and the classification of the chironomid and oligochaete

data. *Polypedilum* spp., was certainly a candidate indicator taxa for this type. *Pagastiella* spp. and Pentaneurini may be candidate indicator taxa but they seemed to be confined to the low alkalinity lakes.

There were sufficient lakes to describe both the low and moderate to high alkalinity deep lake types. However, classification of the chironomid component for moderate to high alkalinity lakes showed that stratification influenced composition and abundance. If the selected candidate reference lakes were typical of their type, in particular the moderate to high alkalinity deep type lakes, then the presence of the chironomid taxa *Protanypus* spp. and an absence of *Chironomus* spp. should have characterised the type. This was not the case. These lakes all stratified and their macroinvertebrate composition and indicator species reflected this. *Chironomus* spp., *Procladius* spp., *Tanytarsus* spp., along with the oligochaetes, *Limnodrilus* group and *Potamothrix-tubifex* group, were all key indicator taxa. *Protanypus* spp. were absent. The lack of indicator taxa for the low alkalinity deep lakes could be attributed to the fact that there was a mix of stratifying and non-stratifying lakes. Furthermore, there were indications that a number of the candidate reference lakes were not in such condition. This complicated the description of reference condition. It is recommended that further work on describing the influence of stratification on profundal communities is carried out. Following this, stratification may be considered for inclusion in the typology.

All the multimetric systems and indices provided information on the profundal community. However, one of the problems in using profundal communities in developing an assessment system is that, the individual, components can return conflicting status reports. Furthermore, the development of a trophic assessment system for low alkalinity lake types was hampered by a lack of TP gradient. The development of an assessment system across all types was complicated by the interaction between phosphorus concentration and depth. The impact of the same phosphorus concentration is greater on a shallow lake compared to a deeper lake (Reckhow & Chapra, 1983, cited in Lang 1990). This has been accounted for in some studies by adjusting TP or Chl *a* concentrations for depth (Wiederholm, 1980). Alternatively, the metrics used can be adjusted for depth. Comparisons of the outputs of chironomid inferred TP models with those of diatom inferred TP models have

shown both good agreement (Brooks *et al.*, 2001) and poor agreement (Clerk *et al*., 2000). The contrasting results have been attributed to differences in TP availability at the sediment interface compared to the epilimnion due to lake depth. Brooks *et al.* (2001) studied shallow lakes (11m) and Clerk *et al*. (2000) lake with depths to 34 m. The results highlighted the interplay between lake morphometry and the impact of eutrophication (Brooks *et al*., 2001). The influence of stratification, particularly on the chironomids, was another complicating factor.

In broad terms, the combined abundance of *Chironomus* spp. and the oligochaetes, *Limnodrilus* group and *Potamothrix-tubifex* group, gave a good indication of deviation from reference condition and ecological status. This is effectively the Quirke Index (Irvine *et al.* 2001) without the modification for *Chironomus* spp. abundance and omitting *Procladius* spp. Further refinement of this system, such as setting of abundance boundaries to reflect changes in trophic status with consideration to lake type and the influence of stratification, would make this system easier to apply. Little more than counting *Chironomus* spp., *Limnodrilus* group and *Potamothrix-tubifex* group (all were relatively easily to distinguish from other taxa within their group) would be involved. However, using this method ignores taxa which may be lost quite early on in the eutrophication process, particularly in naturally nutrient poor lakes. It also ignores all other taxa including key characteristic taxa.

Premazzi and Chiaudani (1992) reviewed oligochaete and chironomid based trophic indices including Saether's (1979) key, all of which were investigated in this study. The disadvantages cited in the review were that the assessment of the ecosystem was incomplete because only one component of the profundal fauna was being examined, indices were study specific and therefore were not necessarily applicable on a broader scale, the taxonomy of oligochaetes and chironomids was difficult (in particular the identification of immature worms and larval chironomids) and trophic status was not often quantified.

It is suggested that a system based on the oligochaete index: IV5 (based on Milbrink, 1980) and the chironomid based Der BQI 4 (based on Wiederholm, 1980) is developed further with due consideration to type. Firstly, it gave a better overall view

of the profundal community by considering both components. Suggested improvements would be to modify the indices to reflect reference conditions with an emphasis on each lake type namely by including relevant taxa only, by adjusting the abundance levels in IV 5 and by introducing a better reflection of *Chironomus* spp. abundance in the Der BQI 4 index. However, it might remain restricted in application by sampling method and study area.

8. An overall lake typology for Ireland

It was apparent from the preceding chapters that the typologies based on the analysis of the individual biological elements had defining parameters in common, with similar bands (Table 8.1, Figure 8.1). This facilitates the construction of an overall lake typology for the island of Ireland (Ecoregion 17).

For each of the System B parameters: altitude, latitude and longitude, one category only is proposed. Most lakes in Ireland lie below 200 m. There were insufficient data on lakes located above 200 m to allow a statistical analysis to be carried out and there was little evidence from the classification of the existing data for an altitude influence (Figure 8.1). Both latitude and longitude have narrow bands due to the island's size and therefore would have little influence.

The additional parameters selected for the overall typology were: alkalinity, depth and lake area. Their selection and order of importance were supported by statistical analysis (Table 8.1). Alkalinity or the correlated surrogate variables: conductivity and % peat, were identified by CVA of the biological elements as the main parameters responsible for discriminating between biological endgroups. Depth was frequently selected as the second most important factor and lake area was found to significantly influence macrophytes (Table 8.1).

At least two alkalinity categories; <20 mg l^{-1} $CaCO_3$ and >100 mg l^{-1} $CaCO_3$; were evident for each biological element, with particularly strong evidence for the former (Table 8.2). There were few reference lakes in the moderate alkalinity category, (20-100 mg l^{-1} $CaCO_3$), consequently this is to be considered a tentative category. Further classification of profundal data (including non-reference lakes) suggested that the types: 20-100 mg l^{-1} $CaCO_3$ and >100 mg l^{-1} $CaCO_3$ may be amalgamated. Depth was clearly a typing parameter for three of the elements with particularly good agreement on boundaries at a mean depth of 4 m (Table 8.2). In the case of the profundal invertebrates, maximum (station) depth (12 m) was equated to mean depth (4 m) using the equation: y=0.314X derived from data in Irvine *et al.* (2001). The

boundaries for lake area were based on the macrophyte endgroups. However, Figure 8.1 suggests that area may also have influenced the littoral and profundal macroinvertebrates, although the development of a typology for each of these elements did not indicate area as a strong typing parameter. Three alkalinity types, two depth types and two area types were recognised. These gave rise to 12 lake types (Table 8.3).

It is suggested that further work be carried out to investigate the possible inclusion of other typology parameters such as stratification, residence time and colour, to better define marl lakes and to incorporate turloughs and high altitude (mountain) lakes (which were not included in the selection of candidate reference lakes) in the typology.

Table 8.1 For each biological element, the significant parameters identified by CVA as discriminating between endgroups are presented. Parameters are numbered in order of importance i.e. variation explained. Shaded cells indicate that a parameter was not used in the original analysis.

			Conductivity	Alkalinity	Colour	TP	Altitude	Lake Area	Lake Perimeter	Depth	Catchment/lake are	% Forestry	% Peat
Biological Element		**CVA- forward selection**											
Phytoplankton				1	4		2				3		
Macrophytes				1				3		2			
Invertebrates	Littoral	cluster endgroups	2	1									3
		TWINSPAN endgroups											1
	Profundal	dataset 1				3		4	5	2		6	1
		dateset 2	1		3		4	6	5	2			

Table 8.2 Boundary settings for the typology parameters (indicated by colour changes) based on the classification of candidate reference lakes for the biological elements.

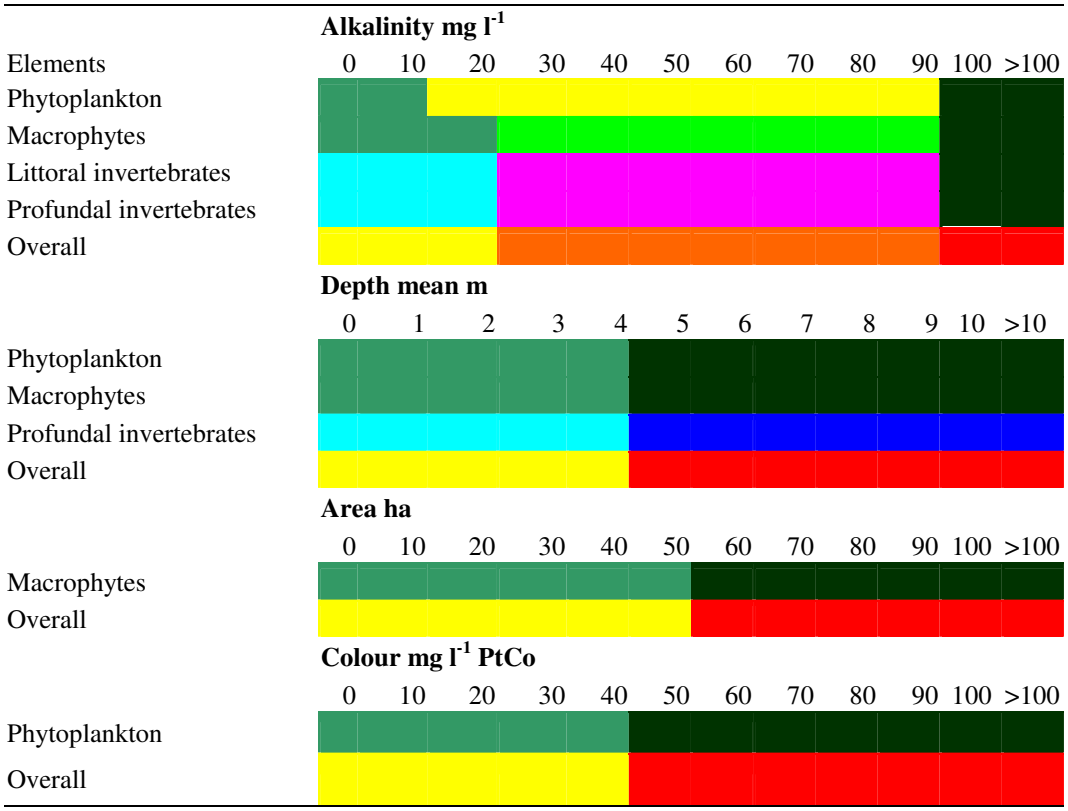


Table 8.3 The overall typology for Irish lakes.

Parameters	**Boundaries**											
Alkalinity (mg l^{-1} $CaCO_3$)	< 20				20 - 100				> 100			
Depth (m)	< 4		> 4		< 4		> 4		< 4		> 4	
Area (ha)	< 50	> 50	< 50	> 50	< 50	> 50	< 50	> 50	< 50	> 50	< 50	> 50
Type	1	2	3	4	5	6	7	8	9	10	11	12

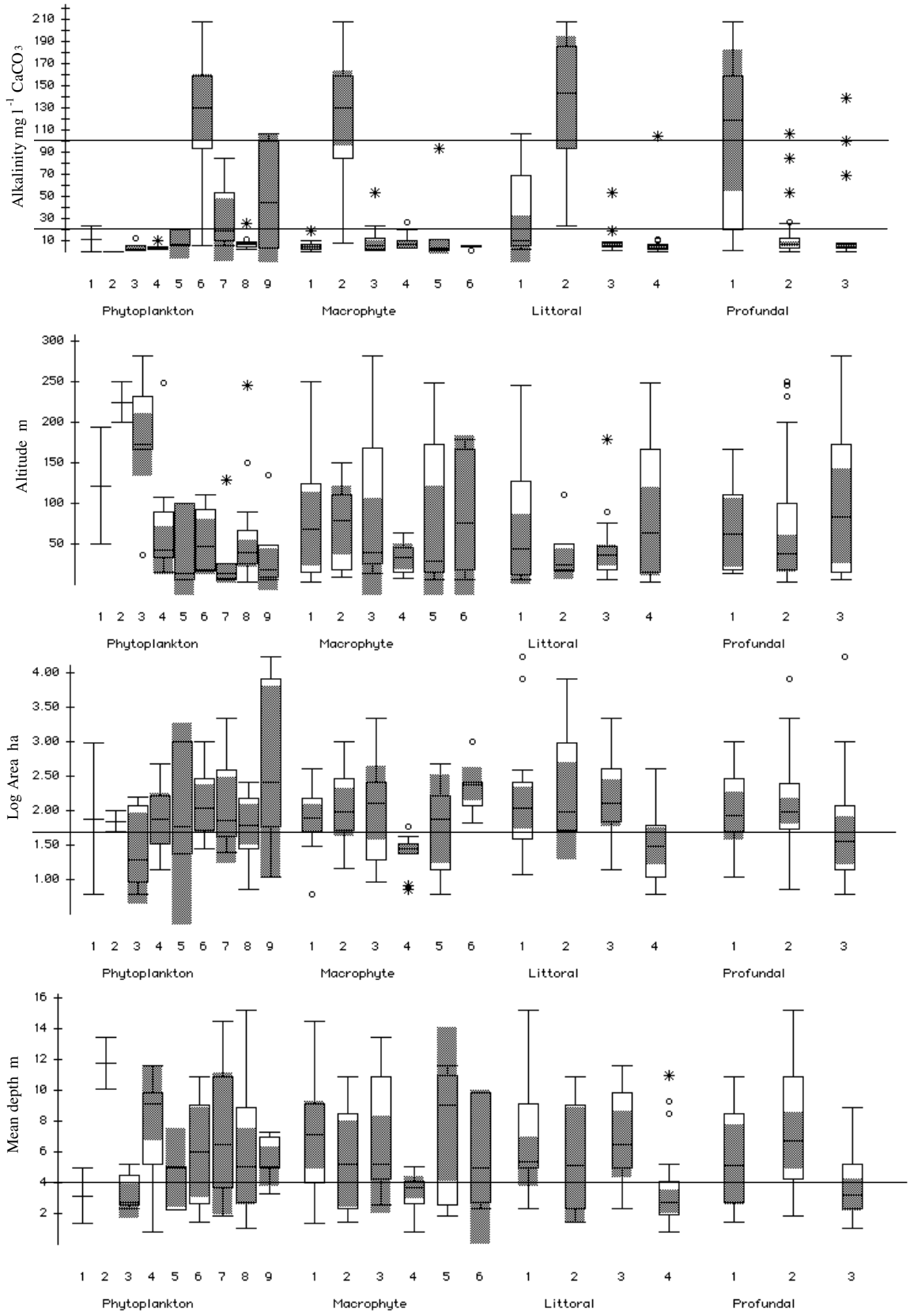


Figure 8.1 Boxplots of typology parameters by classification endgroups of phytoplankton, macrophytes, littoral macroinvertebrates and profundal macroinvertebrates. The parameter boundaries for the overall typology are highlighted by horizontal lines.

9. Defining ecological quality classes

9.1 Introduction

The preceding chapters also focused on the development of ecological assessment tools. This took the form of trying to establish relationships between ecological metrics and a pressure gradient – total phosphorus (TP). The establishment of good relationships between ecological metrics and pressure gradients gives confidence in their ability to detect the influence of pressure. However, the next task is then to divide such ecological assessment metrics up, based on deviation away from reference status, into ecological quality classes. The Water Framework Directive requires that ecological quality is expressed as one of five classes: High, Good, Moderate, Poor or Bad status. These classes cannot be defined by an arbitrary division of an ecological assessment metric but must conform to the definitions as given in the Water Framework Directive (Table 9.1).

The ecological assessment metrics tested in this study had linear or log-linear relationships with pressure. This expresses ecological change as a continuum with little indication of 'breakpoints' that might indicate a suitable place to position a boundary between, for example, high and good status or good and moderate status. The purpose of this chapter is to examine the response of certain metrics to pressure to identify levels of pressure that result in ecological change which can clearly be related to the status classes as defined by the WFD (Table 9.1). Ultimately, such boundaries will be adjusted through an intercalibration exercise with EU countries in order to ensure comparability between member states (CEC, 2000).

9.2 Results

Increased input of total phosphorus was considered to be the main pressure affecting lakes in the Republic of Ireland. Ecologically relevant boundaries for TP concentrations were initially chosen based on the response of macrophyte diversity to TP. Figure 9.1 shows a unimodal relationship between Simpson's diversity index and transformed (log(x + 1)) TP for lakes with an alkalinity greater than 20 mg l^{-1} $CaCO_3$. Four bands of <10, 10-25, 25-70 and > 70 µg l^{-1} TP were selected to correspond to points of change in diversity with TP. The good/moderate boundary was taken to be 25 µg l^{-1} TP on the basis that it corresponds with normative definitions i.e. it is the

point where diversity starts to decrease therefore resulting in an 'undesirable disturbance to the balance of organisms'. The increase in diversity between 10 and 25 μg l^{-1} TP may correspond to normative definitions of good status in that the change in not an 'undesirable' one (Table 9.1).

The TP bands above were examined for further relevance in two sub-sets of lakes in alkalinity bands of the proposed typology that are most subject to pressure: 20 – 100 and > 100 mg l^{-1} $CaCO_3$. In the alkalinity band 20 – 100 mg l^{-1} $CaCO_3$, the 25 μg l^{-1} TP boundary was marked by an increased loss of littoral rosette species including *Littorella uniflora*, an increase in the relative frequency (RF) of Nymphaeids (rooted macrophytes with floating leaves) and a marked reduction in the RF of Charophytes (Figure 9.2). Phytoplankton showed a more linear response to TP although *Pediastrum* spp. only occurred in significant numbers at > 25 μg l^{-1} TP (Figure 9.2) indicating that it may be a more useful indicator over that part of the TP gradient.

In lakes with an alkalinity > 100 mg l^{-1} $CaCO_3$ (typically marl-precipitating lakes) the RF of *Chara* spp. was typically > 40% at TP concentrations below 10 μg l^{-1}(Figure 9.3). The RF of *Chara* spp. declined rapidly at higher TP concentrations from 10 – 25 μg l^{-1}. The RF of Lemnids and the percentage of taxa occurring with filamentous algae also increased after 10 μg l^{-1} TP. As found in the intermediate alkalinity band, *Pediastrum* spp. typically only occurred in significant numbers at > 25 μg l^{-1} TP. *Cryptomonas* and *Scenedesmus* spp. displayed an unclear relationship with TP in this alkalinity band.

Table 9.1 Definitions for high, good and moderate ecological status in lakes for phytoplankton, macrophytes and phytobenthos and benthic invertebrate fauna (CEC, 2000).

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.
Benthic invertebrate fauna	The taxonomic composition and abundance correspond totally or nearly totally to the undisturbed conditions. The ratio of disturbance sensitive taxa to insensitive taxa shows no signs of alteration from undisturbed levels. The level of diversity of invertebrate taxa shows no sign of alteration from undisturbed levels.	There are slight changes in the composition and abundance of invertebrate taxa compared to the type-specific communities. The ratio of disturbance sensitive taxa to insensitive taxa shows slight signs of alteration from type-specific levels. The level of diversity of invertebrate taxa shows slight signs of alteration from type-specific levels.	The composition and abundance of invertebrate taxa differ moderately from the type-specific conditions. 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.

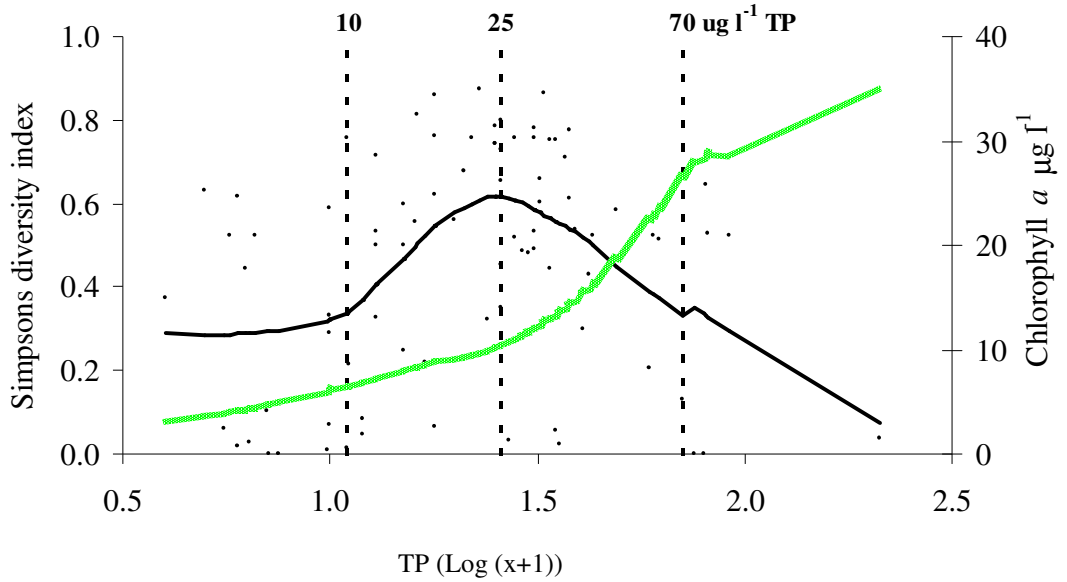


Figure 9.1 Selection of TP bands (**- - -**) based on the lowess smoothed relationship (—) between Simpson's diversity index for the macrophytes samples and transformed TP (Log (x+1)). Smoothed relationship of chlorophyll *a* with transformed TP (Log (x+1)) is overlain (—). Graph refers to lakes > 20 mg l^{-1} $CaCO_3$ only, TP values were mostly measured in Spring.

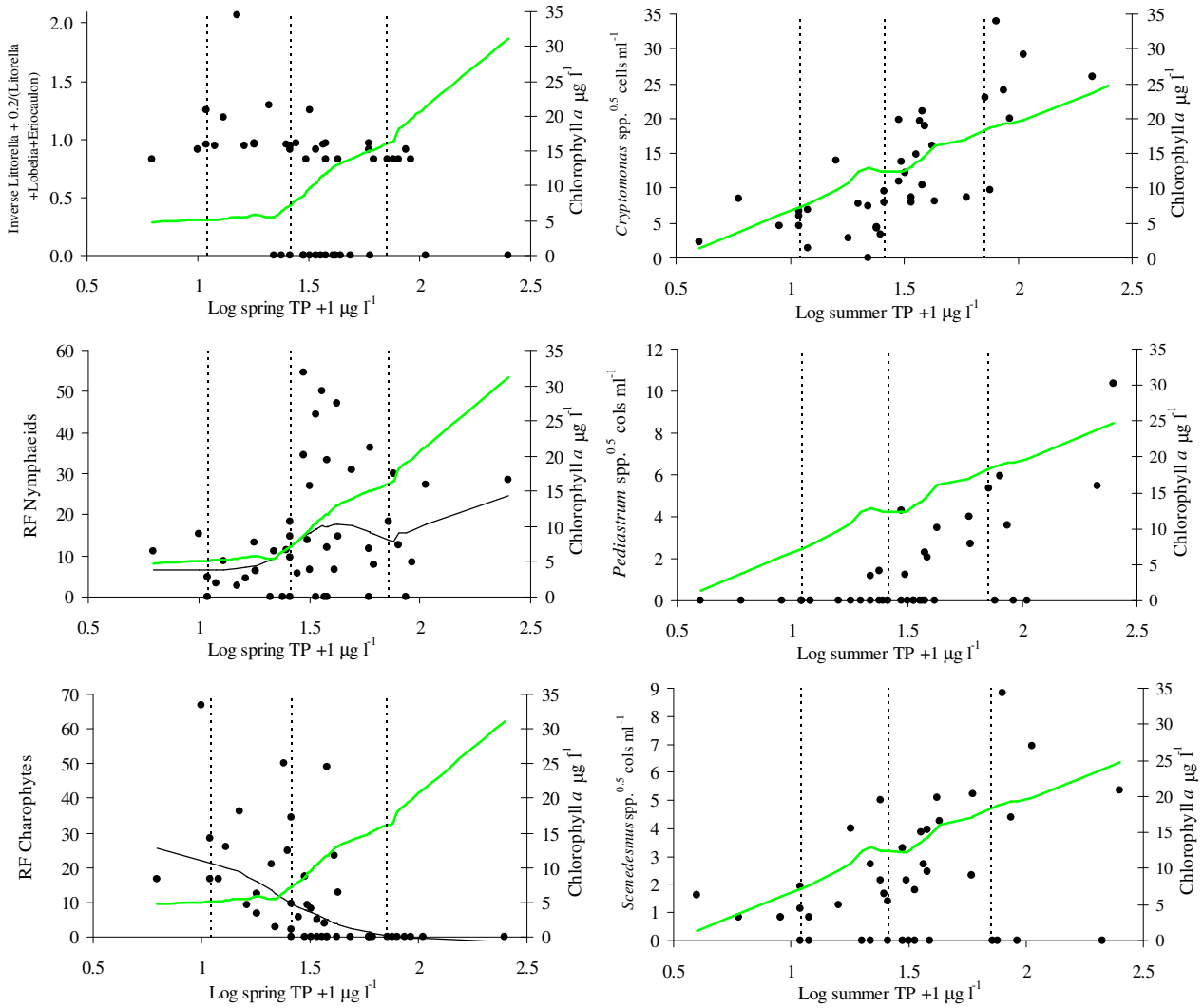


Figure 9.2 Relationship between TP (Spring or Summer) and selected macrophyte metrics (left) and phytoplankton taxa (right) for lakes between 20 and 100 mg l^{-1} $CaCO_3$ alkalinity. The lowess smoothed relationship between TP and summer chlorophyll *a* is overlain (—). Dashed lines represent the proposed boundaries of 10, 25 and 70 µg l^{-1} TP. Continuous black lines represent the lowess smoothed relationship between TP and the metric in question.

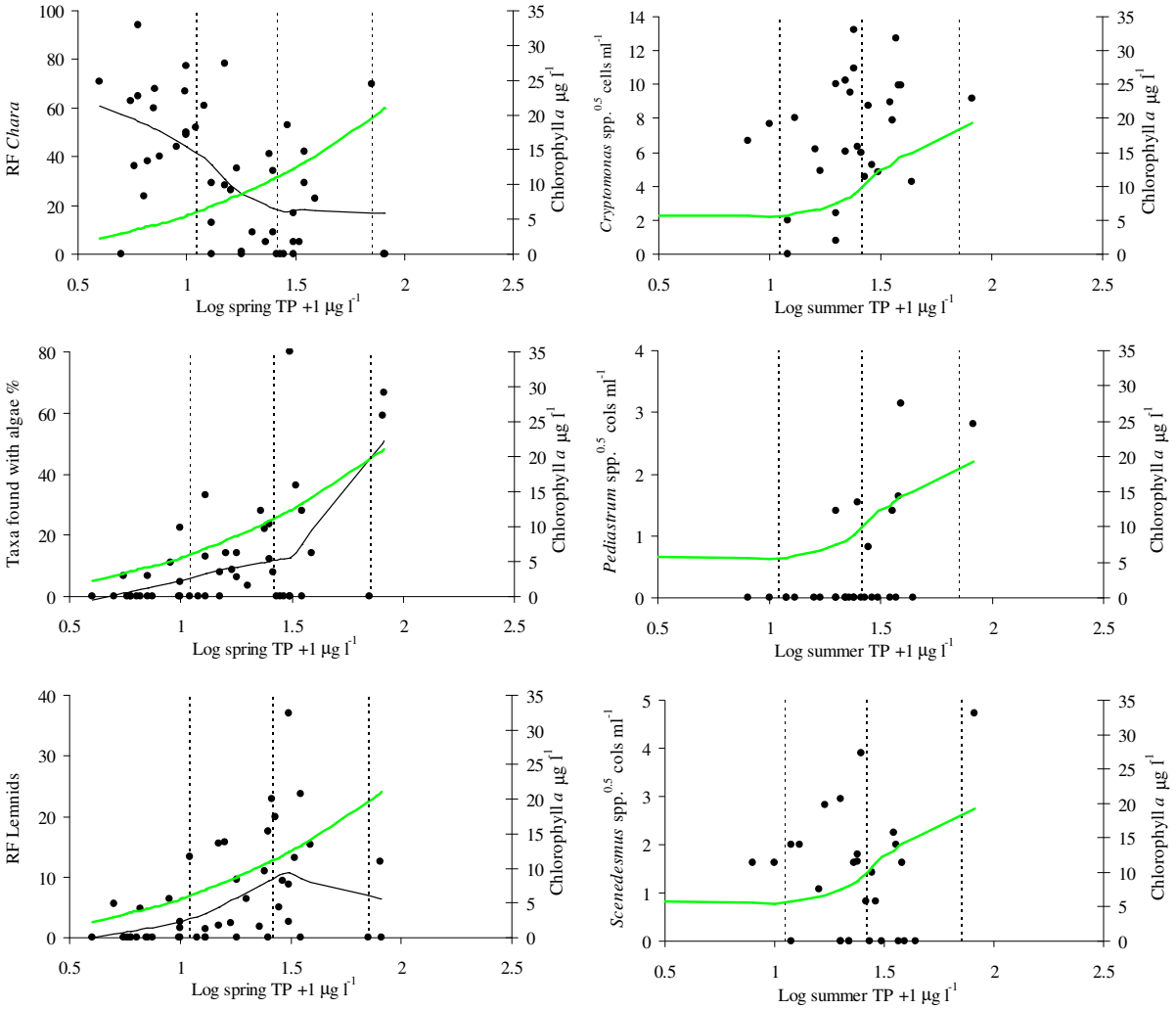


Figure 9.3 Relationship between TP (Spring or Summer) and selected macrophyte metrics (left) and phytoplankton taxa (right) for lakes > 100 mg l^{-1} $CaCO_3$ alkalinity. The lowess smoothed relationship between TP and summer chlorophyll *a* is overlain (—). Dashed lines represent the proposed boundaries of 10, 25 and 70 µg l^{-1} TP. Black continuous lines represent the lowess smoothed relationship between TP and the selected metrics.

An alternative approach may be to examine literature based relationships that might lend support to boundary setting. A series of regression models (Table 9.2), 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.

Figure 9.4 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 µg l^{-1} TP and this appears to be where there is a significant change in slope/response of the depth of macrophyte colonisation to TP concentration for a colour of 30 mg l^{-1} PtCo (Figure 9.4).

The good/moderate boundary was placed at 25 µg l^{-1} TP which corresponded to a point where the depth of colonisation of Charophytes is reduced by 24% from reference condition (assumed to be 10 µg l^{-1} TP). This appears to fit normative definitions (Table 9.1), 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*.

Table 9.2 Models used to generate Figure 9.4. 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* µg l^{-1}	0.92	1.449 log TP µg 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* µg l^{-1}
3	Zc Angiosperms$^{0.5}$		1.33 log Secchi depth + 1.4
4	Log Zc Charophyta	0.83	1.03 log Secchi depth + 0.18

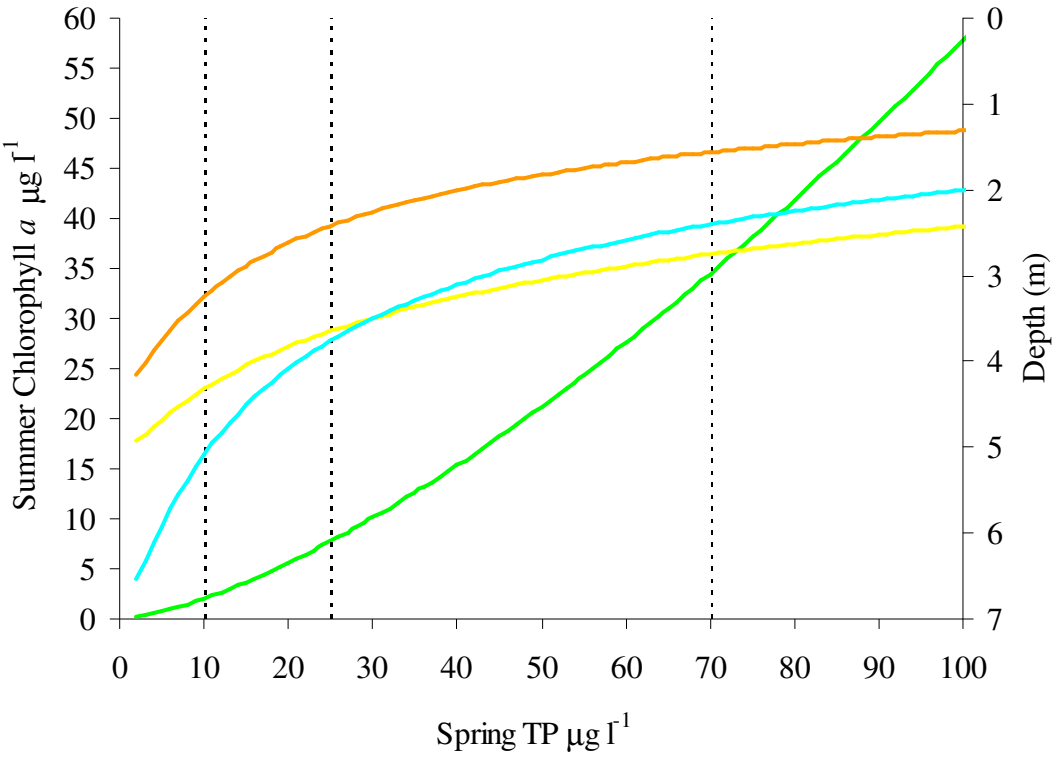


Figure 9.4 Relationship between Spring TP and Summer chlorophyll *a* (—), and predicted Secchi depth (—), predicted depth of colonisation of Charophytes (—) and Angiosperms (—). Sources and models are listed in Table 9.2. Dashed lines represent boundaries of 10, 25 and 70 µg l^{-1} TP.

9.3 Discussion

The Water Framework Directive (WFD) provides only a very generalised description of what constitutes high, good and moderate status for the different ecological quality elements (Table 9.1). It is up to member states to define boundaries to the different status classes in terms of an ecological quality ratio (EQR) that corresponds to their definition as given in the WFD (Table 9.1). The approach explored here was to define boundaries in pressure (TP) that relate to changes in macrophyte diversity as an objective method of defining ecological boundaries. These boundaries were then examined for support using additional macrophyte metrics and phytoplankton taxa.

Once boundaries of pressure (TP) have been assigned, they can be used to select corresponding levels of an assessment metric or EQR. The objective is to separate an EQR into 5 quality classes. This can be done by selecting TP concentrations that correspond to points of ecological change consistent with the normative definitions and then using those TP concentrations to relate the boundaries to an EQR. For example, using 25 $\mu g\ l^{-1}$ TP to select the good/moderate boundary for the macrophyte multimetric produced in Chapter 5 would equate to a macrophyte multimetric value of 0.5. However, such calculations would have to be done on a type-specific basis with an EQR that measures deviation from type-specific reference condition. Thus, e.g., high quality lakes cannot be allowed to change to a good quality status.

The good/moderate boundary is perhaps more important than other boundaries in that member states will have to restore all lakes found to be in moderate status or below to good status by the end of 2015. However, it is also a requirement of the directive that any further deterioration of water quality is prevented.

For the moderate alkalinity lakes (20 – 100 mg l^{-1} $CaCO_3$) the setting of boundaries at 10 $\mu g\ l^{-1}$ TP for the high/good boundary and at 25 $\mu g\ l^{-1}$ for the good/moderate boundary seemed reasonable based on the changes in diversity, increase in Nymphaeids and the tendency of littoral rosette macrophyte taxa to be absent (Figure 9.1, Figure 9.2).

For the high alkalinity lakes (> 100 mg l^{-1} $CaCO_3$), which are predominantly marl-precipitating in the Republic of Ireland, the boundaries suggested by the overall

relationship between macrophyte diversity and TP (Figure 9.1) appeared too high when individual metrics were examined. In particular, there appeared to be a sharp reduction in the relative frequency of *Chara* spp. at TP concentrations above 10 μg l^{-1} (Figure 9.3). Charophytes typically dominate marl lakes in reference condition (Figure 5.4) and owing to their deep depth of colonisation, are sensitive to even small increases in phytoplankton due to the exponential decrease of light with depth. In order to conform with normative definitions for moderate status, the TP boundary for good/moderate status could be placed at 15 μg l^{-1}, which, for *Chara* spp. would correspond to a significant distortion from good quality (Table 9.1, Figure 9.3). A TP concentration of 7 μg l^{-1} may represent the boundary between high and good status, based on the TP distribution of lakes with a high relative frequency of *Chara* spp. (Table 9.3).

Further guidance has been provided on ecological status classification by ECOSTAT (2005) who equate moderate conditions to a situation where "a group of taxa or a species of significant conservation importance normally present at reference conditions is in significant decline." This can clearly be related to the decline of *Chara* spp. in Figure 9.3 which are in significant decline at 15 μg l^{-1} TP.

There was some disagreement between the model run using published equations and the observed response of Charophytes (c.f. Figure 9.3, Figure 9.4). A more rapid decline in the relative frequency of Charophytes with TP was observed than indicated by the published models (Table 9.2, Figure 9.4). However, the outcome of the model is largely dependent on input values (Table 9.2); if they are changed to a colour of 20 mg l^{-1} PtCo (more typical of marl lakes) and a initial reference TP of 5 μg l^{-1}, then a 15 μg l^{-1} TP good/moderate boundary would correspond to a 24% reduction in the depth of Charophytes.

The suggested boundaries must be regarded as preliminary owing to the limited availability of nutrient data for this study. Most of the analysis for the macrophytes used spring TP whereas annual averages would have been preferable. However, as TP shows a sinusoidal annual pattern in Irish lakes - being higher in winter and lower in summer (Gibson *et al*., 1996) - samples taken in the months of April and May tend to

correspond closest to annual averages (Irvine *et al*., 2001). The analysis of phytoplankton used summer TP values (which corresponded to the sample used for phytoplankton analysis), as phytoplankton may be more dependent on current nutrient status.

Table 9.3 Suggested concentrations of TP μg l^{-1} (Spring values) that may correspond to ecological status boundaries.

Status	20-100 mg l^{-1} $CaCO_3$	>100 mg l^{-1} $CaCO_3$ (Marl lakes)
High/Good	10	7
Good/Moderate	25	15
Moderate/Poor	70	
Poor/Bad		

The lower boundaries of TP suggested for the high alkalinity marl lakes take cognisance of sensitivity of charophytes to eutrophication pressure but are also appropriate because of the conservation importance of these lakes (John *et al*., 1982). In addition, lower boundaries of TP are appropriate because natural background nutrient concentrations are likely to be lower in these lakes due to the co-precipitation of phosphorus (Otsuki and Wetzel, 1972) and the lower nutrient inputs associated with groundwater fed rather than surface water fed lakes.

In some respects it is difficult to place boundaries to ecological classes as ecological change is often a continuum with few breakpoints that suggest appropriate places to position a boundary that meets WFD definitions (Table 9.1). However, it is clear from this study that most ecological change takes place at the early stages of eutrophication, and certainly below 25 μg l^{-1} TP. It is a requirement of the WFD that ecological quality boundaries are adjusted through an intercalibration exercise with EU countries in order to ensure comparability between member states (CEC, 2000). This study should help to inform the national position on the relationship between pressure and ecological change. It would be beneficial, however, to broaden the involvement in boundary setting exercises to achieve consensus on what represents high, good and moderate ecological status for Irish lakes.

To some extent, the importance of the positioning of ecological boundaries depends on how the WFD requirement for no further deterioration in ecological quality is viewed. Deterioration may be allowed to continue across a whole ecological class before action is taken. This would be unsatisfactory. It would be more prudent to respond at an earlier stage when an EQR shows signs of initial decline.

10. General Discussion

10.1 Typology

One of the main objectives of the project was to produce a lake typology that is successful in partitioning biological variation in reference condition. Typologies were initially developed for each individual biological element and subsequently combined into an overall typology. The overall typology was described in Chapter 8 and is to be used as the reporting typology for Ireland. Comparison of the typologies developed at individual element level indicated that they were at least as successful as the default typology system of the WFD – system A.

The overall typology presented in this study was developed as a reporting typology. The results of ecological assessment will be expressed as an ecological quality ratio (EQR) and reported through the appropriate lake type. The typology should prove effective in partitioning natural variation and allow ecological change to be detected more accurately. In addition, the typology developed should provide a useful framework to focus research on certain lake types. The typology, especially the alkalinity bands, should broadly reflect contrasts in the Irish environment. The < 20 mg l^{-1} $CaCO_3$ alkalinity band should correspond to lakes in the siliceous uplands that ring the country whereas the > 100 mg l^{-1} $CaCO_3$ alkalinity band should largely encompass those in areas of limestone found mostly in the midlands and County Clare. Lakes in the intervening band (20 - 100 mg l^{-1} $CaCO_3$) may be found in catchments with a mixture of siliceous and limestone rock types and in lakes in the northeast in Cavan and Monaghan based on Silurian and Ordovician strata.

A straightforward use of the typology presented in Chapter 8 would involve the identification of reference lakes in each lake type and the generation of an EQR for each biological element in a lake by dividing the observed metric value by the reference metric value. Final ecological assessment is represented by the lowest EQR recorded for the biological elements and the supporting physical and chemical elements.

Another approach would be to regard the overall typology as a reporting typology only. Ecological assessment could be based on typologies developed separately for

each biological element and an EQR simply reported into the overall typology. This is one of the benefits of the EQR approach, which yields a value from high status (near 1) to bad status (near 0) and theoretically should allow broad interpretation across lake types, water body types (e.g. rivers, lakes, transitional waters) and countries.

The approach taken to develop the typology examined differences in composition and abundance of flora and fauna in potential reference lakes using multivariate techniques. Environmental boundaries to types were then set to partition biological variation in reference condition. An alternative approach would be to develop a typology based on ecological assessment metrics rather than composition and abundance of the flora and fauna. This would entail the examination of both metric variation in reference condition and metric response to pressure for each of the biological elements. Focusing on metrics may be more appropriate as the purpose of a typology is to help detect ecological change by accounting for factors that effect ecology naturally. Developing a typology based on metric variation in reference condition would be likely to lead to fewer types, depending on the metric used. For example, the phytoplankton index appeared to be similar across types of reference lakes and also had low within-type variation (Table 4.9). This may indicate that the phytoplankton index may successfully work across different lake types and that a smaller number of types would be sufficient. The barrier to taking this approach in the current study was that none of the ecological assessment metrics are finalised, so that it would be premature to use them to develop a typology.

One factor that was not directly considered in the development of a typology or ecological assessment system was background nutrient status. Some lake types may be more naturally nutrient rich than others, and may, therefore, appear to be impacted; however, they may be in reference state if their background nutrient status is atypically high. However, it was considered more practical to develop metrics based on a common pressure gradient under the assumption that reference background nutrient concentration was similar for all lakes. It is recommended that research be carried out to estimate the background nutrient status for lakes. A correction factor based on background nutrient status could be applied after the application of an ecological assessment metric. This would be more transparent than developing

assessment systems with integrated, preconceived estimates of background nutrient status.

Other elements that could be considered useful to refine the typology at a later date are information on residence time, more detailed information on stratification and mixing depth, a description of the substrate for macrophytes and invertebrates and a measure of exposure for macrophyte and littoral invertebrate sites. In addition the biological elements: fish and phytobenthos, were not considered in this study but should be incorporated at the earliest opportunity.

The upper alkalinity band to the typology (> 100 mg l^{-1} $CaCO_3$) was generally taken to correspond to marl-precipitating lakes. These lakes are typically situated on limestone and are characterised by their abundance of Charophytes in reference condition. It may be worthwhile to confirm which lakes with > 100 mg l^{-1} $CaCO_3$ are marl-precipitating on a site-by-site basis at a later stage. Some lakes, located in the northeast may have high alkalinity but may not be considered marl lakes, as they are not located on limestone and may be fed by surface rather than ground water. It may be appropriate to consider two subtypes – marl and non-marl lakes – within the > 100 mg l^{-1} $CaCO_3$ lake type. This is important as it was argued in Chapter 9 that marl lakes should have more stringent environmental criteria as they respond to pressure at an earlier stage.

10.2 Ecological assessment

The purpose of ecological assessment is to measure deviation away from reference status along a pressure gradient. A clear relationship must be demonstrated between an ecological metric and pressure before it may be accepted as a useful descriptor of ecological change for a biological quality element. Only preliminary work on developing ecological assessment systems for the biological elements was within the scope of this project.

The success of the ecological assessment metrics developed was found to vary with the biological elements. Some elements such as phytoplankton and macrophytes were clearly more useful in detecting the eutrophication pressure. The phytoplankton index showed a good relationship with total phosphorus (r^2 = 0.67) as would be expected,

given the well-established relationship between indicators of phytoplankton biomass (chlorophyll *a*) and TP (Sakamoto, 1966; Dillon and Rigler, 1974). One drawback is that occasionally higher than expected values of the phytoplankton index may be recorded for a given TP owing to zooplankton grazing and other factors (Chapter 4). In overall assessment terms, this should not prove a problem as it is the lowest EQR of the biological, chemical and physical elements that is used for assessment. The macrophyte multimetric index also had a good relationship with TP (r^2 = 0.59). Macrophytes may show a close relationship with the pressure gradient because they reflect the influence of eutrophication on the light climate over the growing season. In contrast, phytoplankton are more reflective of conditions when sampled and are more affected by nutrient dynamics and grazing. Changes in macrophyte composition are also related to sediment enrichment caused by eutrophication. The multimetric index developed for the macrophytes appears to have achieved similar success in detecting the effects of pressure on ecological quality of the lakes as the long established Q-value system has for river quality in Ireland and should provide a useful tool in ecological assessment (Chapter 5).

The performance of the invertebrate trophic score for littoral macroinvertebrates had limited success (r^2 = 0.43) and needs further development. The response of littoral macroinvertebrates to eutrophication pressure may be complex, perhaps owing to the non-uniform substrate, diverse communities or complex food web. The relationship between profundal invertebrates and pressure was also concluded to require further work. The complicating influence of depth and stratification on the effects of eutrophication may make it difficult to develop an assessment system that can be used in all situations. Thus, some degree of expert judgement may be necessary in the application of ecological assessment metrics for profundal invertebrates.

One approach employed in this study, to help exclude some of the natural variation and allow ecological change along a pressure gradient to be detected more easily was to focus metric development on a certain habitat or season. For example, the phytoplankton metric was developed for summer assemblages because the response of phytoplankton to nutrient enrichment in spring may be complicated by the availability of silica and light. In addition, sampling effort for littoral macroinvertebrates focused on stony shorelines only. Focusing effort like this may be criticised as only

incomplete taxa lists are obtained. However, the objective was to develop a relatively simple and robust ecological assessment tool, an objective that could best be achieved if the amount of inherent natural variation could be controlled. Stratifying sampling effort to partition variability is one method that may be used to increase statistical power (Johnson, 1998). Future work on this could focus on attempting to improve the match of littoral sites sampled for macroinvertebrates or perhaps on using a limited depth range within the sub-littoral and profundal zones to try and reduce the influence of stratification and depth.

Annex II of the WFD allows for a quality element to be excluded from ecological assessment if its natural variability (excluding seasonal variability) prevents reference conditions being established. While success in developing ecological assessment metrics varied between the biological groups, all showed enough promise to justify further research.

Metric development also took place across a varying number of types, from developing metrics for macrophytes for individual lake types using multiple regression (Chapter 5) to developing a metric for all lake types such as the phytoplankton index (Chapter 4). The validity of the particular approach depends on the biological element under consideration and the metrics employed. It may be preferable to initially develop an ecological assessment metric across broad types and then seek to refine it using a more detailed typology.

Many of the metrics were not expressed strictly in WFD terms as deviation away from type specific values of the metric in reference condition. This was a conscious decision as many of the metrics can only be considered in draft form in their current state. In any case, it is relatively simple to rescale metrics based on type specific reference conditions (Chapter 5). It may be simpler from a catchment management point of view to only calculate type specific reference metric values when such metrics are significantly different between types of reference lakes. As previously mentioned, the phytoplankton index (Chapter 4) was found to have very little variation between lake types in reference condition.

One theme common to the work on developing ecological assessment metrics was that most ecological change takes place at an early stage of nutrient enrichment. For example, the littoral macroinvertebrate trophic score (Chapter 6) showed an increase after 15 μg l^{-1} TP. The macrophyte multimetric index (Chapter 5) recorded most ecological deterioration between 10 and 20 μg l^{-1} TP. This, together with the work on defining ecological quality classes in Chapter 9 should help focus attention on appropriate positions to place boundaries to high, good, moderate, poor and bad status. These boundaries must correspond to the definitions in the WFD (Table 9.1).

One criticism of the approach to developing ecological assessment techniques is that TP concentration was focused on as the sole pressure gradient. However, it was felt that this was valid for the Irish situation, where eutrophication is the dominant pressure on lakes. Some pressures such as hydromorphological change may be of lesser importance as this study included few heavily modified water bodies.

Metrics were not developed for acidification pressure in the current study. For biological metrics to be developed, it may be necessary to complement current data with a separate study involving a selection of lakes stratified across a range of known acidification pressures in Ireland, such as the extent of forestry (Allott *et al*., 1997). Some indication of the ecological changes that may take place was given in Chapter 4 where the relative abundance of Desmids declined below pH 6. One of the difficulties of such a study is separating naturally acid lakes from those affected by anthropogenic acidification from anthropogenic acidification as may be caused by forestry. A concurrent palaeolimnological project may be necessary to adequately measure deviation away from reference status.

It is useful to make some remarks about the level of taxonomy required for an effective ecological assessment system. While some of the metrics developed suggest focusing on a very reduced set of taxa, such as ‘eutrophic’ taxa for phytoplankton, it may be premature to adopt such an approach. The development of ecological assessment tools is ongoing in Europe and it is recommended that future work on ecological assessment is carried out at the lowest taxonomic resolution possible so that future assessment tools may be availed of. It is possible to make observations on the level of taxonomy that was considered feasible during the operation of this

project. This was largely a function of the biological group considered, as some groups are more difficult taxonomically than others (Table 10.1).

Table 10.1 Recommended level of taxonomy

Biological group	Level of taxonomy practical
Phytoplankton	Largely Genus with selected taxa to species
Macrophytes	Species where possible
Littoral invertebrates	Species where possible
Profundal invertebrates	Largely Genus with selected taxa to species

11. Conclusions and recommendations

The conclusions and recommendations that follow from the discussion in Chapter 10 are:

- Information on the ecology of 201 lakes was successfully collected. The resulting dataset is the most extensive on lake ecology in the Republic of Ireland.
- The typology described in Chapter 8 is to be used as the reporting typology for Ireland. Comparison of the typologies developed at the individual element level indicated that they were at least as successful as the default typology system of the WFD – system A. The typology should prove effective in partitioning natural variation and allow ecological change to be detected more easily.
- Further work may help the application of the typology. The relative benefits of using the typologies developed at individual element level versus the overall typology level should be further considered.
- Further work may be conducted for typology refinement such as the development of typologies based on the variation of ecological assessment metrics in reference condition for the biological elements.
- Research into the background nutrient status of lakes is needed. Owing to the uncertainty associated with background (reference) nutrient concentration estimates, it may be preferable to apply a correction factor based on background nutrient status after the application of an ecological assessment metric. This would also be likely to improve transparency.
- Other elements that may be useful to refine the typology at a later date are information on residence time, more detailed information on stratification and mixing depth, a description of the substrate for macrophytes and invertebrates and a measure of exposure for macrophyte and littoral invertebrate sites. In addition the biological elements: fish and phytobenthos were not considered in this study but should be incorporated at the earliest opportunity.
- The success of the ecological assessment metrics developed was found to vary with the biological elements. Some elements such as phytoplankton and macrophytes were clearly more useful in detecting pressure.

- The multimetric index developed for the macrophytes appears to have achieved similar success in detecting the effects of pressure on ecological quality in the lakes as the long established Q-value system has for river quality in Ireland and should provide a useful tool in ecological assessment.
- The performance of the invertebrate trophic score for littoral macroinvertebrates had limited success (r^2 = 0.43) and needs further development. The response of littoral macroinvertebrates to eutrophication pressure may be complex, perhaps owing to the non-uniform substrate, diverse communities or complex food web.
- The relationship between profundal invertebrates and eutrophication pressure was also concluded to require further work. The complicating influence of depth and stratification on the effects of eutrophication may make it difficult to develop an assessment system that can be used in all situations. Focusing on a limited depth range such as the sub-littoral as well as the profundal zone may help extract the influence of stratification and depth from that of eutrophication.
- Most ecological change was found to take place at an early stage of nutrient enrichment; generally between 10 and 20 μg l^{-1} TP. Ecological status boundaries need to reflect this.
- Ecological assessment metrics were developed for eutrophication only. It is recommended to complement current data with a separate study whose site selection is stratified across a range of known acidification and hydromorophological pressures in Ireland.
- Further chemical and physical characterisation of lakes is necessary and should be carried out by the River Basin Districts.
- Reference conditions were unavailable or poorly represented for certain lake types – mainly in the 20 – 100 mg l^{-1} $CaCO_3$ alkalinity band. A palaeolimnological project focusing on biological elements such as phytoplankton, chironomids and macrophytes would help to characterise reference conditions for these types. This would complement ongoing work by the EPA funded project INSIGHT, which aims to test the validity of many of the reference lakes selected by this project using palaeolimnological techniques.

- Fundamental research into the functioning of aquatic ecosystems at third level should be supported to provide a basis for continued refinement of ecological assessment techniques.
- Work should be immediately carried out to refine metrics, develop estimates of uncertainty in assessment and an excerise on integrating assessment, based on all biological elements should be performed.

12. References

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Appendices

Appendix 1. The classes of CORINE land cover (standardised for all of Europe) aggregated into the six landuse categories.

Landuse Cateogry	Code	Corine data
Urban	111	Continuous Urban Fabric
	112	Discontinuous Urban Fabric
	121	Industrial or commercial units
	122	Road and Rail Networks
	124	Airports
	131	Mineral Extraction Sites
	133	Construction Sites
	142	Sport and Leisure facilities
Forestry	311	Broad Leafed Forest
	312	Coniferous Forest
	313	Mixed Forest
Pasture	231	Pasture
	2311	Pasture High Productivity
	2312	Pasture Low Productivity
	2313	Pasture Intimate Mix
Other Agricultural land	211	Non-Irrigated Arable Land
	242	Complex cultivation patterns
	243	Land Principally Occcupied by Agriculture
Bogs	4121	Peat Bogs - Unexploited
	4122	Peat Bogs - Exploited.
	412	Peat Bogs
	322	Moors and Heathlands
Other	321	Natural Grasslands
	324	Traditional Woodlands/scrub
	331	Beaches
	332	Bare Rock
	333	Sparsely vegetated areas
	411	Inland Marshes

Appendix 2. Taxonomic keys used for the identification of phytoplankton.

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Appendix 6. A summary of datasets used for the development of typology and relationships between TP and oligochaete community structure and chironomid community structure in the profundal zone.

	Reference Lakes		Oligochaete	Chironomid
No. of lakes	63.00		166.00	93.00
Pseudopecies cut levels	0, 44.44, 222, 444, 888		0, 50, 250, 500, 750, 1000	0, 50, 250, 500, 750, 1000
No. of taxa	31		13	26
Variable Datasets	Dataset 1	Dataset 2		
pH	√	√	√	√
Conductivity	√	√	√	√
Alkalinity	√	√	√	√
Colour	√	√	√	√
Secchi	√		√	√
Chl 'a'	√		√	√
TP	√		√	√
Depth At sites/ max depth	√	√	√	√
CCC				√
CCP				√
ILBS				√
Altitude meters	√	√		√
Lake Area km^2	√	√		√
Lake Perimeter km	√	√		√
Catchment Area km^2	√	√		√
Catchment Perimeter km	√	√		√
Urban	√			√
Forestry	√			√
Pasture	√			√
Other Agriculture	√			√
Peat	√			√
Other	√			√

Appendix 7. Lake and taxon abbreviations with the number of replicates taken at each lake.

Lake Name	abbrev	Replicate No.	Taxon	abbrev
ACROW	**ACROW**	5	Chironimids-total	ChironT
Aleckmore	Aleckmor	5	Chironomids - sp indet.	ChironIN
Alewinghta	Alewingh	5	Chironominae	Chiron
Alua	Alua	5	*Chironomus* spp.	ChironSP
ANNAGHMORERN	**ANNAGHMO**	5	*Cladopelma* spp.	Cladop
ANUREDL	**ANUREDL**	5	*Cladotanytarsus* spp.	Cladot
ARDDERRYGY	**ARDDERRY**	5	*Cryptochironomus* spp.	Crypto
Arrow	Arrow	5	*Demicryptochironomus* spp.	Demicr
Ateduan	Ateduan	5	*Dicrotendipes* spp.	Dicrot
Atorick	Atorick	3	*Endochironomus* spp.	Endoch
Aughrusbeg	Aughrusb	5	*Glyptotendipes* spp.	Glypto
Avaghon	Avaghon	5	*Harnischia* spp.	Harnis
Ballinahinchgy	Ballinah	5	*Microchironomus* spp.	Microc
Ballycullinan	Ballycul	5	*Micropsectra* spp.	Microp
Ballyeighter	Ballyeig	5	*Microtendipes* spp.	Microt
Ballymore	Ballymor	5	*Pagastiella* spp.	Pagast
Ballynakill	Ballykk	5	*Parachironomus* spp.	Parach
BALLYNAKILL(GORUMNA)	**BALLYKG**	5	*Paracladopelma* spp.	Paracl
Ballyquirke	Ballyqui	5	*Paralauterborniella* spp.	Parala
BANE	**BANE**	5	*Paratanytarsus* spp.	Parata
BARFINNAY	**BARFINNA**	3	*Polypedilum* spp.	Polype
BARRADL	**BARRADL**	5	*Phaenopsectra* spp.	Phaeno
Belhavel	Belhavel	5	*Pseudochironomus* spp.	Pseudo
BELTRAMO	**BELTRAMO**	5	*Sergentia* spp.	Sergen
Boderg	Boderg	5	*Stempellina* spp.	StempLI
Bofin	Bofin	5	*Stempellinella* spp.	StempeLA
Bofingy	Bofingy	5	*Stictochironomus* spp.	Sticto
Brackley	Brackley	5	*Tanytarsus*	Tanyta
Bridget	Bridget	5	unidentified	uniden
BUNNY	**BUNNY**	4	Tanypodinae	Tanypo
Calluananyheeda	Calluana	5	*Procladius* spp.	Procla
CARAGHNORTH	**CARAGHN**	3	Pentaneurini	Pentan
CARAGH-SOUTHBASIN	**CARAGHS**	5	Orthocladinae	Orthoc
Carranorth	Carran	4	*Cricotopus/Orthocladius* spp.	Cricot
Carrasouth	Carras	5	*Heterotanytarsus* spp.	HeteroTA
Carrigeencor	Carrigee	5	*Heterotrissocladius* spp.	HeteroTR
Carrowkeribly	Carrowke	5	*Psectrocladius* spp.	Psectr
Carrowmore(n)	Carrowmo	5	*Synorthocladius* spp.	Synort
Cavetown	Cavetown	5	*Tvetenia* spp.	Tveten
Cloneem	Cloneem	4	Diamesinae spp.	Diames
Clonlea	Clonlea	5	*Protanypus* spp.	Protan
Cloonacleigha	Cloonacl	5	Prodiamesinae	ProdiaM
CLOONAGHLIN	**CLOONAGH**	5	*Monodiamesa* spp.	Monodi
CLOONGATGY	**CLOONGAT**	5	*Prodiamesa* spp.	ProdiaMS
Connlower	Connlr	5	Oligochaeta	Oligoc
Connupper	Connupr	5	Lumbricidae	LumbriCI
Corglass	Corglass	5	Tubificidae	Tubifi
CORRIBMIDDLE	**CORRIBMI**	5	*Limnodrilus hoffmeisteri* immature	LHOFFIM
CORRIBUPPER	**CORRIBUP**	5	L. hoffmeisteri mature	LHOFFMA
Corry	Corry	5	Limnodrilus grp	Limnod

CRAGHYDL	**CRAGHYDL**	5	*Aulodrilus pluriseta*	APLUR
CULLAUN	**CULLAUN**	5	*Potamothrix hammoniensis* mature	PHAMMMA
CURRANE	**CURRANE**	3	P. hammoniensis immature	PHAMMIM
Cutra	Cutra	3	*Tubifex tubifex*	tubife
DAN	**DAN**	5	*P. bavaricus* spp.	PHAMMBA
Derg	Derg	3	Pot-tub group	PottuB
Dergdl	Dergdl	5	*Psammoryctides barbatus*	Psammo
Derravarragh	Derravar	5	*Spirosperma ferox*	SFERO
Derrybrick	Derrybri	5	Lumbriculidae spp.	LumbriCU
Derrycassan	Derrycas	4	*Lumbriculus variegatus*	LVARI
Derryclare	Derrycla	5	*Stylodrilus herringus*	SHERI
Derryhick	Derryhic	5	Naididae	Naidid
Doo	Doo	5	*Dero digata*	Derodi
DOODL	**DOODL**	5	*Vejdovskyella comata* spp.	Vejdov
DOOMO	**DOOMO**	5	*Nais* spp.	Naissp
Dromore	Dromore	5	*Slavina appendiulata*	Slavin
Drumharlow	Drumharl	5	Enchytridae	Enchyt
Drumlaheen	Drumlahe	5	others	others
Drumlona	Drumlona	5	Ceratopogonidae	Ceratopo
Duin	Duin	5	Charobus	Chaobor
DUNGLOWDL	**DUNGLOWD**	5	Diptera pupa	Diptera
Dunlewy	Dunlewy	5	Caddis cased - total	CadcdT
EASKYSO	**EASKYSO**	5	Caddis (cased) indet sp.	CadcdIN
Egish	Egish	5	Limnephilidae	Limnephi
Ennell	Ennell	5	Leptoceridae	Leptocer
Errit	Errit	5	Athripsodes cinerus	Athripso
Eske	Eske	5	*Ecnomus tenellus*	Ecnomus
FAD(EAST)DL	**FAD(EAST**	5	*Mystacides longicornis*	Mlongi
FAD(WEST)DL	**FAD(WEST**	5	*Oecetis ochracea*	Oecetis
FEEAGHMO	**FEEAGHMO**	5	*Mystacides* spp.	Mystacid
FEEGY	**FEEGY**	5	Caddis casless - total	CadclT
FINMO	**FINMO**	5	Caddis casless indet sp.	CadclIN
Finn	Finn	3	Polycentropodidae - unided	PolycIN
Finndl	Finndl	5	*Cyrnus trimaculatus*	Ctrima
Forbes	Forbes	5	C. falvidus	cyrnusf
Garadice	Garadice	5	*Holcentropus picicornis*	Holcentr
Garanorth	Garanort	5	*Plectrocnemia conspersa*	Plectroc
GARTANDL	**GARTANDL**	5	*Polycentropus flavomaculatus*	Polyflav
Garty	Garty	5	TRICHOPTERA INDET	TRICHIN
Gill	Gill	5	Hirudinae	Hirudina
Glasshouse	Glasshou	5	**Caddis casless indet**	**Caddisc**
Glen	Glen	5	H. stagnalis	HSTAGN
Glenade	Glenade	5	*Glossoma complanata*	GCOMPL
Glenbeg	Glenbeg	5	*Eropbdella octulata*	Eoctul
GLENCARLM	**GLENCARL**	5	*Hemclepsis marginata*	Hemcleps
GLENCULLINMO	**GLENCULL**	5	*Dina lineata*	Dinalin
Glendollagh	Glendoll	5	*Haementeria costata*	Haemente
Glenicmurrengy	Glenicmu	5	Crustaceans - total	Crustace
Glenmore	Glenmore	5	Fairy shrimps	Fairysh
Glore	Glore	1	*Mysis relicta*	Mysisre
GOLAGH	**GOLAGH**	5	*Asellus aquaticus*	Aaquat
Gowna-north	Gownan	5	*A. meridianus*	Amerid
Gowna-south	Gownas	5	*Asellus* spp.	Asellus
Graney	Graney	2	*Gammarus* spp.	Gammarus
GUITANE	**GUITANE**	5	*Crangonyx* spp.	Crangony
Guladoo	Guladoo	5	ZOOPLANKTON group total	ZOOPLANK

Inagh	Inagh	5	Cladocera	Cladocer
Inchiquincl	Inchiqcl	3	Leptodoridae	Leptodor
INCHIQUINKY	**INCHIQKY**	3	Copepods	Copepods
Inniscarraeast	Innisce	5	Ostracods	Ostracod
Inniscarrawest	Inniscw	5	*Argulus* spp.	Argulus
Invernagleragh	Invernag	5	Ephemeroptera - total	EphemT
Islandeady	Islandea	5	Ephemeroptera - sp .indet	EphemIN
Keel	Keel	5	BAETIDAE	BAETIDAE
KEEL(ROSSES)DL	**KEELDL**	5	*Caenis* spp.	Caenis
Key	Key	5	*Caenis lucuosa*	Caenisl
Kilglass	Kilglass	5	*Caenis horaria*	Caenish
KILTOORISDL	**KILTOORI**	5	*Chloroperla torrentium*	Chlorope
Kinale	Kinale	5	*Ephemera danica*	Edanic
KINDRUMDL	**KINDRUMD**	5	Nematodes	NEMATODS
Knappaghbeg	Knappagh	5	Hydracarina	HYDRACAR
KYLEMOREGY	**KYLEMORE**	5	Bivalves-total	BIVALVES
lannagh	lannagh	5	*Dressina polymorpha*	Dpolym
LENE	**LENE**	5	Sphaeridae	Sphaerid
lettercraffoe	lettercr	5	Unionida	Unionida
levally	levally	5	Gastropoda - total	GastropT
lickeen	lickeen	5	Gastropoda indet.	GastIN
loughanillaun(maum)	loughm	5	*Valvata cristata* spp.	Valvata
loughanillaungy	loughgy	5	*V. piscinalis* spp.	Vpisci
MACNEAN	**MACNEAN**	5	*V.macrostoma* spp.	Vmacros
MASKMO	**MASKMO**	5	*Planorbis* spp.	Planorbi
MAUMEENGY	**MAUMEEN**	5	*Bythina tentaculata* spp.	Btenta
MAUMWEEGY	**MAUMWEE**	5	*Physa fontinalis* spp.	Physafo
MCNEAN	**MCNEAN**	5	*Lymneae pergra* spp.	Lymneae
Meela	Meela	5	*Potamopygrus jenkinsi* spp.	Pjenki
Meelagh	Meelagh	5	Planarians total	PLANT
MELVIN	**MELVIN**	5	Planarians indet spp.	PlanIN
MOURNE	**MOURNE**	5	*Dugesia lugubris/Polychroa*	Dugesia
MUCKANAGH	**MUCKANAG**	5	*Polycelis nigra/tenius*	Polyceli
Mucknon.	Mucknon	5	coleoptera total	coleopte
Mucknosouth	Mucknos	5	Chrysomelidae	Chrysome
MUCKROSS	**MUCKROSS**	5	*elmis* spp.	elmis
Nablahy	Nablahy	5	*Oulimnius* spp.	Oulimniu
Nacungupr	Nacungup	5	*Esolus* spp.	Esolus
Nadgreegeel	Nadgreeg	5	*Limnius* spp.	Limnius
NAFOOEYGY	**NAFOOEYG**	5	Gyrinidae	Gyrinida
NAKIRKA	**NAKIRKA**	5	Lepidoptera	Lepidopt
NALUGHRAMANDL	**NALUGHRA**	5	*Paraponyx* spp.	PARAPONY
NAMBRACKKEAGHGY	**NAMBRACK**	5	Odonata total spp.	ODONATA
NAMINNA	**NAMINNA**	5	Zygoptera spp.	ZYGOPTER
NAMINNDL	**NAMINNDL**	5	*Enallagma cyathigerum* spp.	Enallagm
Oakport	Oakport	5	Coenagrioniidae spp.	COENAGRI
O'FLYNNRN	**O'FLYNN**	5	Corixidae spp.	CORIXIDA
OORIDGY	**OORIDGY**	5		
OWEL	**OWEL**	5		
Pollaphuca(bigbasin)	Pollapbb	5		
Poulalphoucanorthbasin	Poulalpn	3		
Poulalphoucawest	Poulalpw	5		
Ramor	Ramor	3		
REA	**REA**	3		
Ree1	Ree1	9		

Rinn	Rinn	5
Rockfield	Rockfiel	5
Ross	Ross	5
Rowan	Rowan	3
Sallagh	Sallagh	5
SALTDL	**SALTDL**	5
Scur	Scur	5
Sheelin	Sheelin	5
SHINDILLAGY	**SHINDILL**	5
Sillan	Sillan	5
Skannive	Skannive	5
Skeaghuppr	Skeaghup	5
Skean	Skean	5
St.jonhn's	St.john	5
Sunderlin	Sunderli	5
TALTSO	**TALTSO**	5
Tap	Tap	5
TAY	**TAY**	5
Templehouse	Templeho	5
UPPERLAKE	**UPPERLAK**	4
Urlaur(s)	Urlaur	5
WASKELDL	**WASKEL**	5
White	White	5