Assessment of the Trophic Status of Rivers using Macrophytes: Evaluation of the Mean Trophic Rank

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E39

Assessment of the Trophic Status of Rivers Using Macrophytes.

Evaluation of the Mean Trophic Rank

R&D Technical Report E39

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Statement of use
This report presents the main findings from R&D project E1-i694 'Assessment of the Trophic Status of Rivers using Macrophytes'. It describes the evaluation of the macrophyte-based Mean Trophic Rank system and compares this with other methods. The principal application for which the system was evaluated is the designation of Sensitive Areas (Eutrophic) under the Urban Waste Water Treatment Directive. The report will be of interest to those involved with the development, management or implementation of biological methods to assess trophic status.

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CONTENTS

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LIST OF FIGURES

LIST OF TABLES

Keywords: Macrophyte surveys, Urban Waste Water Treatment Directive, UWWTD, Sensitive Areas, eutrophication, trophic status, phosphorus, nutrient, monitoring.

GLOSSARY AND ABBREVIATIONS

CCW	Countryside Council for Wales.
Comparability	a measure of confidence in the physical comparability of a pair of sites based upon the similarity of width, depth, substrata, habitat, shading, water clarity and bed stability.
CVS	Cover Value Score. The score allocated to a species resulting from the multiplication of the Species Trophic Rank and the Species Cover Value.
DoE	The Department of the Environment.
d/s	downstream.
EA	Environment Agency
EN	English Nature.
DQI	The Diatom Quality Index. A transformation of the Trophic Diatom Index (TDI) for use when comparing TDI with MTR results.
highlighted	refers to a plant species within MTR which is believed to be a particularly reliable indicator of trophic status.
IFE	The Institute of Freshwater Ecology.
IRTU	Industrial Research & Technology Unit (Northern Ireland).
LEAP	Local Environment Agency Plan.
macrophyte	higher aquatic plant or alga (includes bryophytes), observable to the naked eye and nearly always identifiable when observed.
MTR	Mean Trophic Rank. A numerical 'score' assigned to a survey length based on its macrophyte presence and abundance characteristics.
nitrate	dissolved, soluble or non-particulate nitrate (concentrations given as mg l^{-1} nitrate-nitrogen).
non-QD	Non-Qualifying Discharge under the Urban Waste Water Treatment Directive (UWWTD), ie less than 10,000 population equivalents (pe).
NRA	The National Rivers Authority, a predecessor to the Environment Agency for England and Wales.
pe	population equivalent.
phosphate	dissolved or non-particulate phosphate, normally analysed as soluble reactive phosphate (SRP) or by the molybdenum-blue method (concentrations given as

mg l^{-1} phosphate-phosphorus).

QD — Qualifying discharge, usually from a waste water treatment works (WWTW), under the Urban Waste Water Treatment Directive (UWWTD), ie greater than 10,000 population equivalents.

RHS — River Habitat Survey: a method for assessing the physical character and quality of rivers, and impacts upon them.

SA[E] — Sensitive Area (Eutrophic). An area of water which is considered to be eutrophic or which in the near future may become eutrophic if protective action is not taken, and recognised as such by designation under the Urban Waste Water Treatment Directive.

SCV — Species Cover Value. A value assigned to a species according to the percentage of the survey area which it covers.

Site — This is the broad location where the survey is to take place, eg *x* km downstream of a waste water treatment works.

SNIFFER — Scottish and Northern Ireland Forum for Environmental Research.

SNH — Scottish Natural Heritage.

STR — Species Trophic Rank. A value assigned to a species on a scale of 1 to 10, designed to reflect the tolerance of that species to eutrophication. Low ranking indicates tolerance or 'cosmopolitan' distribution (ie no preference). High ranking indicates a preference for less enriched conditions or intolerance of eutrophic conditions.

Survey — The collection of data at one site according to the prescribed methodology.

Survey season — The MTR season is from mid-June to mid-September inclusive.

Survey length — This is the sample area - the actual area of river channel which is surveyed, between two fixed points on the bank. The survey length is 100m in length for standard MTR surveys.

TDI — The Trophic Diatom Index. A method for assessing the trophic status of rivers using benthic diatoms.

u/s — upstream.

UWWTD — The European Community Urban Waste Water Treatment Directive.

WWTW — Waste Water Treatment Works.

Probabilities (P) for the significance of statistical results: 'significant' at the 95% level of confidence or P = 0.05 and indicated '*'; 'highly' significant, 99% or P = 0.01 by '**'; or 'very highly' significant, 99.9% or P = 0.001 by '***'.

EXECUTIVE SUMMARY

1. The Mean Trophic Rank (MTR) is a biotic index developed by N.T.H. Holmes for the National Rivers Authority, a predecessor of the Environment Agency, specifically for the purposes required of biological monitoring under the EC Urban Waste Water Treatment Directive (UWWTD). It is based on the presence and abundance of aquatic macrophytes and uses a simple scoring system to derive a single index to describe the trophic status of a site. Species present are assigned a score (Species Trophic Rank - STR) according to their tolerance to eutrophication (the higher the score, the less the tolerance), and a mean score (MTR) for the site is then calculated, weighted according to the relative abundance of the individual species. MTR scores can range from 10 to 100 (there is no score if scoring species are absent) and decrease with increasing eutrophy.

2. The overall objectives of this study were: to evaluate the MTR as a robust transportable system for assessing the trophic status of rivers using aquatic macrophytes; to compare this with other biological methods of assessing the trophic status of rivers (principally the Diatom Quality Index - DQI); and to produce a recommended method to assist in the designation of Sensitive Areas (Eutrophic) under the UWWTD.

3. A workshop on the use of diatoms and macrophytes to assess trophic status was held to obtain feedback from end-users of the MTR and DQI, recommendations from which were incorporated into the project plan. Both a full and a summary report were produced and distributed (included in R&D Project Record E1/i694/01).

4. Data from MTR surveys in England and Wales, undertaken by the Agency for UWWTD purposes during the period 1994-96, were gathered together with comparable chemical data where available. Initial analysis of the data for 1994-95 revealed the need to supplement the Agency dataset, in order to cover a wider range of river types and to check on some of the 'unexpected' results. Supplementary data were obtained from two sources: MTR surveys undertaken specifically for the project during 1996, and data contributed by both the statutory conservation agencies in England, Wales and Scotland and the regulatory authorities in Northern Ireland. The contributed data also served to extend the dataset to cover the whole of the UK. A small number of diatom surveys were undertaken in parallel to MTR surveys, to extend the comparison between MTR and DQI.

5. A small proportion of MTR surveys undertaken for UWWTD purposes or specifically for this project were re-surveyed for quality assurance purposes. The results of these re-surveys were used to highlight areas of inconsistency. Options for reducing these inconsistencies and thus assuring the quality of results are reviewed and recommendations made. Recommended quality assurance measures are described in full in the companion Technical Report: *Mean Trophic Rank - A User's Manual* (R&D Technical Report E38). They include: (i) measures which are integral to the survey methodology, such as on-site checks and multiple-staffing; (ii) training of surveyors; and, (iii) resurveys. Two resurvey (audit) protocols were derived, each offering a different level of specification, and guidance produced on appropriate action to take if the results of the primary and audit surveys are significantly different.

6. All data gathered for the project were collated and organised, together with other supplementary information, into an appropriate relational database (MS-Access) with a general access front-end. Data entries were validated. Although not a specific objective

of the project, this not only allowed analysis for the MTR evaluation within this project, but also produced a version of the database for operational use on a single-user basis. An instructional manual for the operational version of the database is included within the companion Project Record of this project (R&D Project Record E1/i694/01).

7. Analysis of the dataset showed that the MTR system performed sufficiently well to be used as a tool in the assessment of the trophic status of rivers, to provide evidence in support of the designation of Sensitive Areas (Eutrophic) under the UWWTD. The values assigned to plant species (Species Trophic Ranks) were in general related to their tolerance of eutrophication. MTR was found to be negatively correlated with the logarithm of phosphorus concentration and the nitrate concentration, particularly at concentrations less than 1 $mg\,l^{-1}$ P or 10 $mg\,l^{-1}$ N, but even more so at less than 0.5 $mg\,l^{-1}$ P or 5 $mg\,l^{-1}$ N. It may also be influenced, however, by the physical characteristics of the river and by within-season variation. The MTR system may be used to describe downstream changes in trophic status, provided the sites being compared are physically similar and an error margin of 4 MTR units or 15% is allowed for seasonal variation and sampler error. The MTR system is easy to use, relatively cost effective compared to other assessment systems currently in use and is readily integrated with other sampling programmes. The methodology is reproducible and repeatable within limits (about 4 MTR units) and is nationally applicable. The absence of species scoring an STR of 2 in northern Scotland may merit further investigation to improve national applicability.

8. Applications other than monitoring for UWWTD purposes were considered. Although MTR may be used as a tool to assess the impact of discharges not qualifying under the UWWTD, or in longitudinal surveys of individual rivers, the use of MTR values for comparisons between rivers or catchments should, in particular, be treated with caution.

9. Comparative evaluation of the MTR and DQI showed that both methods are of value in the assessment of the trophic status of rivers. The methods are complementary and both should be used wherever possible. DQI (a transformation of the Trophic Diatom Index or TDI) should be used in preference to the TDI when making comparisons with MTR data.

10. Recommendations resulting from the MTR evaluation that relate to the operation of the method are incorporated into the companion Technical Report to this volume, *Mean Trophic Rank: A User's Manual* (R&D Technical Report E38). Only one substantive change to the method is recommended: *Stigeoclonium tenue* should be removed from the list of scoring taxa as it is considered that the distribution of this species is influenced more by non-nutrient pollution than by nutrient enrichment.

11. Further research is required to (i) verify the STRs assigned to certain species; (ii) examine whether key species may be useful in distinguishing between phosphate and nitrate enrichment; and, (iii) establish the precise influence on MTR of the physico-chemical characteristics of rivers. Development of a predictive element to the system, which allows for natural differences between rivers, is recommended. As well as improving the performance of MTR, such a system should also allow monitoring and classification of trophic status and eutrophication impact for applications other than UWWTD, including the proposed EC Water Framework Directive, national eutrophication management strategies and individual catchment/ river-basin management plans.

1. INTRODUCTION

1.1 Purpose of this report

The purpose of this report is to present the main findings and conclusions of National R&D Project E1-i694: 'Assessment of the trophic status of rivers using macrophytes'. Options for the biological assessment of trophic status are discussed and one method (the macrophyte-based Mean Trophic Rank) is evaluated in detail. Particular reference is made to the requirements of the EC Urban Waste Water Treatment Directive (UWWTD, 91/271/EC), and recommendations are given about the methodology (or methodologies) to be used.

The main audience are those involved in the development, management and/or implementation of biological methods to assess trophic status. The report, however, will also be of interest to conservation staff concerned with river eutrophication.

Supporting information, in the form of reports from the MTR-user workshops and data gathered during the project (except for data subject to access restrictions), can be found in the corresponding Project Record (E1/i694/01). Detailed procedural guidance on assessment methodology, recommended on the basis of the project findings, can be found in R&D Technical Report E38 (Holmes et al 1999).

1.2 Overall objective of the project

The main objective of the project was to evaluate the Mean Trophic Rank (MTR) system developed by Holmes (1995), in order to produce a robust, transportable system for assessing the trophic status of rivers using macrophytes. A second objective was to compare this and other biological methods of assessing the trophic status of rivers and evaluate the role of each in such assessment. The third objective was to update and integrate this evidence and produce a recommended method. The principal application focused upon within the project, was the presentation of data as evidence for designation of Sensitive Areas [Eutrophic] (SA[E]s) under the UWWTD. The potential use for other applications, however, was also considered.

1.3 The organisation of this report

The remainder of this introduction outlines the background and justification for the project, highlighting the need for a robust, transportable biological system of assessing trophic status in rivers. The options to fulfil this need are then introduced and considered. The definition of eutrophication used for the purposes of the project is given. The following chapters (2 - 5) then detail the evaluation of the MTR system, describing the overall approach (methods and preparation), the results, and conclusions on the performance of MTR. Variability and error, and the practical aspects of the methodology are considered. The next chapter (6) compares the MTR system with other methods, and the final chapter (7) summarises the conclusions of the project and makes recommendations on applications of the methodology and future work.

1.4 The need

For the purposes of the designation of Sensitive Areas (Eutrophic), SA[E]s, under the UWWTD, the UK is required to determine whether a reach of river receiving a qualifying waste water treatment works (WWTW) discharge is eutrophic or is 'at risk of shortly becoming eutrophic'. A 'qualifying discharge' (QD), for the purposes of the UWWTD, is one with a loading of more than 10,000 population equivalent (p.e.). Evidence submitted in support of such a designation should include biological data, which should specifically relate to the trophic status of the river rather than, for example, the level of organic pollution.

Until recently, however, there has been no nationally validated and routinely used biological system within the UK for assessing the trophic status of rivers. There was thus an urgent need for a such a system(s) to enable the Environment Agency (hereafter referred to as the Agency) to reach scientifically valid decisions as to which rivers to put forward as SA[E]s and to provide robust evidence in support of each case.

Once developed, systems for assessing the trophic status of rivers may be used in the future for applications beyond purely UWWTD monitoring purposes. Potential applications include:

- identification of nutrient inputs overlooked by the more traditional invertebrate-based methods, whether they be point- or diffuse-sources;
- assessment of the cumulative impact of several small, non-qualifying WWTW discharges (or other similar discharges);
- catchment 'audits' of trophic status to aid the production of management plans such as Local Environment Agency Plans (LEAPs) and river basin management plans for the proposed EC Water Framework Directive;
- setting targets for, and demonstrating changes in, trophic status under the national eutrophication management strategies currently being developed by regulatory bodies in the UK (Environment Agency 1998, DoE, NI and SEPA).

Extension to such applications may be of great value in delivering eutrophication control on a catchment basis.

1.5 Biological Methods for assessing trophic status of rivers

1.5.1 The options

To address the need for a nationally validated system(s) to assess the trophic status of rivers, two methods have been developed in parallel by the Agency (and formerly by the National Rivers Authority; NRA). These are the Trophic Diatom Index (TDI), which is based on epilithic diatoms, and the macrophyte-based MTR; both of which are described below (1.5.2 & 1.5.3).

A literature survey was undertaken at the start of this project, to establish whether there were any other potential new methodologies from Britain and Europe, which could be used to assess the trophic status of rivers following on from the pioneer work of Arber, Butcher, Braun-Blanquet, Pearsall and Sculthorpe (Sculthorpe 1967). Algal survey methods were recently reviewed by Round (1993) and for the UK by Kelly and Whitton (1993). Recent developments in algal survey methodologies have also been discussed by practitioners at

regular European meetings (Whitton & Rott 1996, Prygiel et al 1998), from which the standardised TDI method used here, including the assessment of pollution tolerant taxa, was developed. A survey of the literature on macrophytes revealed several methods, including community based indices, but these were either applicable only at a local or an area level (Spence, 1967, Seddon 1972, Kohler et al 1973, Wiegleb 1978, Harding 1981, Meriaux 1982, Caffrey 1987), or if national did not meet other requirements (Newbold & Palmer 1979, Haslam & Wolseley 1981, Holmes 1983, Kelly 1989, Haury et al 1995). Such methods either did not give a sufficiently broad national approach, or did not cover the wide range of habitats (including flow types, etc) found in Britain (Palmer 1989), or did not utilise plant species representative of the UK flora, or were not rigorously enough proven. [Other literature reviewed is included in the bibliography.]

As a result of the review, it was concluded that no other methods had greater potential than the diatom-based TDI and the macrophyte-based MTR. Therefore, this project undertook an analysis of the MTR methodology together with an assessment of the Diatom Quality Index, DQI, a transformation of the TDI recommended for comparison with MTR (see 1.5.2). Towards the end of the project, recently published accounts of the effects of environmental parameters on aquatic plant associations and of morphological/functional groups in relation to nutrient levels were considered (Dawson & Szoszkiewicz, 1998, Murphy & Ali 1998, Ali et al 1999). A method based on functional groups is compared with MTR by Ali et al (1999) and is considered briefly in section 6.4.

1.5.2 Trophic Diatom Index (TDI)

TDI was developed by Kelly and Whitton (1995b) for the National Rivers Authority (NRA) as part of an investigation into the use of plants to monitor rivers and was further refined by Kelly (1996a, b & c). The initial development work was undertaken at a time when the requirements of the UWWTD were becoming apparent. The prime focus of the research was on the use of diatoms to monitor change in trophic status, following the strong record of their use in continental Europe for monitoring acidification and water quality (eg Prygiel et al 1998).

The TDI is derived from the weighted-average equation of Zelinka and Marvan (1961), using taxon sensitivities to nutrient status, indicator value (spread around the mean) and abundance. The TDI was developed using a smaller dataset (70 sites) than used for the development and evaluation of MTR, but at sites free from significant organic pollution (MTR sites included organically polluted sites). Scores were assigned to diatom taxa, often to the genus level, according to their sensitivity to nutrient status. Scores range from 1 for taxa which are favoured by very low nutrient concentrations to scores of 5 for those favoured by very high concentrations. The resulting TDI score, when calculated, indicates the level of nutrients or trophic status - the higher the score, the higher the nutrient level. In complex situations such as WWTW outfalls, the interpretation of results can be complicated by taxa which are responding to components of the discharge other than nutrients, eg elevated suspended solids, ammonia, dissolved oxygen. Thus, a further value is calculated using species tolerant to organic pollution to indicate the influence of organic pollution (Chapter 6).

Following discussions and trials including four NRA areas in Anglian, Thames and Midlands regions, modifications were made to the methodology. This included reversing the direction of the TDI scale to create DQI, to facilitate the presentation of results and comparison with MTR data. DQI operates on a scale of 0-100 and MTR on a similar scale of 10-100, with

both indicating decreasing nutrients with increasing score (see 2.3.6). In addition, a flow-chart was produced giving guidance on sampling methodology, such as the experimental use of artificial substrata if no suitable natural surfaces are present, and sampling from the 'recovery zone'. A grid to facilitate interpretation of results has also been produced. Full details of the TDI and DQI are given in Kelly (1996a & b).

1.5.3 Mean Trophic Rank (MTR)

The Mean Trophic Rank (MTR) system was developed by Holmes (1995) for the NRA, specifically as a macrophyte-based method of assessing the trophic status of rivers for the purposes of the UWWTD. The system is based on a survey of the cover of each macrophyte species in a 100m section of river (modified 'Blue Book' aquatic macrophyte survey method B, DoE Standing Committee of Analysts 1987). Each of 129 species of aquatic plant was allocated a Species Trophic Rank (STR) score according to its response to eutrophication, based upon data from the literature and expert opinion (see Appendix 6 for STR checklist). STRs range from 1 to 10 – a numerically high value indicates that the plant is intolerant of eutrophication, whereas a low STR indicates that the plant is either tolerant of eutrophication or alternatively has no preference and is termed 'cosmopolitan'. The MTR score for a section of river is calculated (Appendix 7) by:

1. assessing the percentage cover of each 'scoring' plant species on a scale of one to nine, to give the 'species cover value' or SCV (the greater the cover the higher the value);
2. multiplying the STR for each 'scoring' plant species by its SCV, to give the 'cover value score' or CVS for the species;
3. dividing the sum of SCV by the sum of CVS, and multiplying by ten to give the MTR score.

The resulting MTR score lies in the range of 10-100 - the lower the score the more eutrophic the site (no score is recorded if scoring species are absent). In undisturbed ecosystems with low levels of nutrient input, a theoretical maximum score of 100 should be achieved. In degraded, higher nutrient and/or disturbed ecosystems a lower score would be expected. The change from the maximum or 'perfect score' can be used as a measurement of the impact or damage caused to the ecosystem by the disturbance. A series of three measures of confidence are assigned to the MTR score for each site, based upon the survey conditions, the physical comparability between sites, and the number of 'highlighted' species (see 4.6). The latter species are considered to be more reliable indicators of trophic status.

NB Where only none-scoring species are present in the survey length, a value for MTR of 'zero' may be recorded for data archiving purposes but this value must ***not*** *be used to indicate trophic status.*

1.6 Specific objectives of the project

These were defined as follows:

1. To collate survey data and information from macrophyte surveys undertaken for UWWTD monitoring purposes throughout the Environment Agency. [Extended to Scotland and Northern Ireland for the purposes of SNIFFER (Scottish and Northern Ireland Forum for Environmental Research.)]

2. To develop a national database for storage and manipulation of such data and information, together with other appropriate data, in order to allow comprehensive evaluation of trophic status.

3. To evaluate how the MTR performs against the following criteria:
   i) adequate assessment of trophic status of rivers;
   ii) robustness;
   iii) reproducibility and repeatability;
   iv) amenability for quality control;
   v) ease of use;
   vi) national applicability;
   vii) cost-effectiveness;
   viii) ease of understanding/interpretation by non-biologists.

4. To outline deficiencies of the MTR system and recommend refinements where appropriate.

5. To define the limitations to the use of the MTR (or refined MTR) system and hence to determine those circumstances under which the MTR can be applied with confidence, and those under which it cannot.

6. To recommend a suitable system of quality assurance for MTR macrophyte surveys.

7. To compare the MTR with other methods, principally the diatom-based Diatom Quality Index (DQI), both on the basis of the criteria listed in (3) above and by addressing the following additional questions.

   i) Do the different methods provide similar answers? (see also point *(v)*)
   ii) Is one method better under some circumstances?
   iii) Are there situations where neither method is suitable?
   iv) Is one method applicable to more seasons than others?
   v) Are the methods alternative or complementary?
   vi) Under what circumstances should each method be used?

8. To produce a report detailing the work undertaken, results and recommendations, plus a revised manual of biological methods for investigating the trophic status of rivers.
   [Note: During the project it became clear that the general manual of biological methods on trophic status was unnecessary, as a TDI/DQI manual was in production at the same time. The manual in this project was, therefore, confined to the MTR, with cross-references to the TDI manual as appropriate.]

1.7 Definition of eutrophication

There are at least three definitions of eutrophication in current usage within the UK.

The definition of eutrophication according to the UWWTD is:

'Enrichment of water by nutrients, especially compounds of nitrogen and/or phosphorus, causing an accelerated growth of algae and higher forms of plant life to produce an undesirable disturbance to the balance of organisms present in the water and to the quality of the water concerned.'

The definition laid down in the Nitrate Directive is:

'Enrichment of water by nitrogen compounds causing an accelerated growth of algae and higher forms of plant life to produce an undesirable disturbance to the balance of organisms present in the water and to the quality of the water concerned.'

The definition adopted by the Environment Agency of England and Wales in the proposed Eutrophication Strategy (Environment Agency 1998), is:

'The enrichment of waters by inorganic plant nutrients, which results in the stimulation of an array of symptomatic changes. These include the increased production of algae and/or other aquatic plants affecting the quality of the water and disturbing the balance of organisms within it. Such changes may be undesirable and interfere with water uses.'

In practice, interpretation of the definition of eutrophication given in the UWWTD may be problematic. These problems were discussed at a workshop held as part of this project (2.3.6; Newman et al 1997a & 1997b, Dawson et al 1999). Examples of the problems are:
What is the objective and target of measurement? Are they the same? Is it the water-column or the whole river system, with or without the influence of sediment chemistry? Such problems are starting to be addressed. The proposed Environment Agency Eutrophication Strategy, if agreed, will be a crucial first step in this process and will provide a standardised working definition of eutrophication for use within the Environment Agency. The definition needs to be expressed in terms which are measurable and achievable so that improvements can be measured against the criteria set out in the definition. A clear statement of what is included in the definition (water column, sediment, ecosystem) should be made.

For the purposes of this project, both the UWWTD definition and that in the Agency's proposed Eutrophication Strategy were used. Both definitions include a chemical (nutrient) and biotic (plant) component, and describe eutrophication as an increase in nutrient level, which causes (or can cause) increased growth of plants and/or a disturbance to the balance of organisms present. The UWWTD definition is the stricter of the two, requiring increased plant growth **and** a disturbance to the balance of organisms present, and the disturbance is **required** to be undesirable. The definition in the proposed Eutrophication Strategy is not as prescriptive and gives increased plant growth and a disturbance to the balance of organisms as example symptoms of eutrophication, which **may** be undesirable. The nutrients considered in this project were phosphorus (as phosphate) and nitrogen (as nitrate). The plants considered were macrophytes (algae and higher plants observable to the naked eye – see Holmes and Whitton 1977), with some reference to microscopic algae in terms of the diatom-based DQI system.

2 METHODS AND PREPARATION

2.1 Introduction

2.1.1 Hypothesis to be tested

The hypothesis to be tested was that the MTR is a useful tool for assessing the trophic status of rivers in terms of the macrophyte community.

2.1.2 General approach to MTR evaluation

The testing of this hypothesis incorporated five main stages:
(1) establishing a set of criteria for the ideal system (see section 2.2);
(2) gathering data to allow evaluation of the performance of the MTR against these criteria (see section 2.3 & 2.4);
(3) assessing the relationship between MTR and nutrient status (see Chapter 3);
(4) assessing the robustness of the MTR (ie sources of variability and error – see Chapter 4); and,
(5) considering the suitability of the MTR as a practical monitoring tool (ie is it easy and cost-effective to use – see Chapter 5).

Throughout the evaluation, deficiencies and limitations of the MTR method were considered and possible refinements suggested, with a view to making recommendations for operational use.

2.2 Performance criteria

2.2.1 Adequate assessment of trophic status of rivers

Performance criterion: the MTR should give an adequate assessment of the trophic status of rivers.

This is the main criterion against which the performance of the MTR was evaluated. To adequately assess trophic status – or more accurately, eutrophication - according to the definitions given in the UWWTD and the Agency's Eutrophication Strategy (1.7), the MTR should detect increased growth of macrophytes and/or a (undesirable) disturbance to the balance of organisms present, caused by the enrichment of the river by nutrients. A 'disturbance to the balance of organisms' is taken, in this context, to mean a change in the macrophyte species composition and/or diversity. Although this incorporates the effects of both nitrate and phosphate, it was assumed for the purposes of MTR development and evaluation that in rivers phosphate concentration is the most limiting to growth, ie the 'controlling factor' in growth. Removal of phosphate is thus likely to be more effective in reducing growth and restoring the 'desirable' balance of organisms present, than removal of nitrate. For this reason, the evaluation focused primarily on phosphate, but with some assessment of the influence of nitrate.

The criterion, therefore, incorporates the following key questions:

- does MTR express the response of the macrophyte community to nutrient enrichment, in terms of increased growth and changes in species composition/diversity?
- does MTR assess nutrient rather than non-nutrient influences?
- can the MTR or individual taxa be used to distinguish between N and P enrichment?
- can the MTR detect downstream changes in trophic status?
- can the MTR detect temporal changes in trophic status?
- can the MTR detect between-river differences in trophic status?

In a 'perfect' system, the answer to all the above questions will be 'yes'. The relationships between biota and physical and chemical factors, however, are often complex and are unlikely to allow 'perfection' in a system of assessment. Should the MTR system prove not to be 'perfect', then the subsequent stage would be to define where it is deficient and to deconstruct the operation of the MTR calculation to evaluate which component(s) were limiting performance. The latter would be aided by constructing a picture of how an 'ideal' MTR system would work, against which the actual evaluation results could be compared. Such an 'ideal' system may incorporate the following elements:

- an equal number of species assigned to each STR, there being an equal probability that at any one site, any one plant recorded will 'score';
- each plant species to respond to trophic status in a statistically 'normal' manner, ie to have a restricted range within which it can grow, with a 'preference' range towards the centre of this 'tolerance' range;
- species ranges to serially abut, but not overlap, with plants with a similar response to trophic status;
- the mean trophic status per species to be correlated with STR;
- within each STR, species to be selected to represent the range of potential environmental conditions (such as pH, alkalinity, geology or flow) found in the UK.

In such an 'ideal' system, the MTR would be derived from species assigned to only the upper, mid- and lower adjacent STR values corresponding to the trophic status of the site.

The analyses required to evaluate the performance of MTR against this criterion, and where necessary to deconstruct the system to understand its deficiencies, included the following.

1. Determination of the underlying relationship between each MTR-scoring species and nutrient status. This included the range and pattern of the relationship; and its comparability with the relationships found for other species.
2. Determination of the relationship between nutrient status and the species rankings (STRs) to establish the variation in nutrient status at any one STR or any anomalous rankings and thus to confirm or refute the STRs.
3. Determination of the relationship between the MTR and nutrient status to establish the variation and/or regular bias resulting from practical, methodological and 'natural' causes (see 2.2.2).
4. Comparison between the STR- or MTR-phosphorus relationship and the corresponding relationships with nitrate and the N:P ratio.
5. Comparison of results between geographical regions, years and rivers.
6. Determination of the relationship between nutrient status and the number of scoring species; and, between nutrient status and the percentage cover of macrophytes.

2.2.2 Robustness

Performance criteria:

- the baseline error in the MTR, in terms of the natural background variation, sampler errors and the inherent variation in operation of the method, should be minimal;
- the sensitivity of the MTR to the physical nature of the site should be minimal;
- the sensitivity of the MTR to the chemical nature of the site (other than nutrient status) should be minimal .

These criteria were assessed by examining the repeatability and reproducibility of the method, analysing the temporal variation in MTR within the survey system, and determining the relationship between MTR and selected physical and chemical variables.

2.2.3 Repeatability and reproducibility

Performance criteria:

- the MTR should be repeatable;
- the MTR should be reproducible.

The definitions used in the evaluation of the MTR system were as follows.

Repeatability: The characteristic of the MTR system which makes it possible for the same surveyor to use the same system at the same site, to produce MTR scores which are not significantly different from each other.

Reproducibility: The characteristic of the MTR system which makes it useable on a regular basis by different surveyors at the same site, producing MTR scores which are not significantly different from each other (given the constraints of interfering factors).

The first criterion, that of repeatability, was evaluated by means of a mathematical exercise. Performance against the second criterion was evaluated by analysis of data from Quality Assurance (QA) surveys.

2.2.4 Amenability for quality control

Performance criterion: quality control should be straightforward and confirm the original findings within acceptable limits.

This was assessed by considering options to reduce the various sources of variability and error within the method (assessed under 2.2.2 and 2.2.3).

2.2.5 National applicability

Performance criterion: the method should be applicable on a national basis.

With any plant based system, national applicability can be achieved by reducing the number of species assessed to a minimum number, which ensures that local rarities are not overvalued in some regions while being treated as commonplace in others. The system should use common species for assessment.

Evaluation of performance against this criterion thus addressed the following questions:

- is there regional variation in STR or MTR within the UK?
- is the taxa list sufficiently representative across geographical regions?
- are regional 'additions' or weightings required?

Ease of use

Performance criterion: the effort and expertise spent on gathering information must be proportional to the quality and usefulness of the information obtained.

Evaluation of performance against this criterion involved consideration of ways in which the operation of the method could be made easier, including modifications suggested by practitioners, and of the accuracy required of the data.

2.2.7 Cost-effectiveness

Performance criteria:

- the method must be at minimum cost commensurate with the ecological information required;
- the method must compare with the cost of other alternative methods.

This evaluation involved the consideration of whether more effort would provide better data upon which to judge trophic status, or whether the effort could be reduced, and a comparison with the cost of other methods used for routine biological monitoring.

2.2.8 Ease of interpretation/understanding by non-biologists

Performance criterion: survey findings must be in a form easily interpreted and communicated to non-biologists, to facilitate appropriate management decisions and assessment of priorities.

Evaluation of performance against this criterion involved consideration of different applications, including comparison of trophic status along rivers and between catchments.

2.3 Sources of data and information; preparation for analysis

In order to undertake the analyses required to evaluate the performance of the MTR against the above criteria, data were gathered from various sources. The primary source of data was the Agency's UWWTD MTR dataset (Table 1). A preliminary analysis of data from 1994 and 1995 UWWTD MTR surveys, however, revealed the need to supplement the Agency dataset in order to: cover a wider range of river types; check on some of the 'unexpected' results; extend the cover of the dataset to Scotland and Northern Ireland; and extend the comparison between the MTR and Diatom Quality Index (DQI). 'Unexpected' results were defined as those results not conforming to the hypothesis of decreasing scores with increasing P-loading. This need was confirmed in discussions with practitioners at the workshop described in 2.3.6. Additional data were, therefore, gathered as described in 2.3.2 - 2.3.5. [Data on direct seaward discharges and their seasonal variation in quantity, or on the consequences of multiple non-qualifying discharges, which could equate to QDs, were not considered in this study.]

Data were converted as closely as possible into a standard form for analysis. Thus, for example, the small number of cases where macrophyte cover had been recorded on a 3- or 5-point scale, were converted to the 9-point scale, as used in the standard MTR survey. Similarly, data contributed by the conservation agencies (2.3.5) relating to 500m survey reaches were transformed to give an equivalent MTR for a 100m reach (100m is the standard MTR survey length). Details of transformations, which include the assumptions made, are given in Appendix 1. Supplementary data, including environmental and chemical data, were used as available (Appendix 2).

Plant data were combined into a relational database, validated by staff from the Institute of Freshwater Ecology (IFE) and the Agency Regions, and any errors found were corrected. Under formal written agreement with collaborating partners, two versions of the database were produced for retention by the Agency. The first holds the complete data gathered for the project and is subject to access restrictions (see Project Record E1/i694/04, Dawson et al 1999, for details of access arrangements). The constituent components of this database, in terms of the different sources of data, are summarised in Table 1. The second version of the database holds only those data not subject to access restrictions, this including all data except those contributed by Scottish National Heritage (SNH), Countryside Council for Wales (CCW) and English Nature (EN). Although not a specific objective of the project it can be used for operational purposes on a single-user basis. A reference copy of this second version of the database, with its pre-programmed data input and outputs, is held with the Project Record on CD-ROM in a form compatible with Agency datasets (Microsoft Access 97, Dawson et al 1999). The Project Record includes instructions on how to operate the database.

Table 1a. Description of the datasets used within this project: Environment Agency, IFE and 'Conservation Rivers' datasets.

'Agency MTR'		
Source	Environment Agency (EA)	
Description	Approximately 230 sets of sites, mostly pairs, surveyed upstream and downstream of QDs in England and Wales for UWWTD monitoring purposes. Mostly 4 surveys in 3 years at each site; 1994-96, with some in 1993. Dedicated MTR surveys, using 100m survey length and 9-point macrophyte cover scale, although with some variation before 1995 (eg optional use of 3-point cover scale).	
No of sites/surveys	Sites: 523	Surveys: 1655
Physico-chemical data available	Physical: Yes (85% of sites)	Chemical: Yes
Purpose of using dataset	Data directly applicable to use of MTR for UWWTD designation purposes.	
IFE		
Source	Institute of Freshwater Ecology	
Description	Dedicated MTR surveys, undertaken specifically for the purposes of this project in 1996. Targeted sites in selected catchments. Standard survey methodology using 100m survey length and 9-point macrophyte cover scale	
No of sites/surveys	Sites: 105	Surveys: 117
Physico-chemical data available	Physical: Yes	Chemical: Yes
Purpose of using dataset	To check 'unexpected' results in EA dataset and to provide additional data on unpolluted rivers, rivers with differing geology, comparison of MTR and DQI and quality assurance.	
Conservation Agencies'		
Source	Conservation Agencies' Rivers Database. courtesy of English Nature (EN), Countryside Council for Wales (CCW) and Scottish Natural Heritage (SNH).	
Description	Surveys in England, Wales and Scotland, from 1978-1992. Not dedicated MTR surveys: usually two 500m surveys per site and a 3- or 5-point cover scale. Sites selected to determine conservation status over a wide range of physical and chemical types, particularly those of high conservation value in Britain.	
No of sites/surveys	Sites: 1563	Surveys: 3128
Physico-chemical data available	Physical: Yes (some different categories to MTR methodology)	Chemical: No
Purpose of using dataset	To supplement EA dataset with surveys from a range of high quality rivers; and to test applicability of the methodology to Scotland.	

Table 1b. Description of the datasets used within this project: 'Northern Ireland' and RHS Benchmark datasets.

'Northern Ireland database'		
Source	DoE/IRTU Surveys undertaken by Industrial Research & Technology Unit (IRTU) for Department of the Environment (DoE), Northern Ireland.	
Description	MTR surveys in Northern Ireland in 1995. One survey per site at routine biological monitoring sites, using 100m survey length and 9-point macrophyte cover scale.	
No of sites/surveys	Sites: 271	Surveys: 271
Physico-chemical data available	Physical: RIVPACS data recorded but were not used in this project	Chemical: Yes
Purpose of using dataset	To supplement EA dataset and test the applicability of the methodology in Northern Ireland.	
'RHS Benchmark',		
Source	Environment Agency (R&D Project 611)	
Description	Data from selected surveys in England, Wales, Scotland and the Republic of Ireland, were made available for MTR evaluation. Not dedicated surveys; macrophyte surveys undertaken alongside River Habitat Surveys (RHS) in 1994-98, usually 500m surveys lengths and a 5-point cover scale. Site selected were high physical quality river habitat, to allow calibration of RHS habitat quality scores.	
No of sites/surveys	Sites: 110	Surveys: 110
Physico-chemical data available	Physical: Not fully compatible	Chemical: No
Purpose of using dataset	To supplement the EA dataset with data from near pristine sites such as low-nutrient lowland sites.	
Total MTR database		
No of sites/surveys	Sites: 2572	Surveys: 5281

2.3.1 Environment Agency/NRA UWWTD surveys

MTR surveys were undertaken by the NRA/Environment Agency for UWWTD monitoring purposes mainly between 1994 and 1996, but with some also undertaken in 1993. The standard survey strategy was to survey pairs of physically similar sites, upstream and downstream of QDs, four times over a period of three years. The standard survey length was 100m and only plants in the channel, not the bankside, were recorded. This included scoring and non-scoring taxa. The physical characteristics of the site were also recorded. Survey data from all Agency regions (mostly as paper copies of field sheets) were collated by IFE and, except for data on non-scoring taxa, were input to the database for this study.

As the programme of surveys was, from necessity, being undertaken in parallel with development of the method, there were some changes between years to the standard guidance on methodology issued to surveyors. Although these changes must be allowed for when interpreting results of the MTR evaluation, they were either not believed to significantly change results, or were amenable to suitable data transformations. Examples of the changes include the following.

- The recommended species checklist for 1994 surveys was that used in the field sheet of Harding (Standing Committee of Analysts, DoE 1987). In 1995 and 1996, the checklist of MTR scoring species had been developed and was recommended for use.
- The guidance for surveys in 1994 allowed greater flexibility in operation of the method, which resulted in differences between Agency Regions and Areas. For example, one Region undertook ten 10m surveys at each site, rather than one 100m length, the data from which required transformation to the standard form to make it equivalent to a 100m survey length. In another example, the option to use a 3- or 5-point cover scale in 1994, instead of the 9-point scale subsequently recommended, required mathematical conversion (Appendix 1). Survey length and cover scales were standardised, however, by 1995.

Chemical data were input, where available, as mean values over a period of 2-5 years, for sites both upstream and downstream of WWTWs or close by. Additional data on the position of WWTWs and some data on phosphate stripping were also input to the database.

2.3.2 IFE (current project)

One hundred MTR surveys together with a small number of DQI surveys were specified and undertaken by IFE, within the terms of this project. The choice of sites was based primarily upon analysis of the initial database but was also as a result of discussions and the need for representation from each region of the Agency. Sites on the Rivers Coquet, Creedy, Danby, Dove, Eden, Edenbrook, Erewash, Erme, Frome, Gram, Great Stour, Meden, Mole, Otter, Ryton/Anston Brook, Severn, Sowe, Stour, Teme, Waveney, Weaver, Wye/Lugg and Yeo were chosen. The sites were selected to fulfil the following specified criteria:

i) to examine the unexpected results from the UWWTD MTR dataset by re-surveying upstream and downstream pairs of sites and then expanding, as necessary, to select more appropriate alternative sites in the locality (35, 47, 52);

ii) to determine the natural variation in MTR along relatively unpolluted river systems by surveying along the length of such rivers (20, 23, 35; Rivers Eden, Wye/Lugg & Coquet);

iii) to compare MTR and DQI (TDI) surveys in small catchments (15+10, 26+13, 30, Rivers Lodden/Blackwater & Kennet);

iv) quality assurance of Agency and IFE surveys (20+10, 10+12, no overlap but 30

additional re-surveys of Agency surveys 2 weeks to one month after the primary survey; rivers from each region of the Agency); and,

v) to determine the variation in rivers with differences or changes in geology along their length (10, 5, 27, Rivers Stour & Avon).

[Note: numbers of sites given within brackets for a) fulfilment of the contract, b) the achieved number of sites and c) the resultant number of surveys as a result of overlap of criteria.]

Methodology was as used for the Agency UWWTD surveys in 1996.

Macrophyte and physical data were input from completed field survey forms. Chemical data on the water quality were input for each site after analysis using standard methodologies.

2.3.3 Northern Ireland

MTR surveys on rivers in Northern Ireland were undertaken by IRTU during 1995 for the Environment and Heritage Service of DoE, NI, to determine the quality of the whole river network rather than just the impacts of WWTWs. Assessment of the latter were not a priority as many of the larger inland qualifying discharges (QDs) are already subject to phosphate removal. Phosphate removal is in place in virtually all QDs in catchments that do not discharge directly to the sea, ie Lough Neagh and Lough Erne (Hale, pers. comm.). The Lower River Bann, which is the outlet river from Lough Neagh, is further enriched by discharges from significant population centres such as Ballymoney.

Rivers in Northern Ireland tend to be enriched by the intensive use of fertilisers and by numerous small WWTWs and septic tank discharges. Conversely, the River Lagan, which discharges directly to the sea, receives WWTW inputs from 1 million people and the associated industries in the catchment in addition to the intensive farming of the middle reaches of the river. No nutrient-stripping is currently undertaken in this catchment (Hale, pers. comm.).

The survey methodology used was broadly similar to that of the Agency in England and Wales except in respect of the site selection. As sites were not selected as upstream-downstream pairs in relation to QDs, and as the positions of QDs have not yet been established in relation to the survey sites, MTR survey data from N. Ireland were not incorporated in analysis of downstream changes at QDs.

Physical data were recorded at corresponding RIVPACS sites but were not used in this project. Data on water chemistry were supplied by DoE, NI.

2.3.4 RHS surveys in the Republic of Ireland and Scotland

Plant species data collected as part of 'benchmarking' for the Agency's River Habitat Survey project (R&D Project 611) in the Republic of Ireland and Scotland during summer 1996, were made available for the MTR evaluation. These data were transformed into MTR survey data form by the main surveyor N.T.H. Holmes, and provide data on low nutrient, lowland river sites. Data for such sites were required for calibration purposes to establish the MTR expected for un-enriched lowland rivers. The sites included the River Unshin in west Ireland, which is a high-quality, moderately-sized, predominantly un-dredged river with a WWTW located in the middle reaches of the surveyed length of river.

Chemical data on the water quality were input for each site after analysis using standard methodologies. Physical data were input from completed field survey forms.

2.3.5 Scottish Natural Heritage (SNH), Countryside Council for Wales (CCW) & English Nature (EN)

Data owned by and used courtesy of SNH, CCW and EN were extracted and converted from their Rivers Database (called herein the Conservation Rivers (CR) Database) for use in clarifying the potential MTR ranges along river systems; to extend the range of trophic states and river types covered in the MTR database; and to assist the setting of standards of confidence for cross river or region comparisons. The survey data were based upon two adjacent surveys of 500m lengths initiated in the early 1970s. Conversion of some parameters was necessary, particularly for early data (see Appendix 1) and for the absence of the smaller category of river width (< 1m), the smallest width group being 0-5m.

The use of these additional data courtesy of SNH, CCW and EN was of great value to the MTR evaluation undertaken within this project. Although suitable transformations of data were possible, future development of the method would be enhanced by continued collaboration between the national regulatory and conservation agencies, and by standardisation of data collected between the agencies.

Data on chemistry were not available for sites in the Conservation Rivers database but were obtained by matching Environment Agency data from the 7510 GQA monitoring sites regularly sampled in England and Wales, on the basis of a GQA site being within a short distance of a sampled site. No similar data were available for Scotland, reducing the usefulness of the Conservation Rivers database by 25% and to 101 of the 129 scoring species as many of the rarer species were recorded in Scotland. A range of distances from GQA sites were considered from plus or minus one kilometre to ten kilometres in the easting and northing directions (Table 2). For 1 km, the GQA site was within a 4 km^2 area centred on the plant survey site and so forth. All matches up to ± 2.5 km were checked on maps and discarded if they were not on the same river or if there was an input from a WWTW or tributary between the sites. Above this distance it was estimated that a coincident phosphate match would be available for 90% of cases and thus a distance of 5 km would seem to be an optimum distance for more general comparisons.

Analysis was generally undertaken on the more cautious basis of ± 1 km, which resulted in a set of 394 sites (788 surveys) with matching CR plant and GQA phosphate and nitrate data, widely distributed throughout England and Wales (Figure 1a). However, of these sites, only 85 had been surveyed for plants after 1985 (Figures 1b & 2). The effects of changes of nutrient concentrations over time, or of plant colonisation rates, were not investigated

Table 2. Number of Conservation Rivers sites with a GQA chemical sampling point occurring within a short distance.

Distance (km)	Number of matches	Percentage of total
0	91	5.8
± 1	394	22.0
± 2.5	862	55.2
± 5	1072	68.6
± 10	1157	74.0

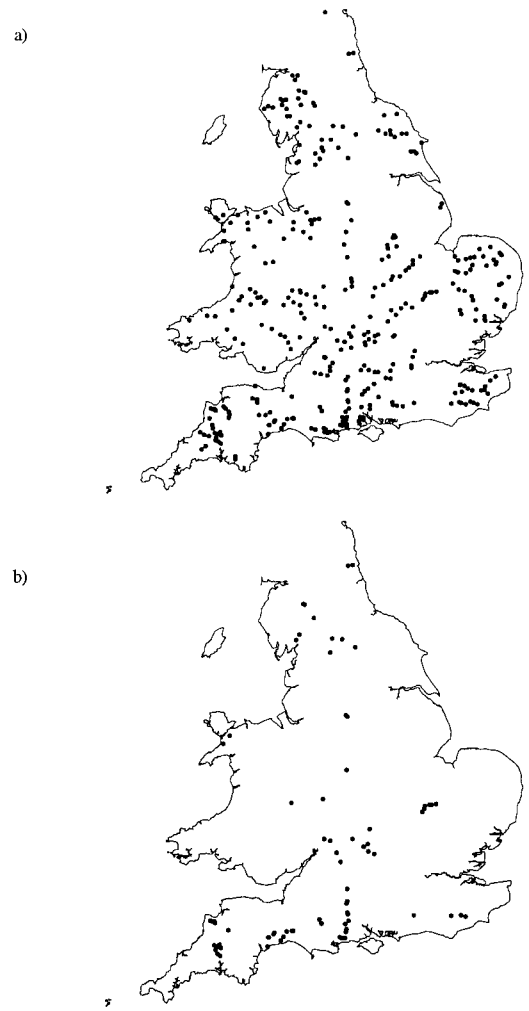


Figure 1. Distribution of Conservation Rivers sites in England and Wales for which a GQA chemical sampling point occurs within one kilometre on the same river: (a) all matched sites and (b) sites where CR surveys were undertaken after 1985.

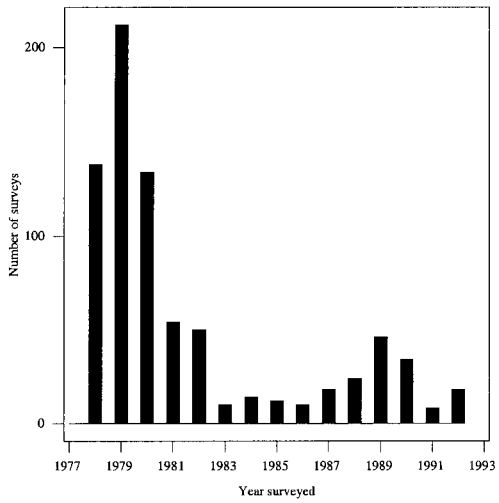


Figure 2. Time schedule of the Conservation River surveys for which a GQA chemical sampling point occurs within one kilometre on the same river.

2.3.6 Feedback from practitioners

Useful comment and feedback from practitioners of both the MTR and Trophic Diatom Index (TDI) were gained at a discussion workshop held in Lancaster shortly after commencement of the project (March 1996). The workshop served both this and the related TDI R&D project (Environment Agency R&D Project i618, Kelly 1996a) and was attended by delegates from all NRA Regions, in addition to the authors of the two methods and other external interested parties. Experiences and ideas on the two methods were exchanged; the usefulness of the methods for both UWWTD and other applications discussed; and recommendations made relating to the two R&D projects, the management of the Agency's trophic status monitoring programme, and future R&D requirements. A full and a summary report of proceedings were produced and distributed to delegates and are included in the Project Record (Newman et al 1997a & b, Dawson et al 1999). Key conclusions and recommendations of relevance to this project were as follows.

1. Both TDI and MTR are capable of detecting differences in the trophic status downstream of QDs, although the sensitivity of the methods may be dependent upon the level of phosphate and/or other nutrients upstream of the discharge.

2. The importance of complicating factors such as direct organic pollution is separable with the TDI but not at present with the MTR and should be investigated in the latter. A grid to facilitate interpretation of results by taking into account the relative influence of organic pollution and nutrient enrichment, as has been developed for TDI, should be investigated for MTR.

3. The validity of 100m reaches for MTR survey purposes should be established.

4. Minor revisions to, and clarifications of, the way MTR data are collected and recorded are necessary.

5. Consideration should be given to the reproducibility of biomass estimates in MTR surveys. Guidance on how and when to record this, plus interpretation, is required.

6. The introduction into the MTR system of regional weightings for local taxa (nationally rare) and for taxa at the edge of their geographical range should be investigated.

7. The application of a weighted average value to the MTR should be considered (cp. indicator value as used in the TDI).

8. Consideration should be given to identifying those taxa which respond quickly to nutrient inputs and/or changes in nutrient levels, on either a spatial or temporal scale of response.

10. The inherent variability of the MTR method should be defined.

11. Situations in which the MTR gives values which do not correspond to the hypothesis of decreasing scores with increasing P-loading ('unexpected results') should be investigated further.

12. Further trialing of both TDI and MTR is necessary in all Agency Regions to establish the relationship between the two methods.

13. Both the TDI and MTR should operate to the same scale and direction. Actual scores and percentage downstream changes in scores will be used for the purposes of the UWWTD. For other purposes, such as large scale national maps of eutrophication, a banding system should be developed if appropriate.

14. Both TDI and MTR have much wider applications than monitoring eutrophication for the purposes of the UWWTD, such as assessing the impact of non-qualifying discharges, tracking non-point source pollution and measuring improvements in the aquatic habitat. English Nature and the Department of the Environment (Northern Ireland) have already trialed the use of MTR for such purposes.

As a result of the workshop, modifications recommended in (4) and detailed in the workshop report (Newman et al 1997b) were incorporated into the draft standard methodology produced by the Agency for UWWTD monitoring (Environment Agency, May 1996). The methodology was circulated for use by Agency surveyors for their 1996 UWWTD surveys. These modifications have been retained in the procedural manual for MTR surveys produced from this project (Holmes et al 1999).

The recommendation given in (13) concerning the scale and direction of both methods was also adopted subsequent to the workshop. As a first step, the scale of the TDI was changed to 0-100 (Kelly 1996a). Secondly, the Agency's Regional Biologists endorsed the recommendation that both methods should operate in the same direction and decided that this direction should give decreasing eutrophy (increasing 'quality') with increasing score. As this was the direction of operation of the MTR, a transformation of the TDI was devised and a new name created for this transformed index to avoid confusion:

Diatom Quality Index (DQI) = 100 - TDI

This maximised compatibility between the two methods. Thus, when comparing diatom data with MTR, DQI should be used in preference to TDI.

The other recommendations were incorporated into the project plan for the evaluation of the MTR, the results of which are reported in the following sections of this report.

2.4 General description and suitability of data

The majority of the data gathered were not originally collected specifically and solely for the evaluation of the MTR, but for other purposes, these purposes differing between the datasets from the various sources. Sites were selected on a non-random basis, being targeted according to the specific purpose of the survey programme, and variations exist between the site selection criteria of the various datasets. For example, as Agency data were collected for UWWTD monitoring purposes, the distribution of sites related to QD distribution, based on a 100m survey length and a 9-point cover scale. This resulted in a bias towards large, low-gradient, lowland, enriched rivers, often with modified channels. Such sites are frequently less shaded (especially downstream of QDs) as a result of enlargement by dredging and/or re-sectioning to accommodate enhanced flows, and a relatively frequent requirement for tree removal to allow access for machinery (4.5.3). The selection of sites in the Conservation Rivers database, in contrast, was targeted towards rivers of high conservation value across a range of river types and based upon two 500m survey lengths and a 3- or 5-point cover scale.

In order to judge whether such non-random and varied data could be used to evaluate the performance of the MTR with any statistical validity, it was first necessary to establish: whether the range of the data was sufficient; whether the biases demonstrated within individual datasets, towards particular types of rivers, balanced each other out (ie whether the datasets complemented each other); and whether data from different sources could be used with the same confidence. In other words, the suitability of the data needed to be confirmed.

Questions to be addressed included:

- is the size of the database sufficient to allow statistically valid analyses?
- is the geographical distribution of sites of sufficient range, in terms of national and regional coverage?
- is there a sufficient range of physical river types represented, and are there biases between datasets?
- is there a sufficient range of chemical river types represented, and are there biases between the datasets?
- is there a sufficient range of macrophyte characteristics represented, and are there biases between datasets?
- are the data from the various sources of equal and suitable quality?

2.4.1 Size of database

The total data input to the database related to 2572 sites or 5281 surveys from Britain and Ireland. This was considered sufficient to give statistical validity to the analyses.

2.4.2 Geographical distribution of sites

The NRA/Agency survey sites reflect the distribution of QDs, which are fairly widespread throughout England but sparse in Wales (Figure 3). The number of surveys completed per year by each Agency Region has increased in the period from 1994 to 1996 (Figure 4) due to a greater number of sites being visited twice in a year in 1995 and 1996 (four surveys required over three years). The number of surveys completed by each Region reflects the number of QDs, which are most abundant in Midlands and Anglian Region, and least in Welsh Region.

The four additional datasets gave a fairly comprehensive national coverage (Figure 5) but a

full range of physical and chemical data were not available for all sites, giving a bias to the analyses of results (Appendix 2). There is a moderate degree of overlap in the location of Agency surveys and Conservation Rivers surveys. For several analyses, however, the CR dataset was restricted to surveys after 1985 as these could be matched to chemical data with more confidence than for earlier surveys.

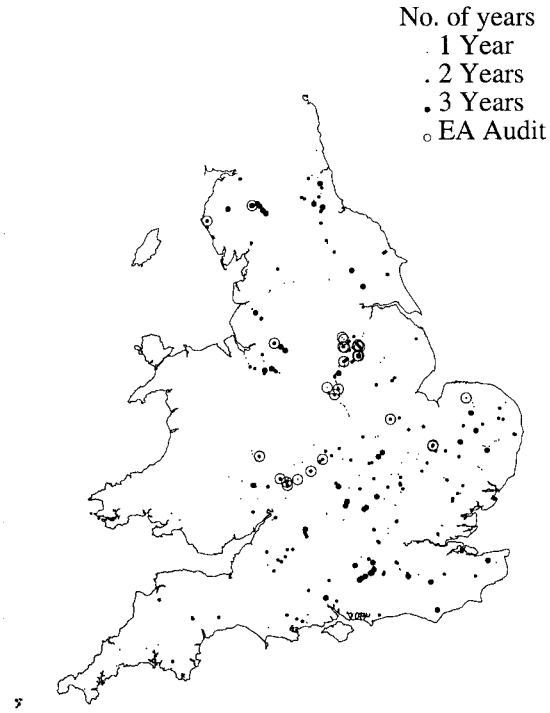


Figure 3. Location of MTR sites surveyed by National Rivers Authority or by Environment Agency, 1993-1996. The number of years surveyed at each site indicated by the size of closed circles; audit surveys indicated by open circles.

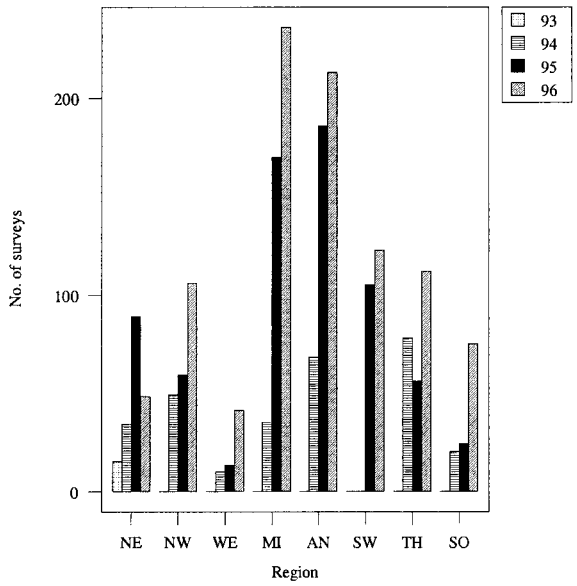


Figure 4. The number of surveys completed within each Environment Agency region in the years 1993-1996. Key to EA Regions: NE = North East; NW = North West; WE = Welsh; MI = Midland; AN = Anglian; SW = South West; TH = Thames; SO = Southern.

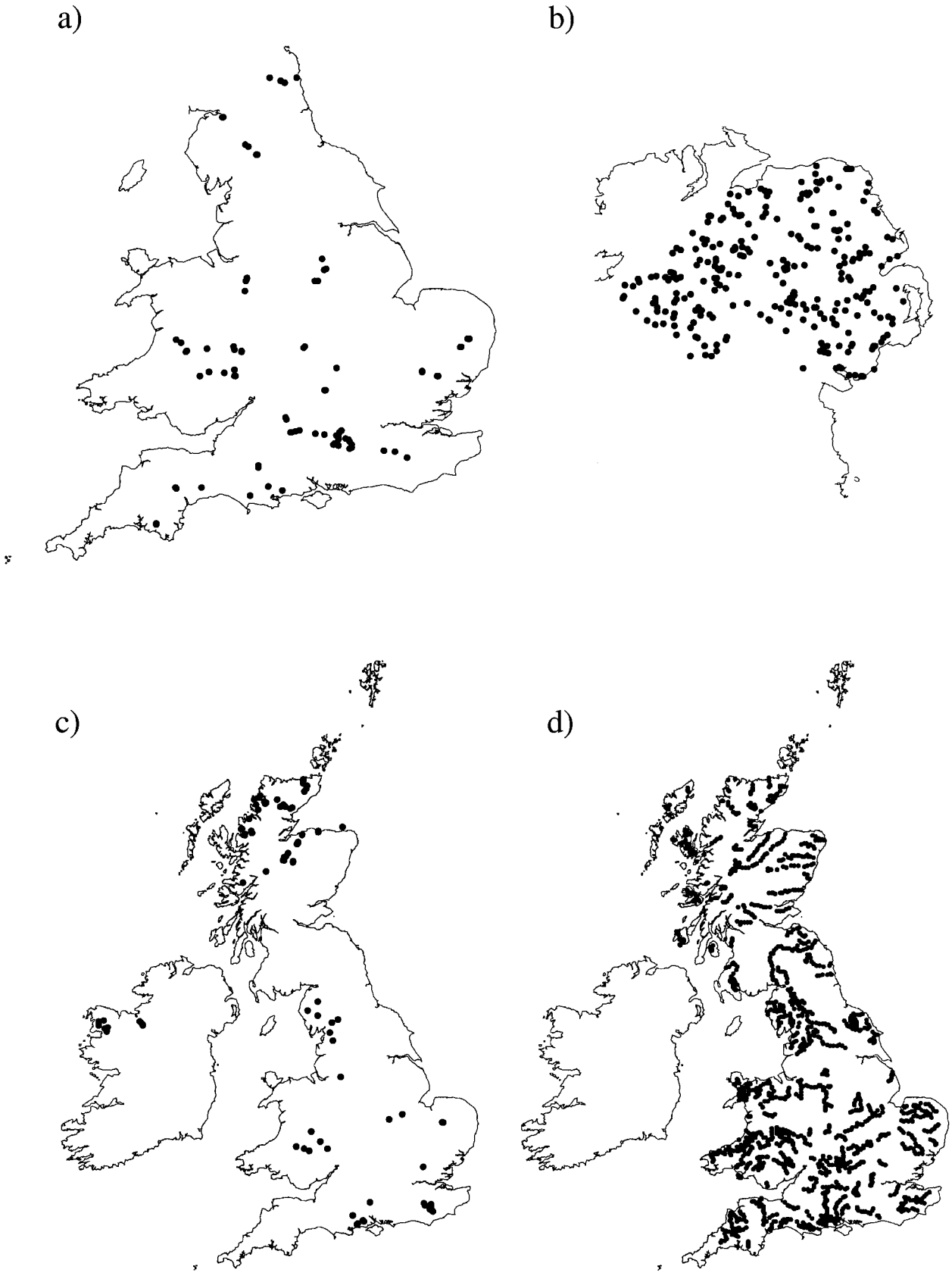


Figure 5. Location of sites surveyed by a) IFE, b) DoE/IRTU Northern Ireland, c) RHS benchmark sites and d) Conservation Agencies Rivers database.

2.4.3 Physical characteristics

Most plants have a specific or preferred range of habitat requirements, in addition to the regular supply of inorganic carbon for growth, macro- and micro-nutrients. These include light at their leaf surfaces, an acceptable water flow and depth, and a suitable substrate for attachment or rooting. If the habitat is less than optimum, growth may be reduced, other competitors may invade and the species may be lost from the reach. In such situations, however, a WWTW discharge or increase in nutrient concentration may coincidentally be identified as the cause of the loss of the plant species. It is important, therefore, to assess the influence of the physical habitat on the MTR. To make this assessment the database should ideally include a wide range of physical river types.

Physical data were available for most surveys. The rivers varied in depth, width, substrate size and the degree of shading, with differences apparent between different datasets. By comparison with the NRA/Agency surveys, the Conservation Rivers surveys were undertaken on sites which were generally shallower, narrower and with larger mean substrate size, ie smaller phi values (Figure 6). Data for comparison of shade were not available for Conservation Rivers. These differences may reflect the greater number of upper river sites in the Conservation Rivers dataset, or may relate to the greater number of lowland rivers or the greater management at the Agency survey sites. Research during the development of the River Habitat Survey (RHS) clearly shows that managed rivers, typical of downstream of QDs, are deeper, broader and have finer substrates than semi-natural rivers of the same type (Raven et al 1997). IFE surveys from 1996 also have, on average, larger substrates, due to the disproportionate effect of the inclusion of the upper Wye and Eden in this small dataset, cobbles and pebbles predominating in these rivers. Details of the completeness of the datasets are given in Appendix 2.

Although vegetation itself can influence the physical character of a river (for example, surface floating vegetation can increase shade for other plant species) no data were available apart from named species (eg *Lemna* spp or *Cladophora* spp) when present.

The initial classification of rivers to RHS river types was not undertaken for the MTR sites although a preliminary assessment showed that the Agency survey sites mainly lie in the lowland classes and on mixed or soft rocks. The Conservation Rivers sites, however, are more likely to reflect the general distribution of river types in Britain, despite the survey focusing on main river channels without a sufficient representation of tributaries.

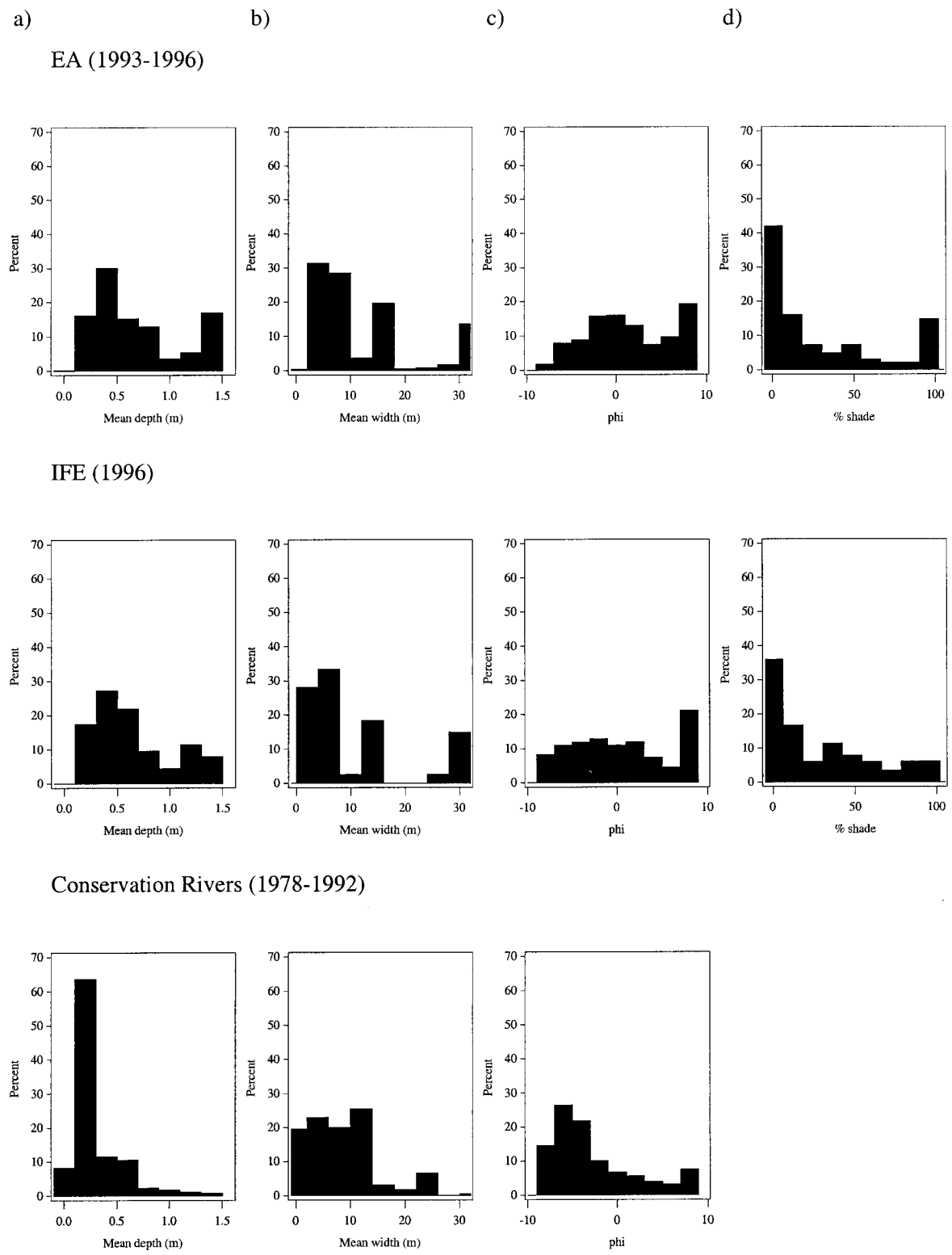


Figure 6. Frequency histograms of a) estimated mean depths, b) estimated mean widths, c) mean substrate size and d) shade for EA, IFE and Conservation Rivers datasets. Note that there were no shade data for Conservation Rivers (full details in Appendix 2). Phi sizes range from -8 for boulders through 3 for pebbles, to 5-10 for coarse to fine silt; see appendix for method of estimating mean depth and width.

2.4.4 Chemical characteristics

Given a favourable physical habitat, growth of a plant species will also be dependent on the chemical characteristics of the site. These include a sufficient concentration of nutrients and micro-nutrients required for growth, but also the absence of those chemicals which inhibit growth. Species-specific differences in growth responses to these chemical characteristics will influence the resulting species assemblage. The MTR is based upon the assumption that it is possible to detect the particular response of the assemblage to one of these chemical characteristics – nutrient status, and in particular, phosphate-phosphorus. Evaluation of the performance of MTR thus requires data over a wide range of phosphate concentrations, but also a range of nitrate-nitrogen concentrations and other chemical determinands which may influence the species assemblage and hence the performance of MTR in assessing trophic status.

Data on phosphate were needed to test both the relationship with MTR scores and with growth of individual plant species in order to validate the STRs assigned to species. Data were used for soluble reactive phosphate, dissolved, or analysed as being present in this form in the water, as this is generally considered to reflect the phosphorus available to biological processes. Results given throughout this report for concentrations of nitrogen and phosphorous are expressed in terms of phosphate-P and nitrate-N.

Although regular efforts were made to ensure a complete or at least a uniform geographic representation of all chemical determinands in relations to QDs, data on water chemistry were absent from several sites and from some areas of the country, particularly from parts of Cornwall, Wales and north-east England. Despite this, however, the combination of the data on QDs, Conservation Rivers and the survey of Northern Ireland provided a reasonably comprehensive set of the water chemistry data. A summary of the completeness of the chemical part of the database is given in Appendix 2.
The database incorporated a wide range of phosphate concentrations, although with much bias in individual datasets. Annual mean concentrations at sites up- and downstream of QDs (NRA/Agency & IFE data) ranged from above 4 mg l^{-1} in 4% of cases to less than 0.03 mg l^{-1} (solid in histogram, Figure 7). The concentrations in Northern Ireland and Wales were generally lower than in England and this brings the mean value for England, Wales and Northern Ireland downwards to just below 1 mg l^{-1}. It should be noted, however, that the detection limit used for the analyses of Agency and IFE samples (0.02 mg l^{-1}) was lower than that used by DoE/IRTU for samples from Northern Ireland (0.05 mg l^{-1}). The geographical distribution of higher phosphate concentrations reflects the distribution of rivers flowing away from the inland centres of population in England and Wales (Figure 7).

Nitrate concentrations showed a similar general regional pattern to phosphate although there were regional differences in the ratio between nitrate and phosphate (compare Figures 7 and 8). These differences reflect the lower nitrate levels in the east and centre of England and in Northern Ireland, compared to the elevated phosphate levels of the north-west midlands and a broad belt of southern England on more calcareous rock.

Data on determinands other than phosphate and nitrate were of more limited availability. Although eight more easily available determinands showed a variety of distributional patterns, none would exclude the growth of plants (Figure 9a-h). The presence or effects of toxic compounds or 'metal impacted' streams were not investigated.

a)

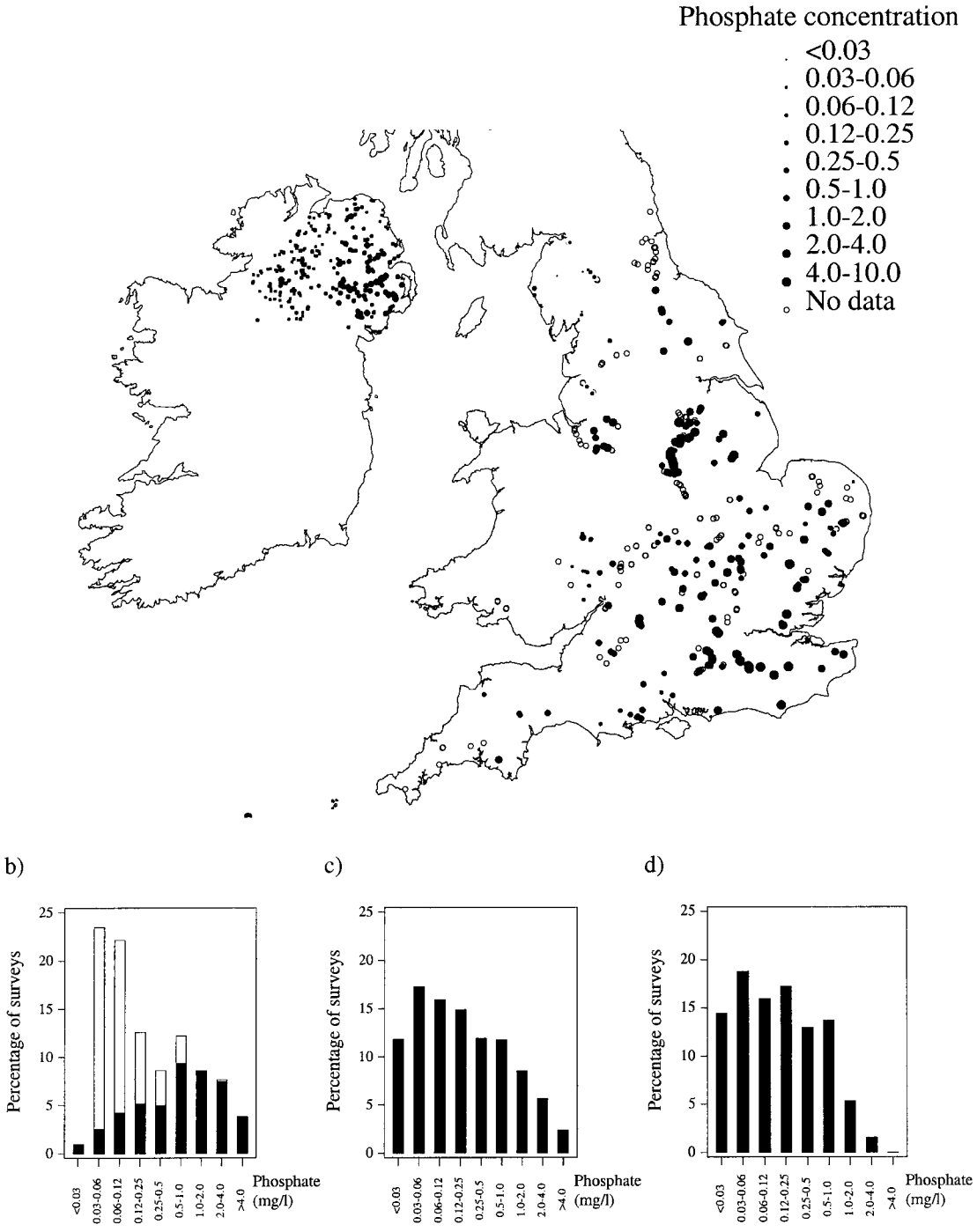


Figure 7. Distribution of the annual mean soluble phosphate concentrations (mg l^{-1}) for MTR survey sites in England, Wales and Northern Ireland. (a) & (b) 1994-96 MTR survey sites, with EA/IFE sites as solid blocks and DoE, NI, as open blocks in the histogram (phosphate data were not available for all MTR sites). (c) The complete GQA phosphate data of England and Wales. (d) Phosphate data matched for the Conservation Rivers for sites within 5 km for all years.

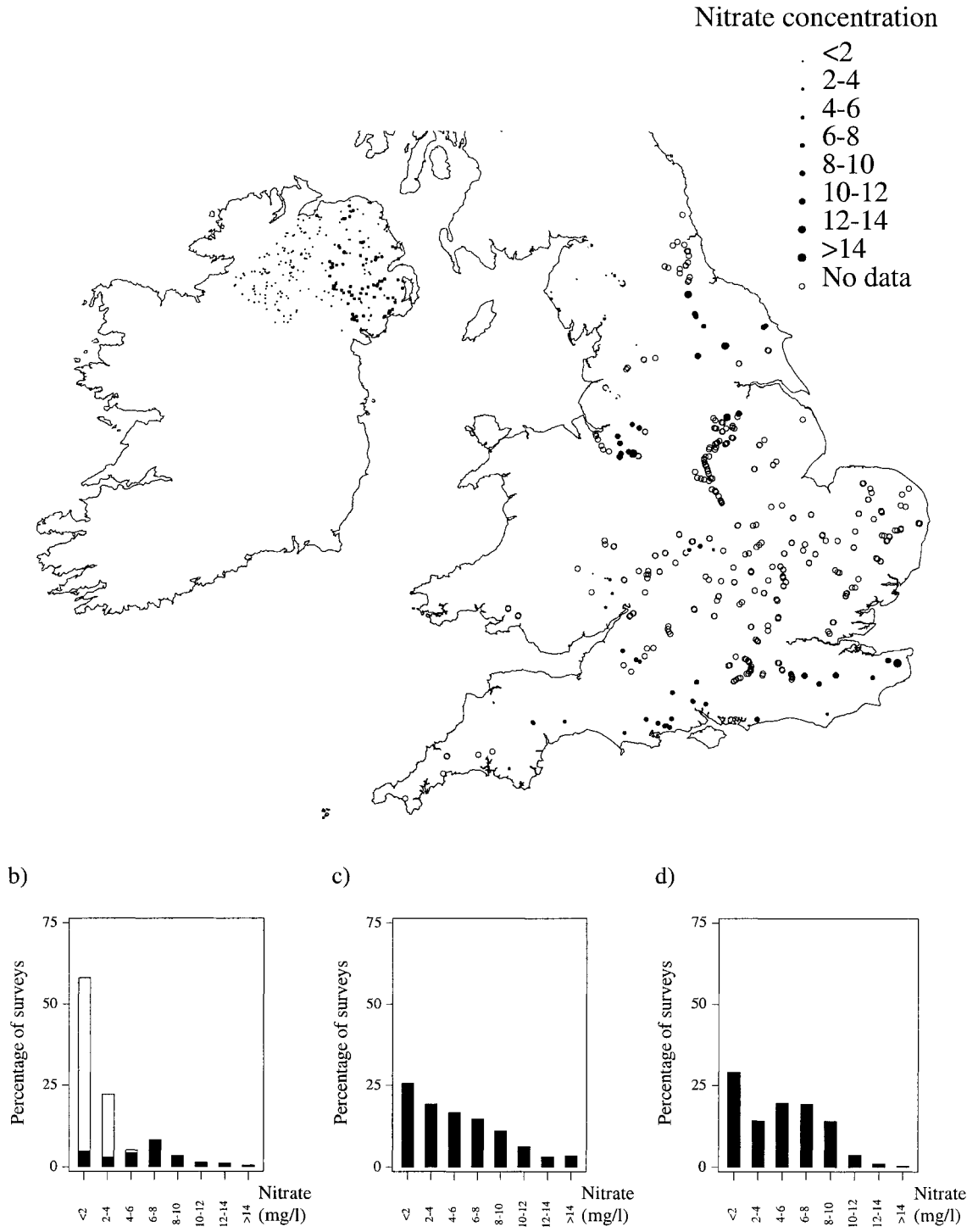


Figure 8. Distribution of annual mean soluble nitrate concentrations (mg l^{-1}) for MTR survey sites in England, Wales and Northern Ireland. (a) & (b) 1994-96 MTR survey sites with EA/ IFE as solid blocks and DoE, NI, as open blocks in the histogram (nitrate data were not available for all MTR sites). (c) The complete GQA nitrate data of England and Wales. (d) Nitrate data, matched for the Conservation Rivers dataset within 5 km for all years.

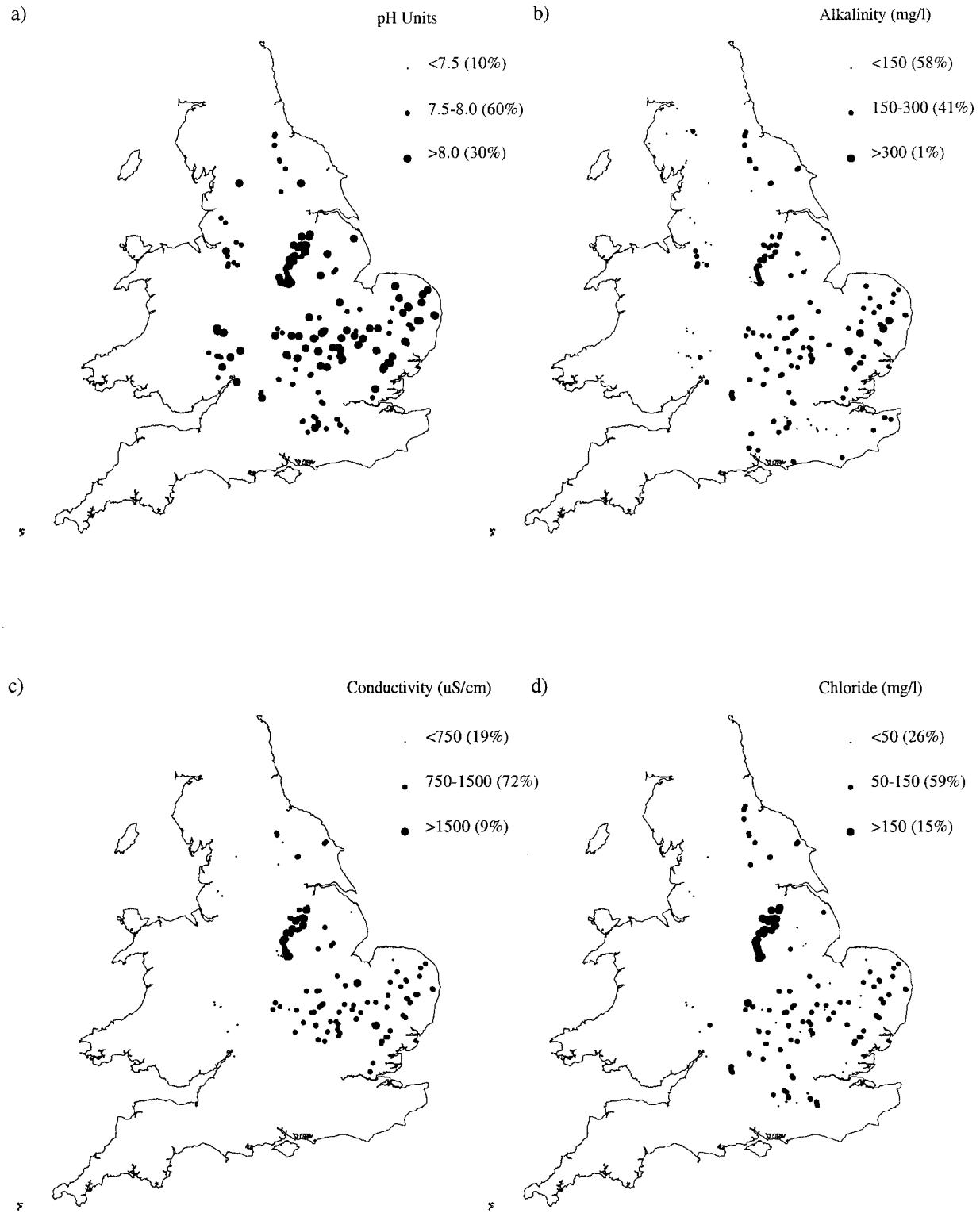


Figure 9. Distribution of annual mean a) pH, b) alkalinity, c) conductivity and d) chloride concentrations for EA MTR sites in England and Wales for 1994-1995. The percentage frequency of sites for each categories is given in parenthesis. Mean pH used as an approximation to the pH derived from the mean OH^- concentration.

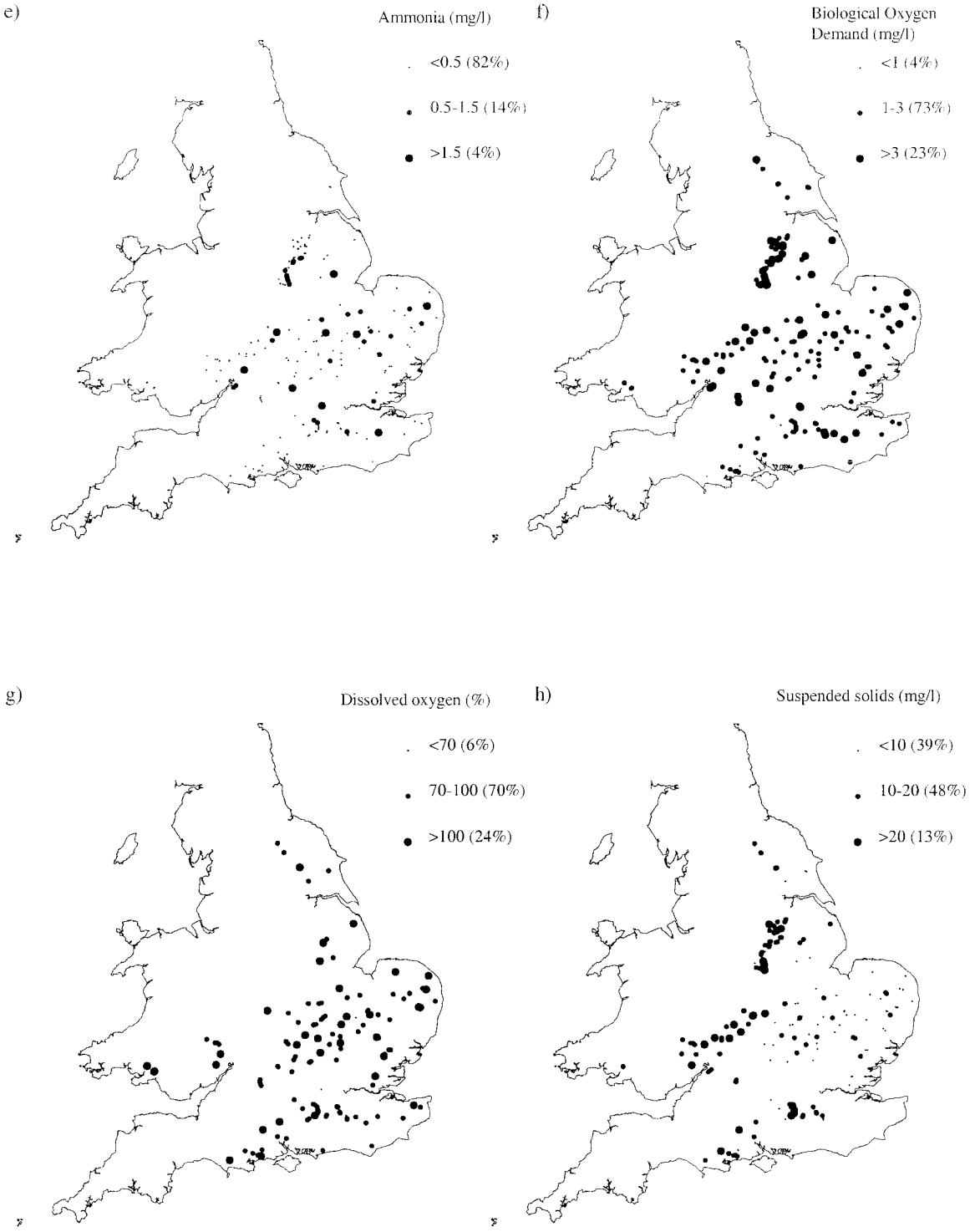


Figure 9. Distribution of annual mean e) ammonia, f) Biological Oxygen Demand (5-day) g) dissolved oxygen and h) suspended solids for EA MTR sites in England and Wales, 1994-1995. The percentage of sites in each category is given in parenthesis.

2.4.5 Macrophyte characteristics

All 129 scoring species were recorded in the combined database, although not in all component datasets (Figure 10a-c). All scoring species were recorded in the Conservation Rivers dataset, including several rare species. A more restricted set of 109 scoring species were recorded in the Agency dataset; three species of liverworts, seven of mosses, five of dicotyledons and five of monocotyledons were not recorded in the Agency dataset.

An average of 41.7 species were recorded per Conservation River survey, of which an average of 16.3 (39%) were scoring species and an average of 9.1 (22%) were highlighted species. Only half the number of scoring species were recorded for surveys in the Agency dataset compared to the Conservation Rivers dataset, with an average of 8.3 species per survey, but these had a higher proportion of highlighted species (6.6 scoring species per survey). Non-scoring species were not entered into the database for Agency surveys. Highlighted species represented 77% of the commoner species, ie those occurring at more than 10% of surveys.

When the macrophyte characteristics are expressed in terms of the MTR, the Agency dataset is shown to be biased towards low MTR sites with fewer scoring species in comparison to the Conservation Rivers dataset (Figure 11). The latter is biased towards higher conservation status (and higher MTR), but as such compliments the Agency dataset. Distributions of the number of highlighted species are similar for both datasets and hence confidence in scores should be comparable

The differences noted above in the number of species recorded during Conservation River surveys compared to Agency surveys are likely to be a reflection of the differences in the way the data were collected, as well as differences in the nature of the sites sampled. The Agency and IFE surveys were predominantly of 100m using a 9-point cover scale, whereas the Conservation Rivers and RHS benchmark datasets were from 500m surveys with a 3- or 5-point cover scale. The longer survey length results in more species being recorded per survey, but does not appear to affect the MTR score (5.3.2, Figures 52 & 53). Similarly, although the 3- or 5-point cover scale data require transformation before they could be used in this project, the resulting MTRs should not differ significantly from if a 9-point scale had been used (at least for the 5-point scale, see 5.2, Figure 51). The differences in MTR recorded in the different datasets are thus likely to reflect real differences in the flora and trophic status of the sites surveyed rather than methodological differences.

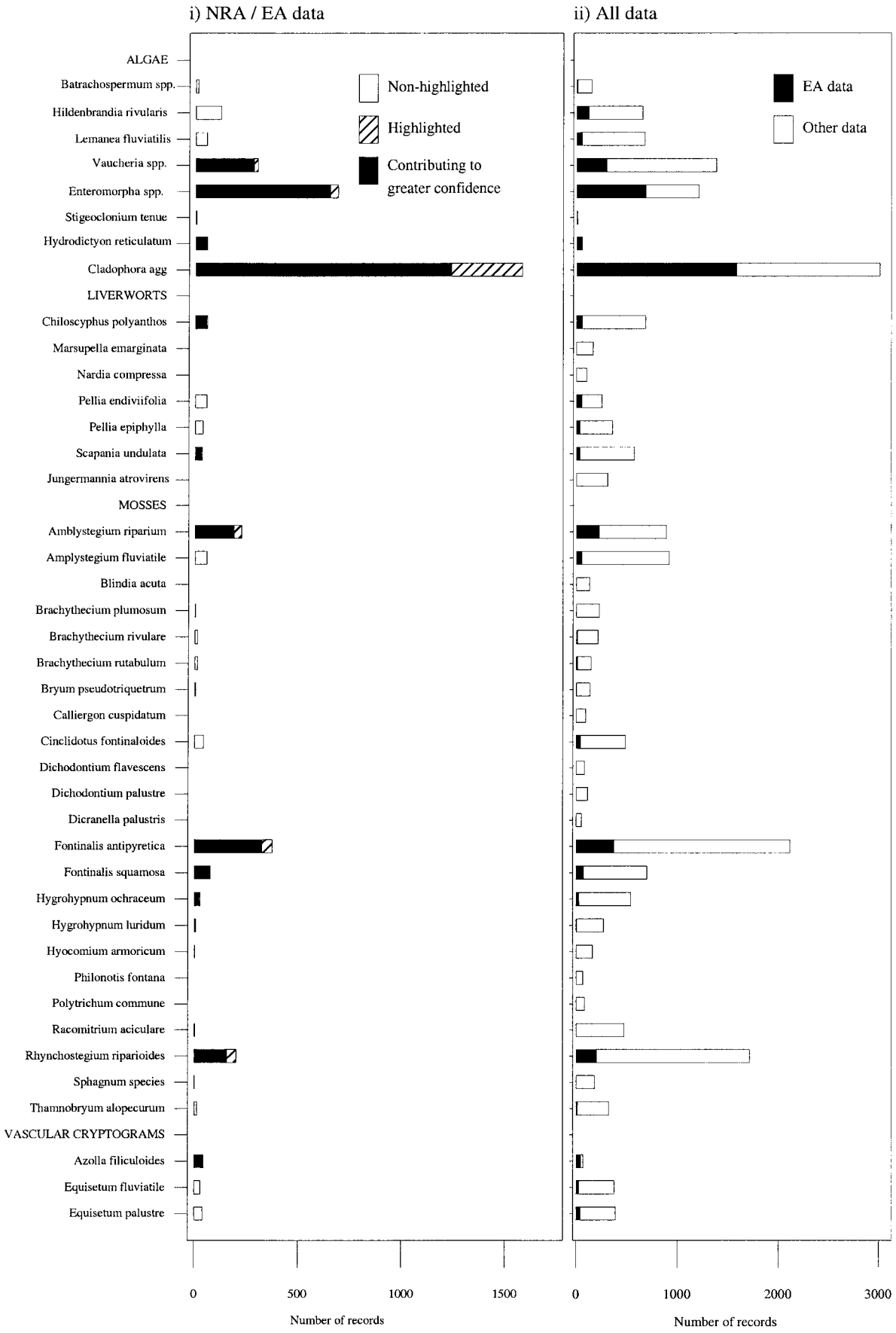


Figure 10a. The frequency of recording of scoring species of algae, bryophytes and vascular cryptograms during i) NRA/EA MTR macrophyte surveys 1993-96 and ii) all surveys used in this study). Records where more than 5 highlighted species were present at the site are shown as solid bars.

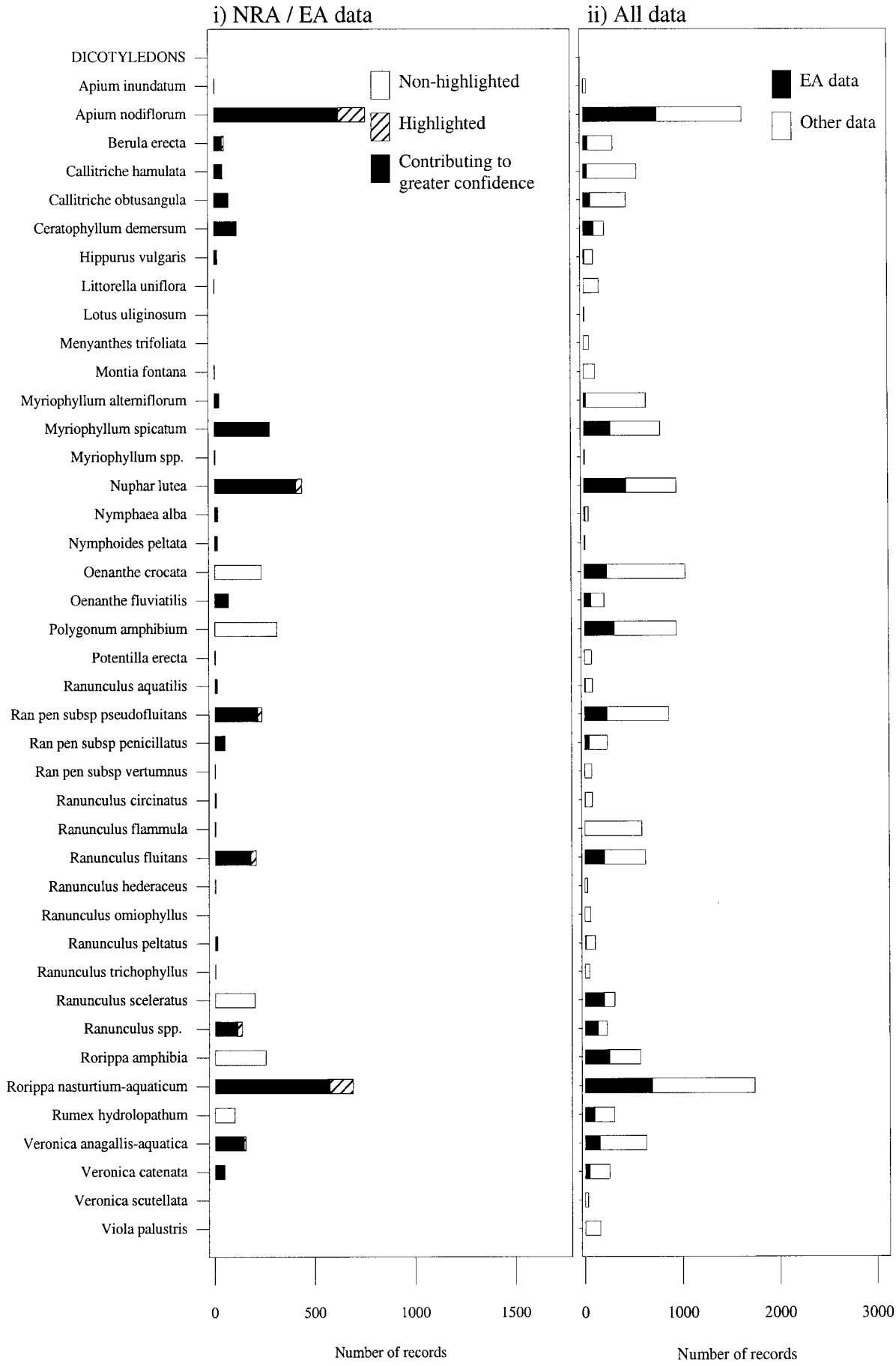


Figure 10b. The frequency of recording of scoring species of dicotyledons during i) NRA/EA MTR macrophyte surveys 1993-96 and ii) all surveys used in this study.

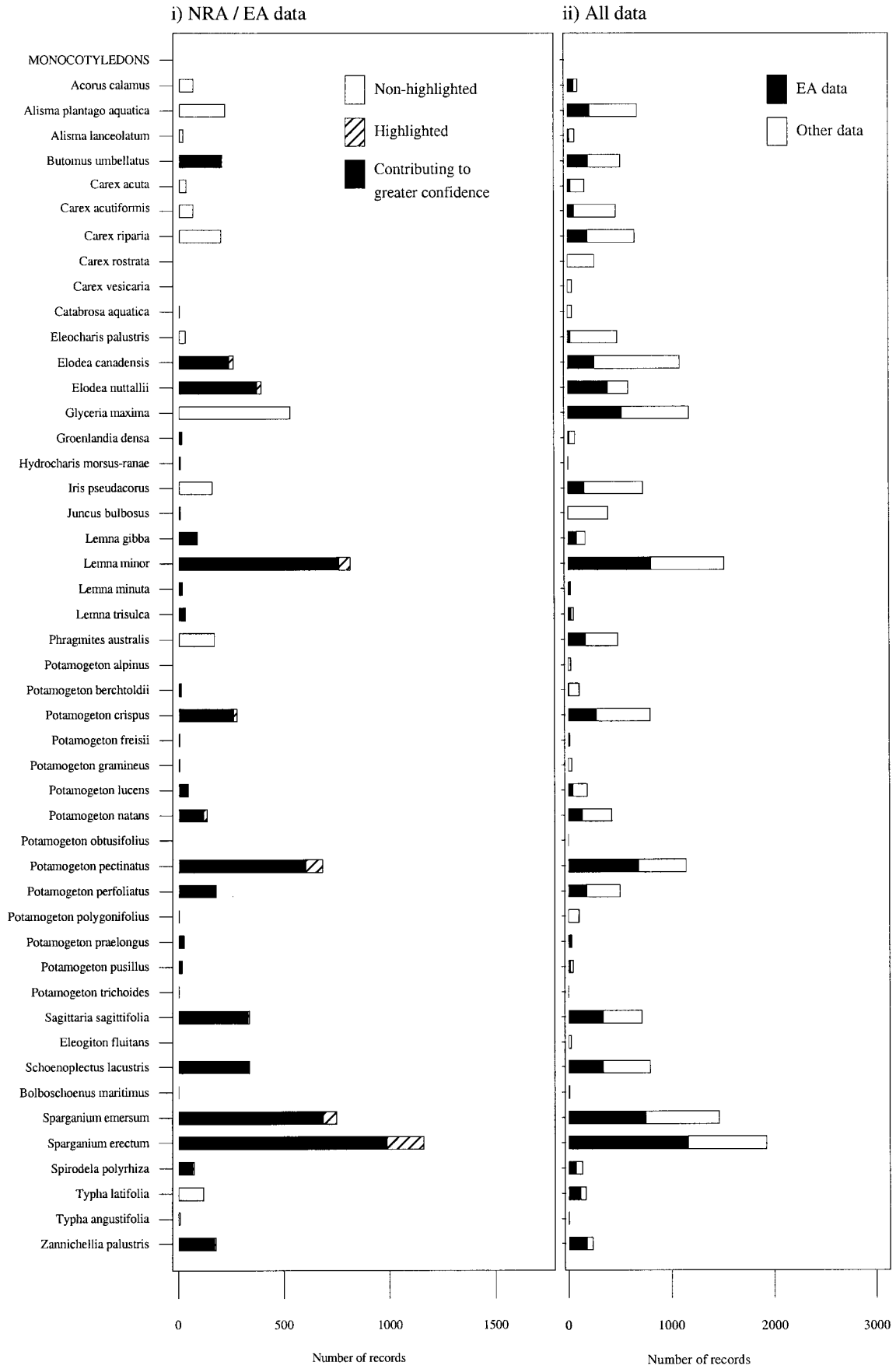


Figure 10c. The frequency of recording of scoring species of monocotyledons during i) NRA/EA MTR macrophyte surveys 1993-96 and ii) all surveys used in this study.

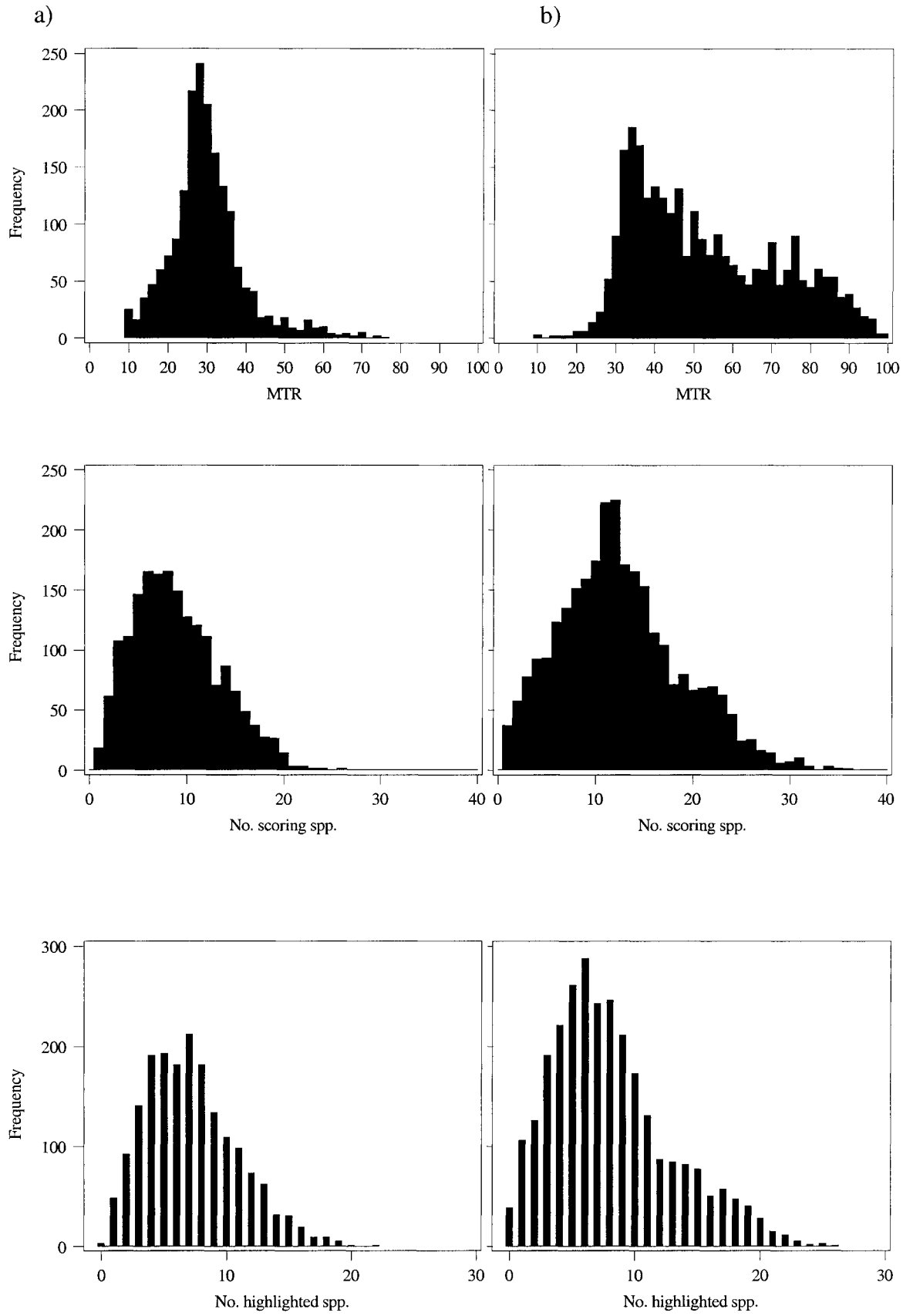


Figure 11. Frequency distribution of MTR score, number of scoring species and number of 'highlighted' species from (a) EA dataset and (b) Conservation Rivers dataset.

2.4.6 Quality of the macrophyte data

Quality assurance and quality control procedures relevant to MTR surveys are discussed in Chapter 4, together with an outline of results and recommendations. The purpose of this section is to determine whether the macrophyte data used for the MTR evaluation were of sufficient quality for this purpose, and whether there were any differences in the quality of data between datasets from different sources.

A full quality assurance (QA) of the available data was not possible. There are several reasons for this. Much of the data had been collected prior to the project commencing; there were slight differences in the MTR methodology used by the Agency over the period for which the data were collected; and where QA re-surveys were undertaken, there was no standardised feedback mechanism in place to correct mistakes detected as a result of the QA procedures. Added to this are the inherent problems associated with quality assurance of macrophyte surveys, as described in Chapter 4.

Assessment of the quality of macrophyte data was thus limited to analysis of the results of a small number of re-surveys of Agency sites, the results of which are described in Chapter 4. Such re-surveys do not give the definitive survey result: they only indicate the consistency of the survey quality. The analysis indicated consistency in quality for approximately two-thirds of Agency surveys and highlighted several reasons for inconsistencies in the remainder. There was also some evidence, from comparisons of surveys in different years, that the quality of macrophyte data increased slightly during the survey period 199(3)4-1996; this was assumed to be due either to increased surveyor experience, or to changes in the species checklist used, within this period.

As a full assessment of the consistency of quality throughout the Agency's MTR survey programme was not possible, and as no such assessment could be undertaken on data contributed by DoE, NI, or the conservation agencies, the working assumption was made for the purposes of this project that the quality of the macrophyte survey data was suitable for evaluation of the MTR, and that the quality of each component dataset was equal.

2.4.7 Conclusion: Confirmation of suitability of data

The overall size of the database was considered sufficient for statistical confidence to be obtained in results of analyses.

The geographical distribution of sites was considered to give sufficient national coverage, although the incompleteness of some of the datasets (eg no chemical data for Scotland; no direct phosphate data available for the Conservation Rivers; few data for Wales in the UWWTD dataset) will bias applicability of the MTR evaluation towards England.

The range of physical, chemical and macrophyte characteristics of sites was also considered to be sufficient to analyse the influence of the former two on the latter. The datasets complemented each other, extending the range from that available purely from the Agency UWWTD dataset and thus justifying the data collation strategy. The incompleteness of some of the datasets (eg most of the direct chemical data related to the UWWTD sites, which are biased towards large, lowland, enriched rivers) will, again, bias analyses towards a limited range of types of sites. Future development of the method would be benefited by gathering data from sites with a wider range of physical substrate.

The working assumption was made that the quality of the macrophyte data was consistent between different datasets and was sufficient for evaluation of the MTR. Although differences in species diversity were recorded in the Conservation Rivers and Agency datasets, it was considered that those resulting from methodological differences would not significantly affect either the MTR scores or the MTR evaluation.

In general, the data gathering strategy was found to be justified, the various datasets complementing each other and balancing out inherent biases due to different site selection criteria. This provided a suitable geographical distribution and range of river characteristics upon which to analyse the performance of the MTR. The incompleteness of some of the datasets, with reference to physical and chemical characteristics, will, however, bias the applicability of the analysis towards those types of sites for which a full complement of data are available, ie the UWWTD sites which were mainly in England. As phosphate and nitrate data were only available for 116 of the 129 species, analyses of data for several of the rarer species was not possible. Results for data analysis must therefore be treated with a degree of caution. For this reason, data analyses are presented in Chapter 3 for either subsets of compatible data within the database, and/or the full database, as appropriate.

3 MTR AND NUTRIENT STATUS

3.1 Introduction

The hypothesis under test was 'that MTR expresses the trophic status of a river in terms of the response of the macrophyte community to nutrient status'. This hypothesis is based upon the premise that within the aquatic macrophyte flora there is a spectrum of tolerances to nutrient enrichment which can be represented on a scale from one to ten - the higher the score (STR), the lower the tolerance to nutrient enrichment. The response of the assemblage of plant species (or 'community') at a site to the prevailing nutrient status can be assessed by integrating the STRs of the individual species present as a mean value (MTR), weighted according to the relative abundance of the individual species.

To test this premise, the relationship between nutrient status and (i) the distribution of individual species, (ii) STRs and (iii) MTR, was explored. In addition, the use of species diversity and overall percentage macrophyte cover in the assessment of trophic status was considered.

The nutrients considered were compounds of phosphorus and nitrogen. The growth of freshwater plants are generally considered to be more limited by phosphorus than nitrogen in most cases (eg Sculthorpe 1967, Kern-Hansen & Dawson 1978), although both may be artificially high as a result of wastewater discharges. The evaluation, therefore, focused mainly on the relationship between MTR, and its components, and phosphorus concentration (as phosphate-phosphorus). In some situations, however, nitrogen may be limiting (eg Kelly & Whitton 1998), and hence some analyses were also undertaken of the relationship with nitrate concentration.

3.2 Species distribution and phosphate

The distributions of individual species were initially examined graphically against the logarithm of the phosphate concentration, in terms of (i) the percentage occurrence in bands of phosphate concentration and (ii) the mean cover when present (Figures 12a-f). The percentage occurrence gives an indication of the chance that a particular species will occur at a given phosphate concentration. The logarithmic conversion of phosphate was chosen as it produced a satisfactory normalisation of the data. The analysis was limited to the more abundant species (species recorded at more than 10% of sites), as the distributions for the less frequently occurring species could not be determined with confidence. This was mainly due to the low number of records for high-scoring species for which phosphate data were available.

The results of this investigation indicated that many of the more abundant species are fairly 'cosmopolitan', occurring over a wide range of phosphate concentration. Within this minimum–maximum tolerance range, however, individual species' preferences were apparent. For example, some species, although fairly cosmopolitan, were found either rarely or only at low abundance at low phosphate concentrations, and were found more frequently or at greater abundance as the logarithm of phosphate concentration increased. *Cladophora* agg., *Enteromorpha* spp., *Potamogeton pectinatus*, *Sparganium erectum*, *Apium nodiflorum* and *Lemna minor* fall into this category. Some of these species, such as *Cladophora*, show marked increases in cover with increasing phosphate concentration, this being a symptom of

eutrophication as defined by the UWWTD and the Agency's Eutrophication Strategy. In contrast, other species were found either rarely or only at low abundance at high phosphate concentrations, and were found more frequently or at greater abundance at low phosphate concentrations. Examples of the latter include *Fontinalis antipyretica* and *Rhyncostegium riparioides*, although phosphate concentration does not explain all the variability in their distribution. Species such as *Vaucheria* spp. displayed no particular preference in relation to phosphate concentration.

There was no relationship between phosphate concentration and the percentage occurrence or cover of an individual 'common' species, which was strong enough for the species to be used alone as an indicator of trophic status.

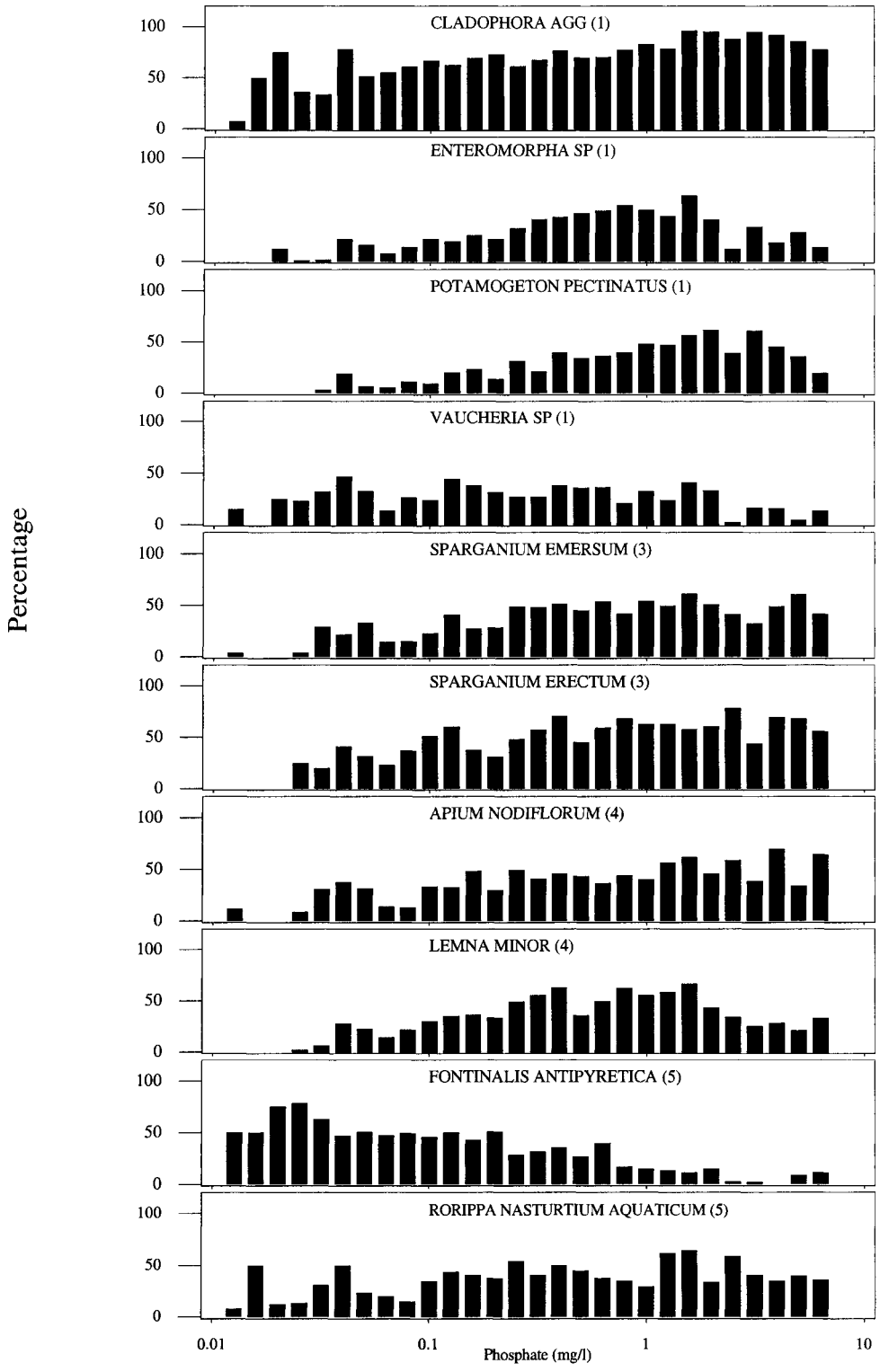


Figure 12a. Percentage occurrence in each phosphate band against the logarithm of soluble phosphate concentration, for surveys where phosphate data were available I: species recorded at more than 25% (575) of surveys. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets for 1985-97. Macrophyte data matched with phosphate data either from the same site or from a site within 1 km. Species names are capitalised to indicate that they are 'highlighted' species. STR values are given in brackets and data is presented in ascending STR order.

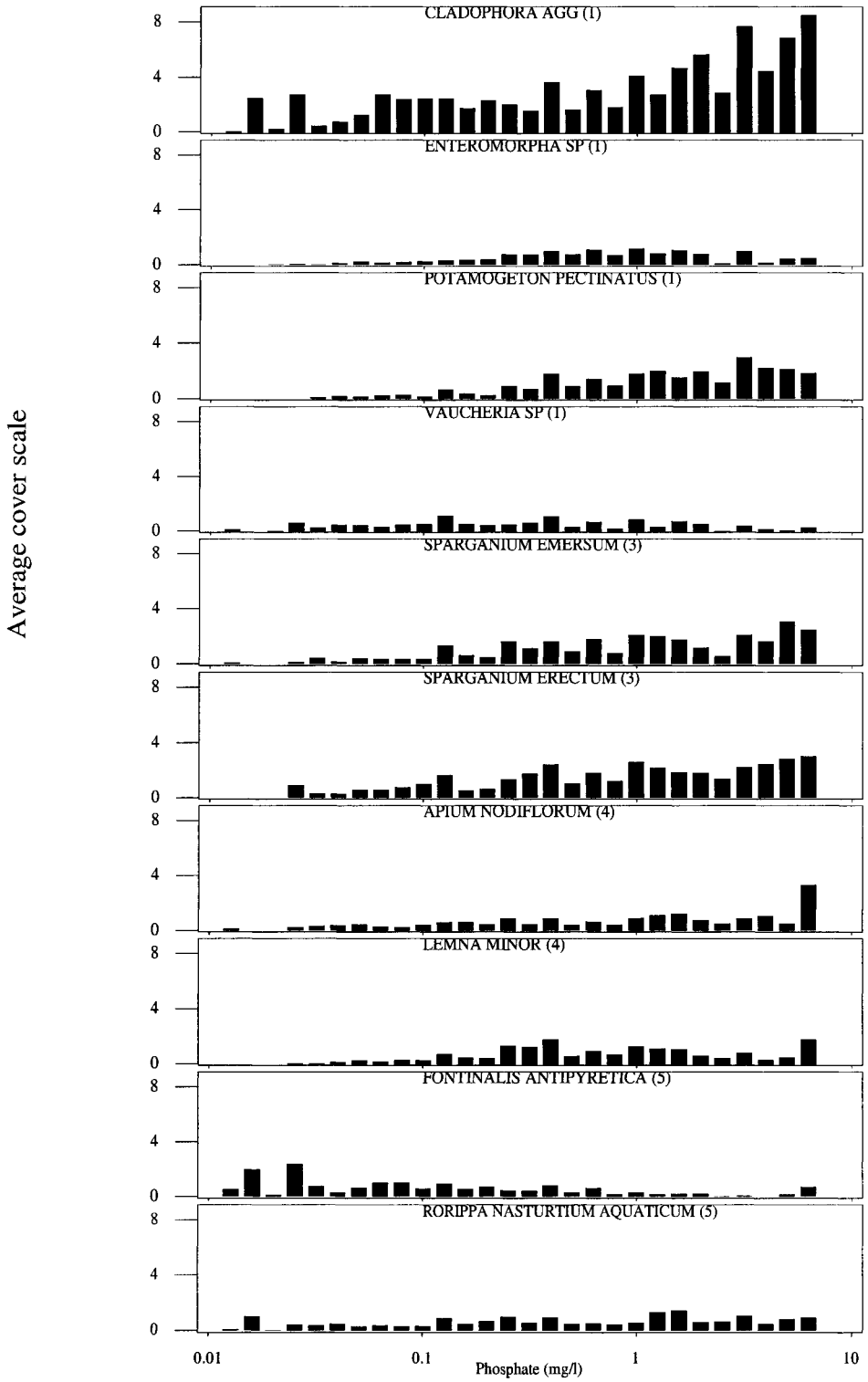


Figure 12b. Mean species cover against the logarithm of soluble phosphate concentration, for surveys where phosphate data were available. I: species recorded at more than 25% (575) of surveys. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets for 1985-97. Macrophyte data matched with phosphate data either from the same site or from a site within 1 km. Species names are capitalised to indicate that they are 'highlighted' species. STR values are given in brackets and data is presented in ascending STR order. Cover was recorded on a 9-point scale (see Appendix 1); cover ratings of zero were not included in the calculation of mean cover.

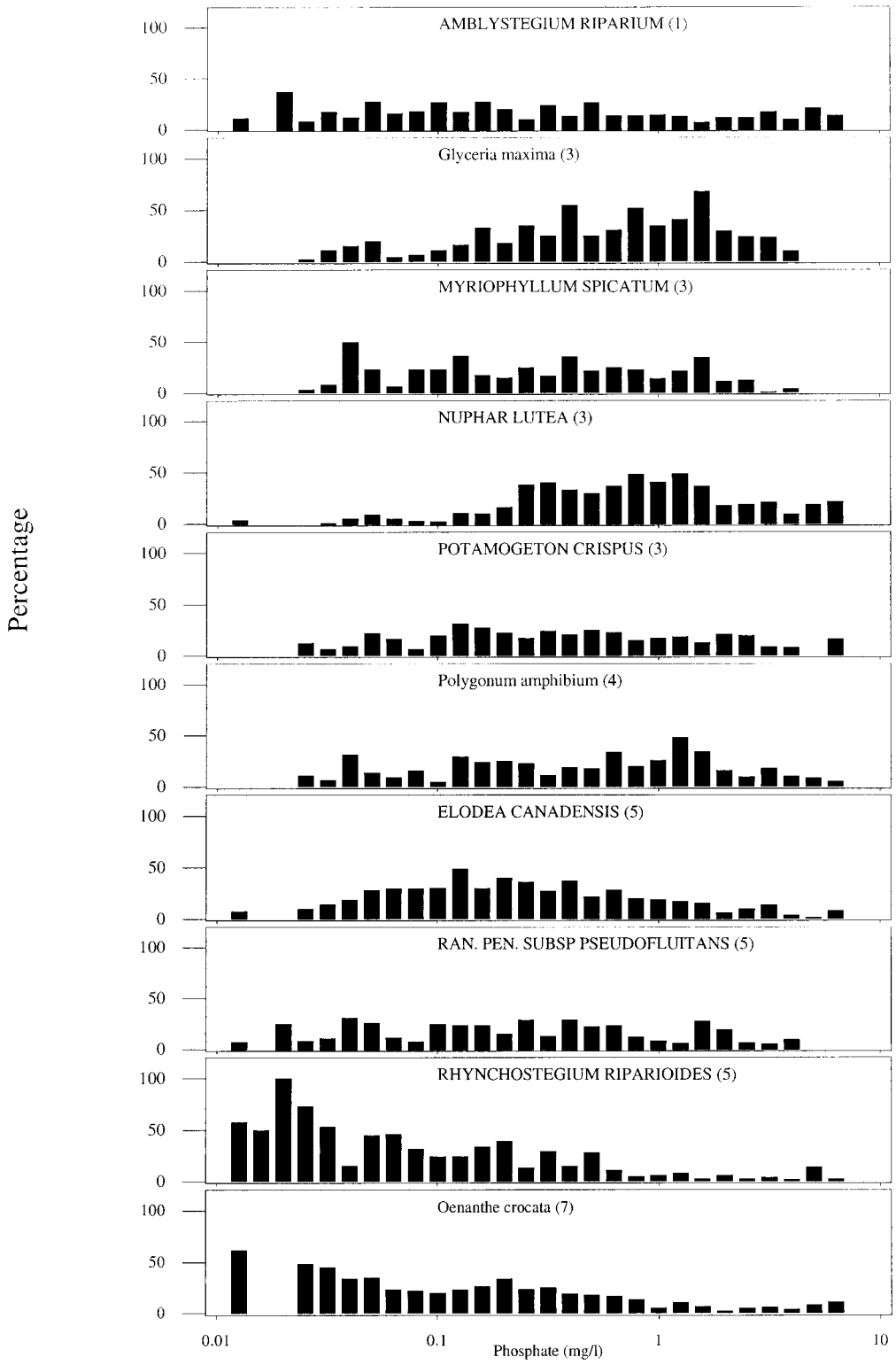


Figure 12c. Percentage occurrence in each phosphate band against the logarithm of soluble phosphate concentration, for surveys where phosphate data were available. II: species recorded at 15–25% (350–575) of surveys. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets for 1985-97. Macrophyte data matched with phosphate data either from the same site or from a site within 1 km. Species names are capitalised to indicate that they are 'highlighted' species. STR values are given in brackets and data is presented in ascending STR order.

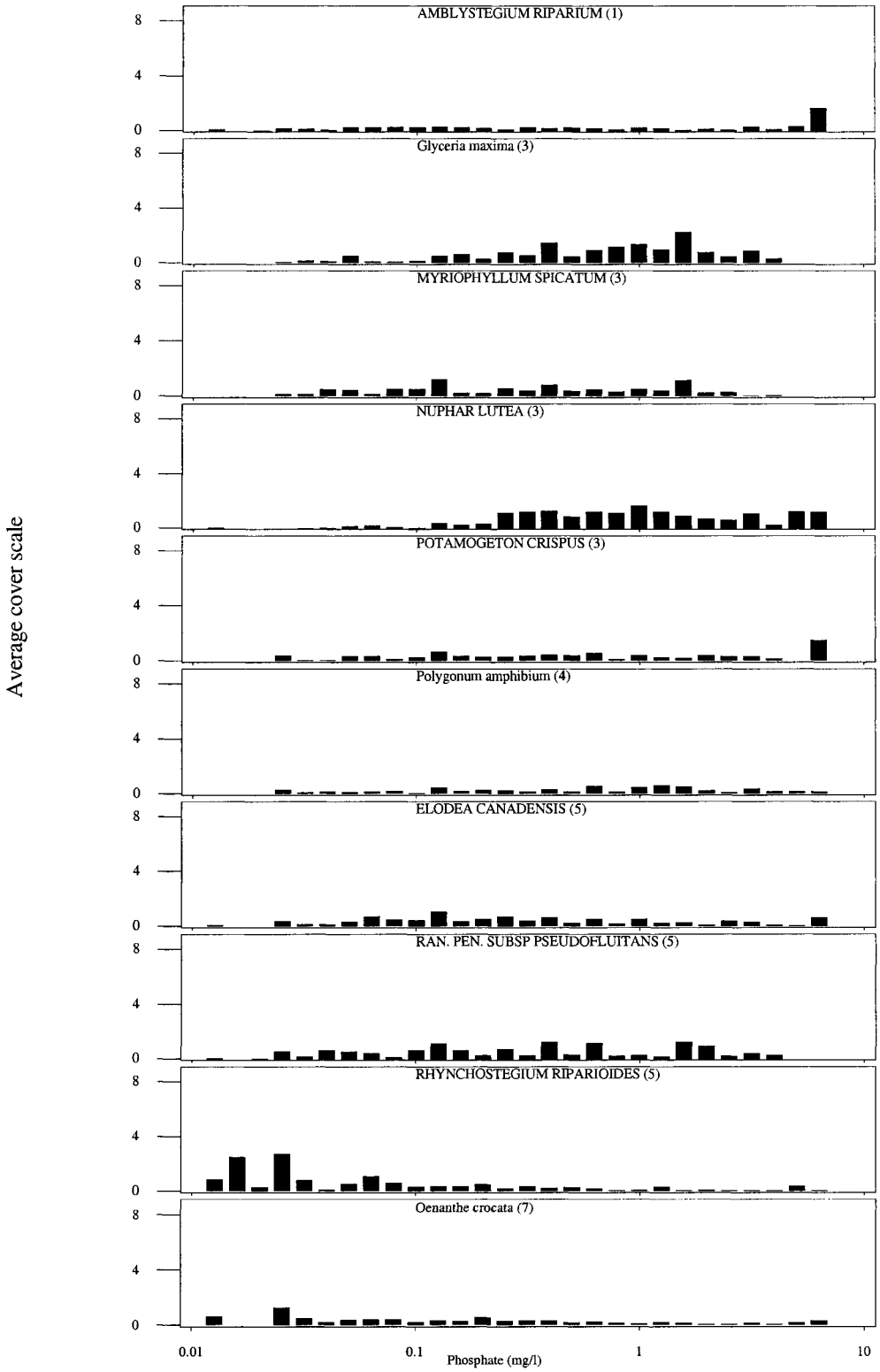


Figure 12d. Mean species cover against the logarithm of soluble phosphate concentration, for surveys where phosphate data were available. II: species recorded at 15–25% (350–575) of surveys. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets for 1985-97. Macrophyte data matched with phosphate data either from the same site or from a site within 1 km. Names of highlighted' species are capitalised. STR values are given in brackets and data is presented in ascending STR order. Cover was recorded on a 9-point scale (see Appendix 1); cover ratings of zero were not included in the calculation of mean cover.

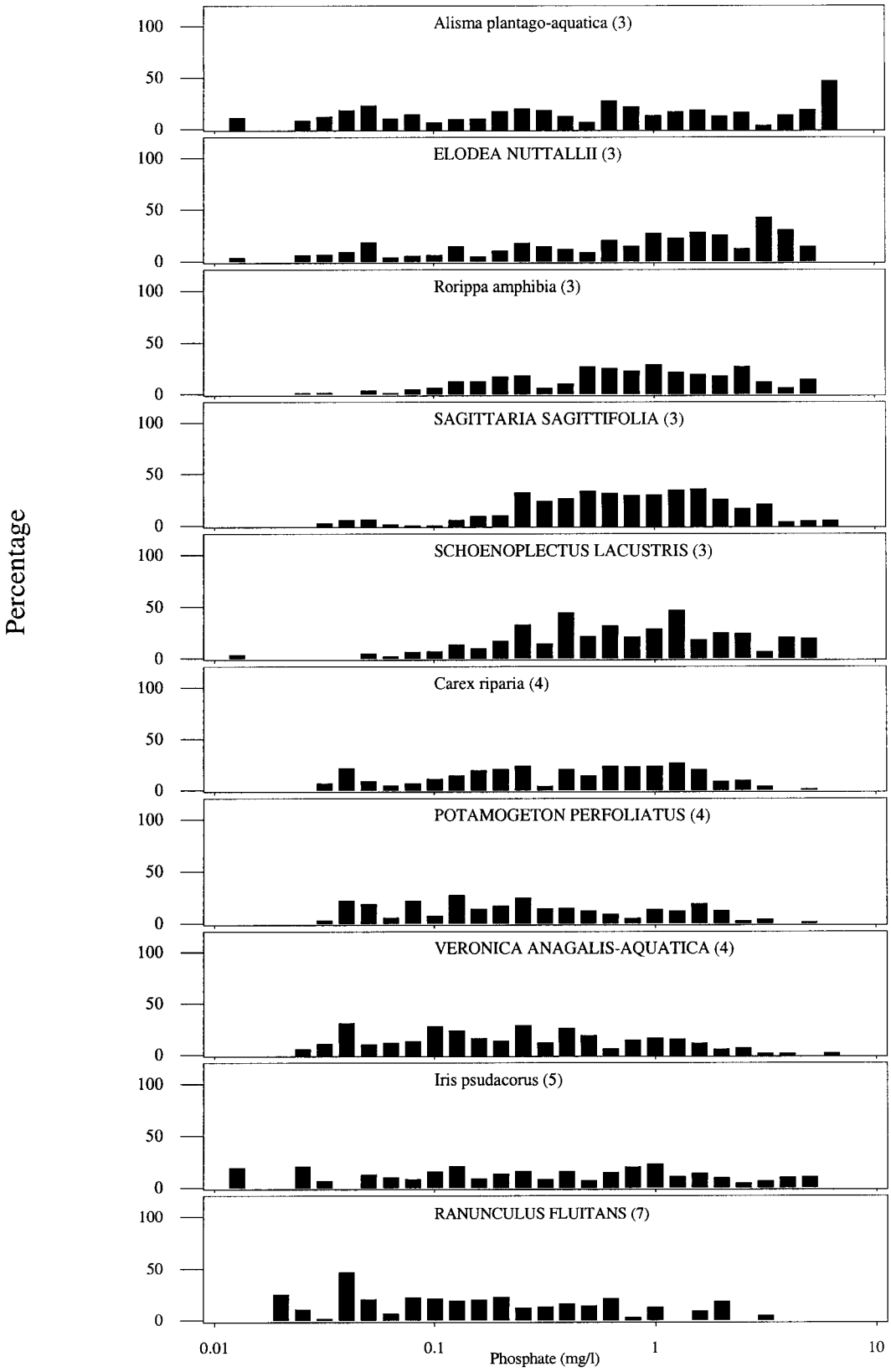


Figure 12e. Percentage occurrence in each phosphate band against the logarithm of soluble phosphate concentration for surveys where phosphate data were available. III: species recorded at 10–15% (250–350) of surveys. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets for 1985-97. Macrophyte data matched with phosphate data either from the same site or from a site within 1 km. Species names are capitalised to indicate that they are 'highlighted' species. STR values are given in brackets and data is presented in ascending STR order.

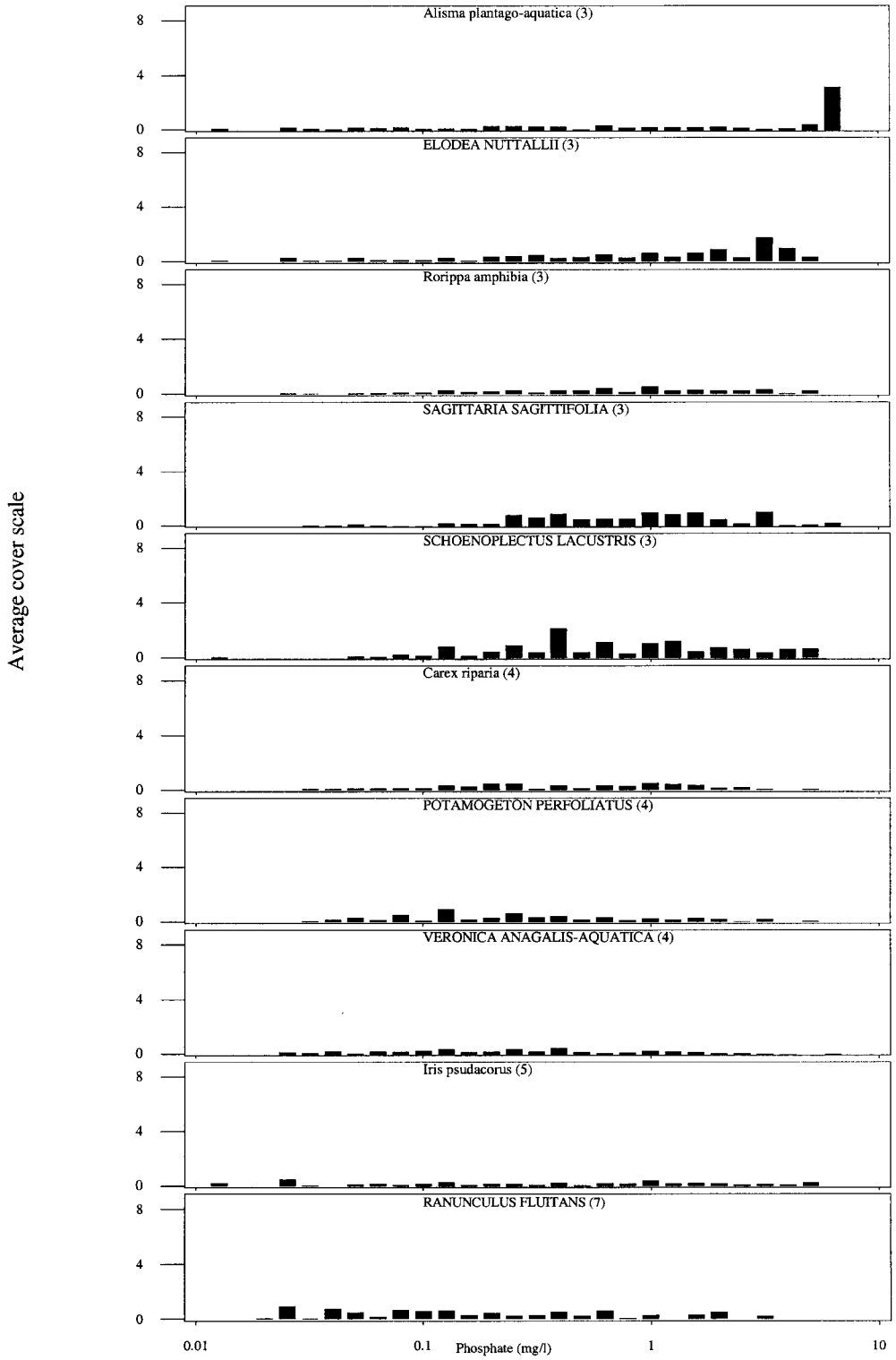


Figure 12f. Mean species cover against the logarithm of soluble phosphate concentration, for surveys where phosphate data were available. III: species recorded at 10–15% (250–350) of surveys. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets for 1985-97. Macrophyte data matched with phosphate data either from the same site or from a site within 1 km. Names of highlighted' species are capitalised. STR values are given in brackets and data is presented in ascending STR order. Cover was recorded on a 9-point scale (see Appendix 1); cover ratings of zero were not included in the calculation of mean cover.

3.3 Species distribution and nitrate

The distribution of individual macrophyte species in relation to nitrate concentration was examined graphically in a similar manner to that for phosphate, but in less detail. The analysis was restricted to the mean cover of the commoner species against the logarithm of the mean nitrate concentration (Figure 13). This analysis showed a generally similar pattern to that shown for phosphate. The abundance of several low STR species significantly increased as the logarithm of nitrate concentration increased, over a range of 0.1–25 mg l^{-1}. The cover of some of these species, such as *Lemna minor* and possibly *Enteromorpha*, appear to increase more with nitrate concentration than with phosphate, whereas the cover of species such as *Cladophora* and *Potamogeton pectinatus* increase with both nitrate and phosphate. The abundance of *Fontinalis antipyretica* and *Rhyncostegium riparioides* (data for the latter not shown) decreased with nitrate concentration, as with phosphate concentration (cp Figures 12 &13).

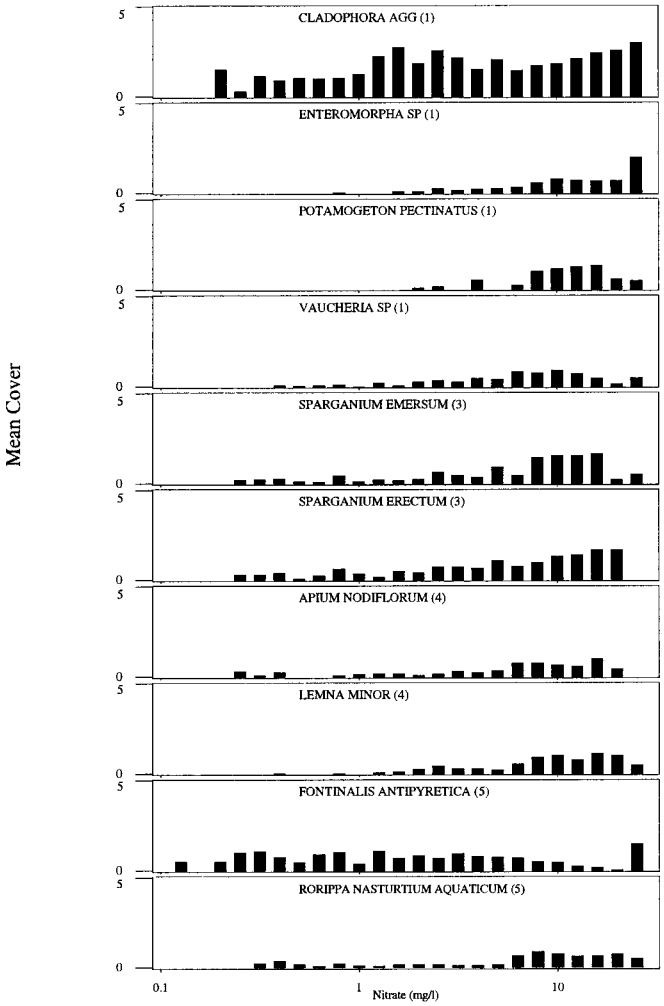


Figure 13. Mean cover of commonly recorded species against the logarithm of nitrate concentration, for surveys where nitrate data were available. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets for 1985-97. Macrophyte data matched with nitrate data either from the same site or from a site within 1 km. 'Commonly recorded' species are those recorded at more than 25% (575) of sites. Species names are capitalised to indicate that they are 'highlighted' species. STR values are given in brackets and data is presented in ascending STR order. Cover was recorded on a 9-point scale (see Appendix 1); cover ratings of zero were not included in the calculation of mean cover.

3.4 STR and phosphate

3.4.1 STR - phosphate relationship

The species' phosphate preferences analysed in terms of STR scores (Figures 12a–f), support the STRs assigned to species in relation to phosphate concentration, although there is a moderate degree of variability. Further analysis of this pattern of STRs against phosphate concentration was undertaken by condensing the distribution data into summary statistics for each species. Data transformations were tested and the logarithmic conversion of the phosphate concentration was found to give an acceptable normalisation of the data. Species were ordered by STR group and data expressed as the mean phosphate concentration at which individual species occurred, together with the standard deviation and range (Figures 14a-b). The overall pattern confirms that although the correct STR was assigned to many individual species, there is considerable overlap between species, even within the relatively narrow range of ± 1 standard deviation. This is a departure from the ideal model introduced in 2.2.1 but is not unexpected. The obvious outliers are those species with few occurrences, which have lower confidence attached to their summary statistics.

The wide variation in growth-phosphate statistics for high-scoring species is reduced if data for Conservation Rivers pre-1986 are added and data for all Conservation Rivers matched with phosphate concentrations from GQA sites within 5 km (Figures 15a-b). The net effect of this reduction in variation is to strengthen the overall relationship between STR and phosphate concentration. Although the reduction in variation could be a result of greater accuracy or consistency of identification of some high-scoring species within the larger dataset (most of the additional data for pre-1986 Conservation Rivers were collected by one surveyor), it could equally reflect a reduction in the bias of the dataset towards larger lowland rivers, the wider range of physical/chemical river types in the larger dataset giving a more accurate picture of the species distribution. Confidence in the relationship demonstrated by the larger dataset rests on whether it is valid to use non-contemporaneous phosphate and macrophyte data.

The relationship between STR and mean phosphate (Figures 14 and 15), is confirmed when the logarithm of the mean phosphate concentration at which individual species occurred is plotted against the STR of the species (Figures 16 & 17). On the graph, each data point represents one species. STR broadly reflects the mean phosphate concentration, but with a range of responses apparent within each STR group, and some overlap between adjacent groups. The relationship between STR and mean phosphate concentration is improved when the entire database is used (Figure 17), compared to when Conservation Rivers surveys from pre-1986 are excluded (Figure 16). The possible reasons for this difference, and the assumption necessary regarding the validity of using non-contemporaneous phosphate and macrophyte data, are outlined above (Figures 14 and 15). No change in the overall pattern of the relationship was found when species were grouped according to growth habit (such as submerged, free-floating, emergent etc), or their water acidity mid-point, and the data re-plotted for the different groups (results not shown).

There are several 'outliers' apparent in the relationship between STR and mean phosphate concentration (Figures 16 and 17). Two of these, *Stigeoclonium tenue* and *Hydrocharis morsus-ranae*, were recorded on fewer than 10 occasions and thus confidence in the relationship with mean phosphate concentration is low. For the other species either the STRs could be incorrect or the specimens could have been misidentified. As no herbarium samples

were available, however, it was not possible to verify the identification.

The 'outliers' may also be identified by listing those low-scoring species (STR < 4) recorded at sites with an MTR greater than 65; and those high-scoring species (STR > 4) recorded at sites with an MTR less than 35 (assuming MTR increases with decreasing phosphate concentration; Table 3). Examples of such 'outlier' species are *Callitriche hamulata*, *Myriophyllum alterniflorum* and *Juncus bulbosus*, for which the STR could be too high, and *Vaucheria* spp. and *Brachythecium rutabulum* for which the MTR could be too low. However, there is insufficient evidence for changing the STR for any of these species. All could be subject to mis-identification, *B. rutabulum* is rarely in water, and *Vaucheria* spp. are commonly recorded algae with a wide distribution and unlikely to be found at high abundance at a low phosphate concentration (Figure 12).

Table 3. List of a) species with an STR less than 4, occurring at sites with an MTR greater than 65 and b) species with an STR greater than 4, occurring at sites with an MTR less than 35, with numbers and percentages of total occurrences of each species.

a) MTR > 65			b) MTR < 35		
Species (STR)	**Occurrences**		**Species (STR)**	**Occurrences**	
	n	**%**		**n**	**%**
Amblystegium riparium (1)	19	2	*Equisetum palustre* (5)*	2	0.4
Cladophora (1)	25	0.9	*Ranunculus* subs *pseudofluitans* (5)*	1	0.2
Enteromorpha sp. (1)	3	0.3	*Potamogeton natans* (5)*	1	0.2
Vaucheria sp. (1)	34	2.9	*Rhynchostegium riparoides* (5)*	1	0.1
			Rorippa nasturtium-aquaticum (5)*	10	0.7
Ranunculus sceleratus (2)	1	0.2			
			Myriophyllum alterniflorum (8)	3	0.6
Alisma plantago-aquatica (3)	15	2.2			
Brachythecium rutabulum (3)	68	11.1	*Callitriche hamulata* (9)	3	0.6
Glyceria maxima (3)	4	0.4	*Dichodontium pellucidium* (9)	4	1.4
Myriophyllum spicatum (3)	2	0.3	*Apium inundatum* (9)	1	3
Nuphar lutea (3)	11	1.3	*Hygrohypnum ochraceum* (9)	1	0.2
Rumex hydrolopathum (3)	3	1.1	*Menyanthes trifoliata* (9)	2	3.7
Sparganium emersum (3)	26	1.9	*Potentilla erecta* (9)	1	0.3
Sparganium erectum (3)	55	2.2			
			Blindia acuta (10)	2	1.9
			Hyocomium armoricum (10)	3	1.2
			Juncus bulbosus (10)	9	1.7

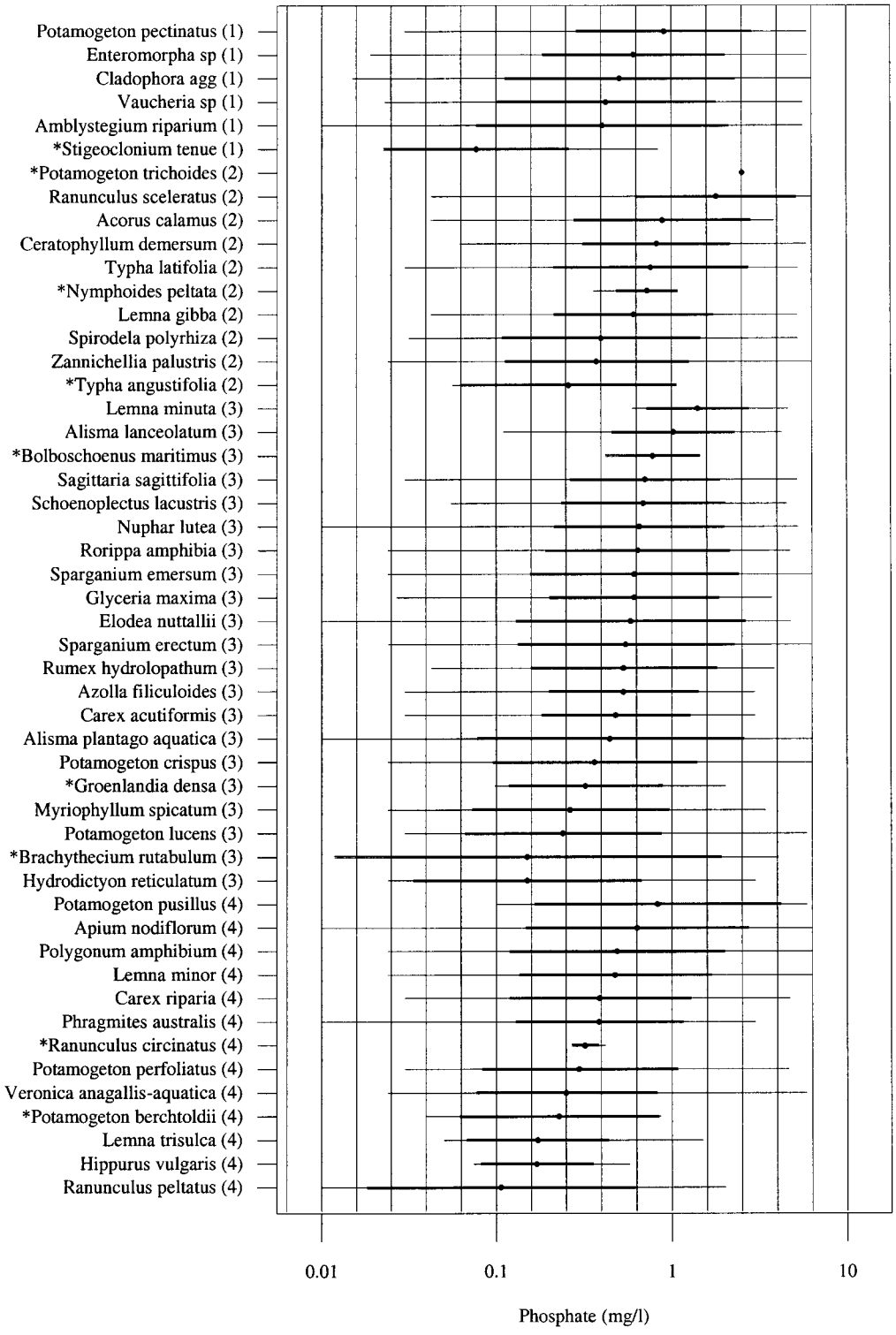
(* STR 5 species occurrences only tabulated for sites with MTR < 15)

In an attempt to confirm those species for which a change in STR may be justified, a new set of rankings was derived based on the data collected during this study. Species were ordered according to the mean annual phosphate concentration at which they occurred (Figure 18), then divided into ten equal groups and the species in each group assigned a 'phosphate rank'. Several species were assigned a different rank to the STR, some of these being significantly different and several candidates for a change in STR were found (Table 4 & Figure 19).

It is recommended that *Stigeoclonium tenue* is removed from the MTR checklist as it is considered that this species is more an indicator of non-nutrient pollution than of eutrophication. Other candidate species would require further detailed studies before any firm recommendation of changes to STR status could be made. These studies would need to provide contemporaneous MTR and phosphate data and to have increased numbers of records for these species. Future recalculation of MTR based upon 'phosphate ranks', and subsequent re-evaluation of the MTR-phosphate relationship, may be useful in confirming changes; this may also involve re-assigning phosphate ranks by weighting mean phosphate concentration according to the cover of the species.

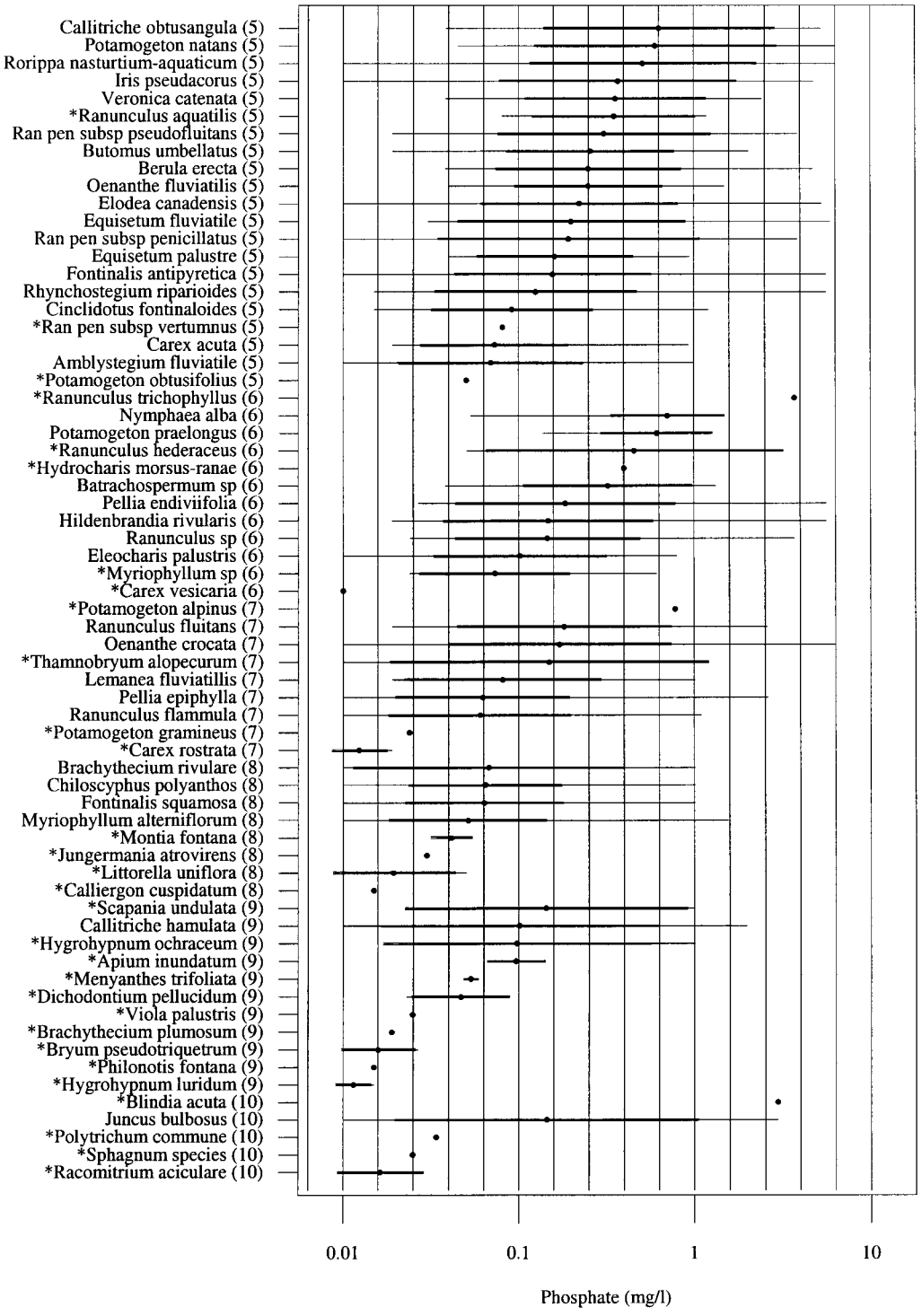
Table 4. List of species for which 'phosphate rank' differs significantly from STR and the relationship with nitrate. (>) indicates that the 'phosphate rank' is higher than the STR for the species; (<) indicates that the 'phosphate rank' is lower than the STR for the species.

Species	STR	'Phosphate ranking'	Species present at sites with low nitrate but high phosphate
Stigeoclonium tenue	1	> 6	
Amblystegium riparium	1	> 5	
Vaucheria	1	> 4	
Nymphoides peltata	2		*
Brachythecium rutabulum	3	> 6	
Groenlandia densa	3	> 5	
Potamogeton freisii	3	> 8	
Ranunculus peltata	4	> 7	
Ranunculus circinatus	4		*
Ranuculus pen. vertum.	5	> 8	*
Oenanthe fluviatilis	5		*
Veronica catenata	5		*
Berula erecta	5		*
Catabrosa aquatica	5		*
Hydrocharis morsus-ran.	6	<3	
Carex vesicaria	6	> 10	
Batrachospermum	6		*
Ranunculus hederaceus	6		*
Potamogeton praelongus	6	< 2	
Nymphaea alba	6	< 1	
Ranunculus trichophyllus	6	< 3	
Potamogeton alpinus	7		*
Menyanthes trifoliata	9		*
Juncus bulbosus	10		*
Blindia acuta	10	< 6	



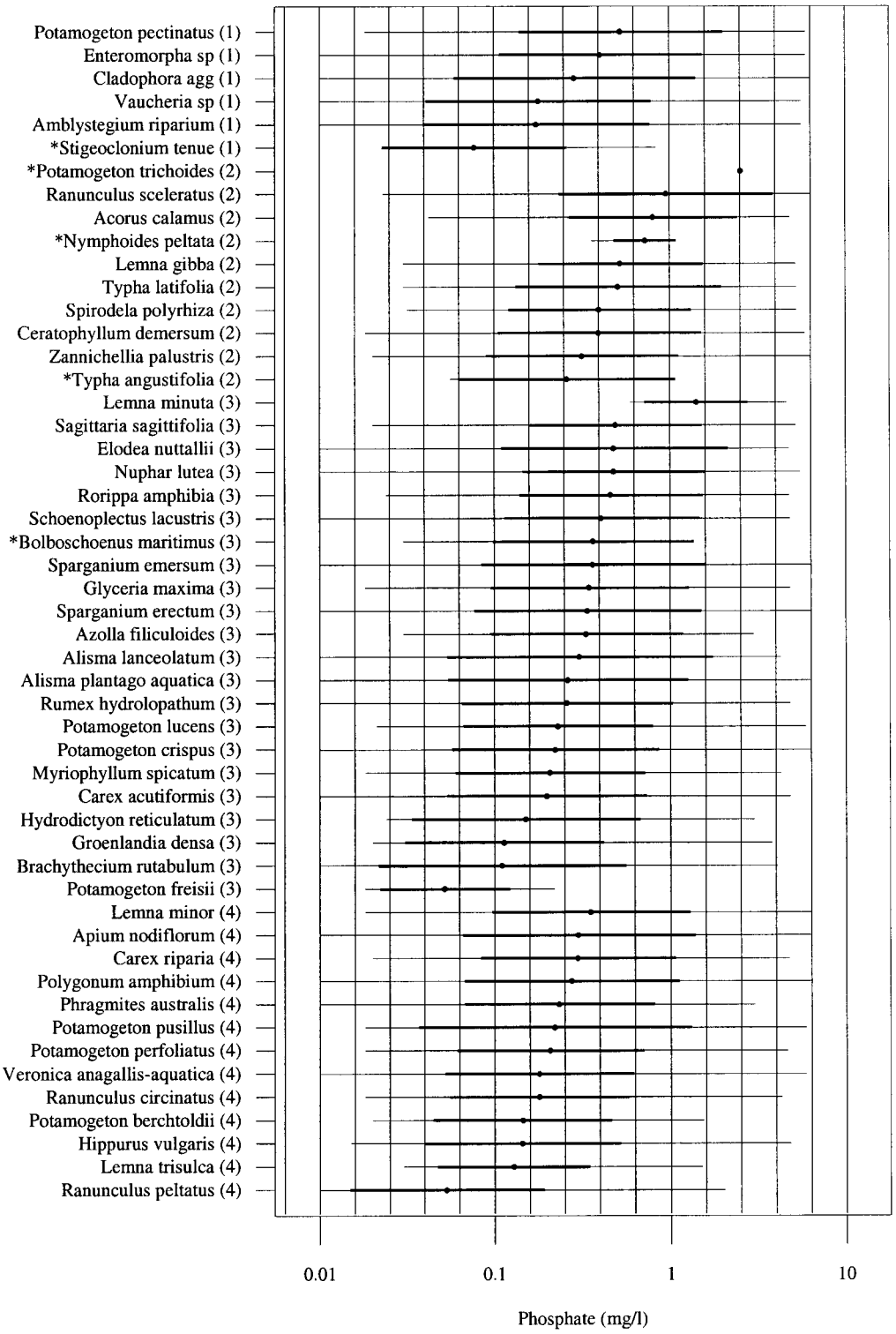
* Species with fewer than ten occurrences

Figure 14a. Species occurrence against the logarithm of the phosphate concentration within 1 km (summary data). I: low-scoring species. Mean phosphate concentration given as a dot, with a thick line giving plus/minus one standard deviation (SD) and a thin line giving minimum/maximum. Where no SD or minimum/maximum is shown, this indicates that one or more records for the species, but only a single phosphate datum. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets (Conservation Rivers data for post-1985 surveys only). Macrophyte data matched with phosphate data either from the same site or from a site within 1 km. STR values given in brackets after the species name.



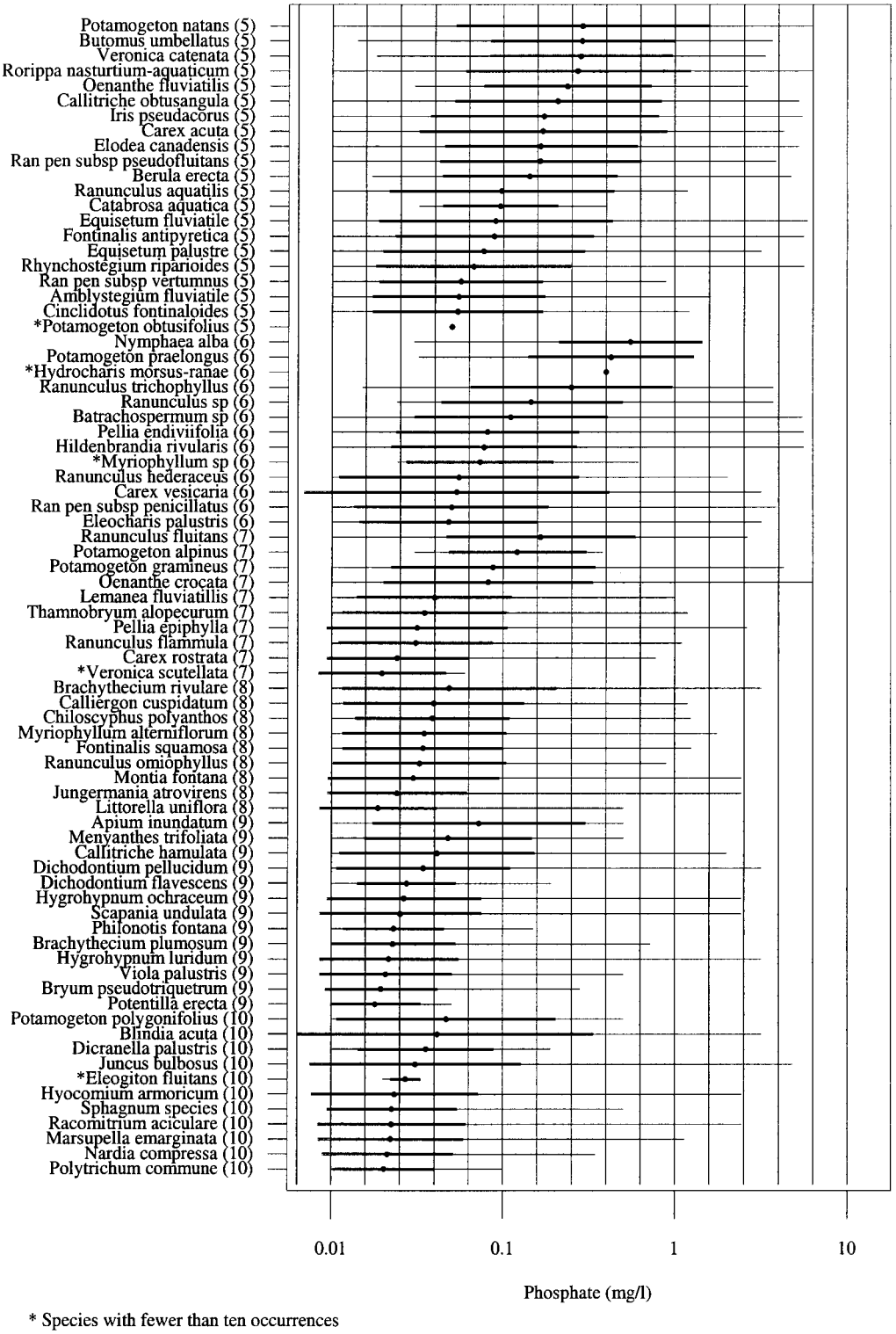
* Species with fewer than ten occurrences

Figure 14b. Species occurrence against the logarithm of the phosphate concentration within 1 km (summary data). II: high-scoring species. Mean phosphate concentration given as a dot, with a thick line giving plus/minus one standard deviation (SD) and a thin line giving minimum/maximum. Where no SD or minimum/maximum is shown, this indicates that one or more records for the species, but only a single phosphate datum. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets (Conservation Rivers data for post-1985 surveys only). Macrophyte data matched with phosphate data either from the same site or from a site within 1 km. STR values given in brackets after the species name.



* Species with fewer than ten occurrences

Figure 15a. Species occurrence against the logarithm of the phosphate concentration within 5 km (summary data). I: low-scoring species. Mean phosphate concentration given as a dot, with a thick line giving plus/minus one standard deviation (SD) and a thin line giving minimum/maximum. Where no SD or minimum/maximum is shown, this indicates that one or more records for the species, but only a single phosphate datum. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets (all years). Macrophyte data matched with phosphate data from either the same site or from a site within 5 km. STR values given in brackets after the species name.



* Species with fewer than ten occurrences

Figure 15b. Species occurrence against the logarithm of the phosphate concentration within 5 km (summary data). II: high-scoring species. Mean phosphate concentration given as a dot, with a thick line giving plus/minus one standard deviation (SD) and a thin line giving minimum/maximum. Where no SD or minimum/maximum is shown, this indicates that one or more records for the species, but only a single phosphate datum. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets (all years). Macrophyte data matched with phosphate data either from the same site or from a site within 5 km. STR values given in brackets after the species name.

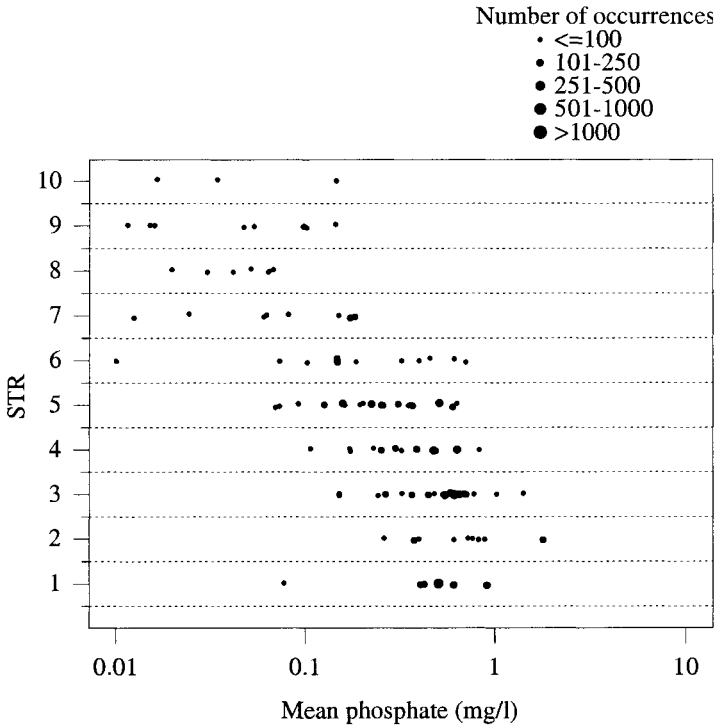


Figure 16. Relationship between STR and the logarithm of the mean phosphate concentration recorded for individual species. Data from EA. IFE, DoE/IRTU and Conservation Rivers datasets (Conservation Rivers data for post-1985 surveys only). Macrophyte data matched with phosphate data either from the same site or from a site within 1 km. A small random movement has been added to the Y-axis position to separate points and hence to improve clarity.

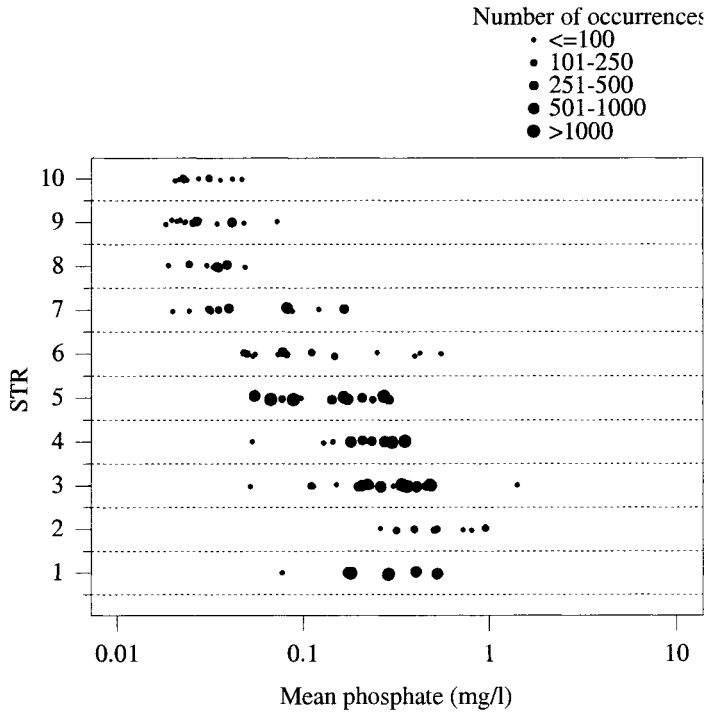


Figure 17. STR against the logarithm of the mean phosphate concentration recorded for individual species. Data from EA. IFE, DoE/IRTU and Conservation Rivers datasets (all years). Macrophyte data matched with phosphate data either from the same site or from a site within 5 km. A small random movement has been added to the Y-axis position to separate points and hence to improve clarity.

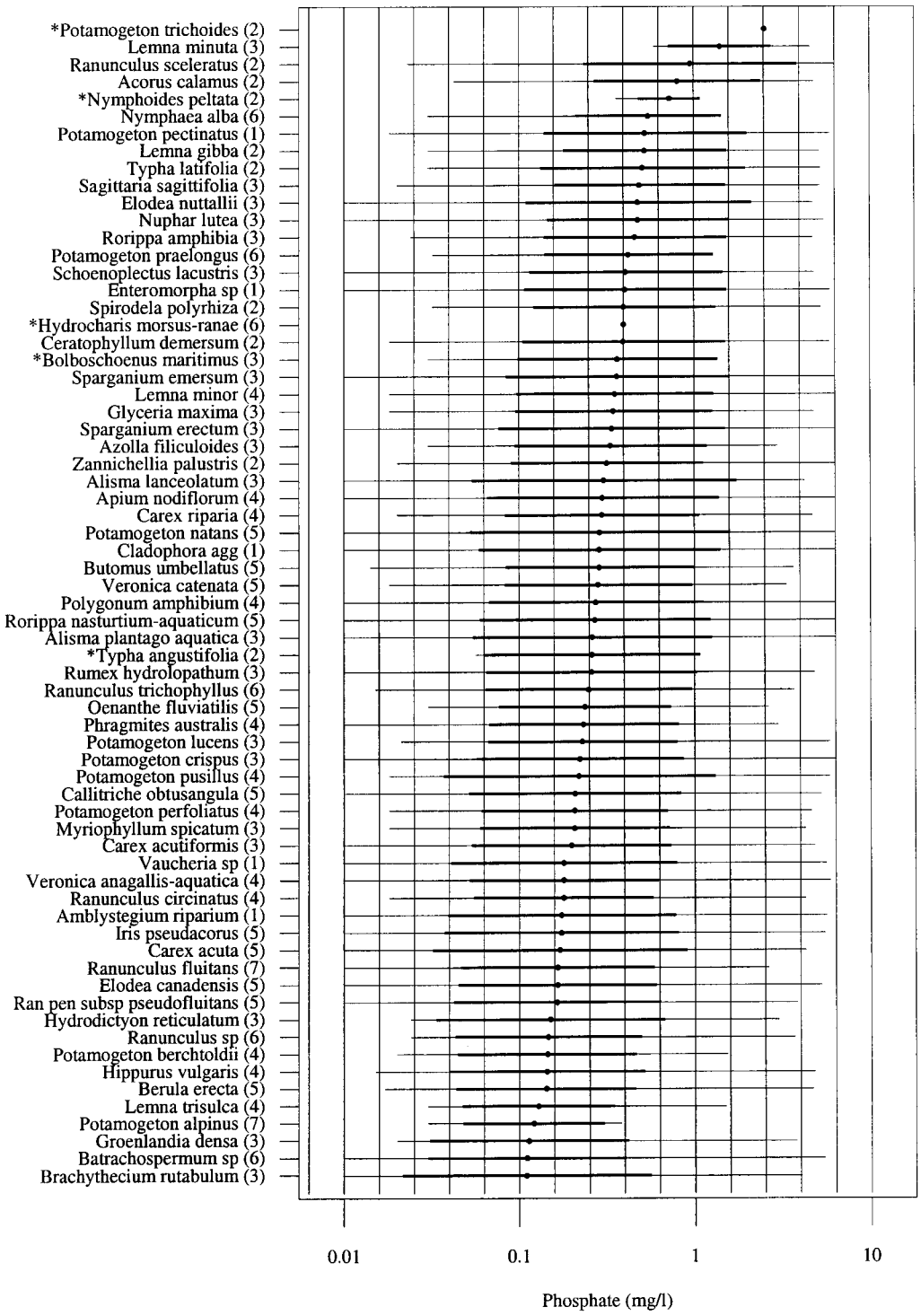
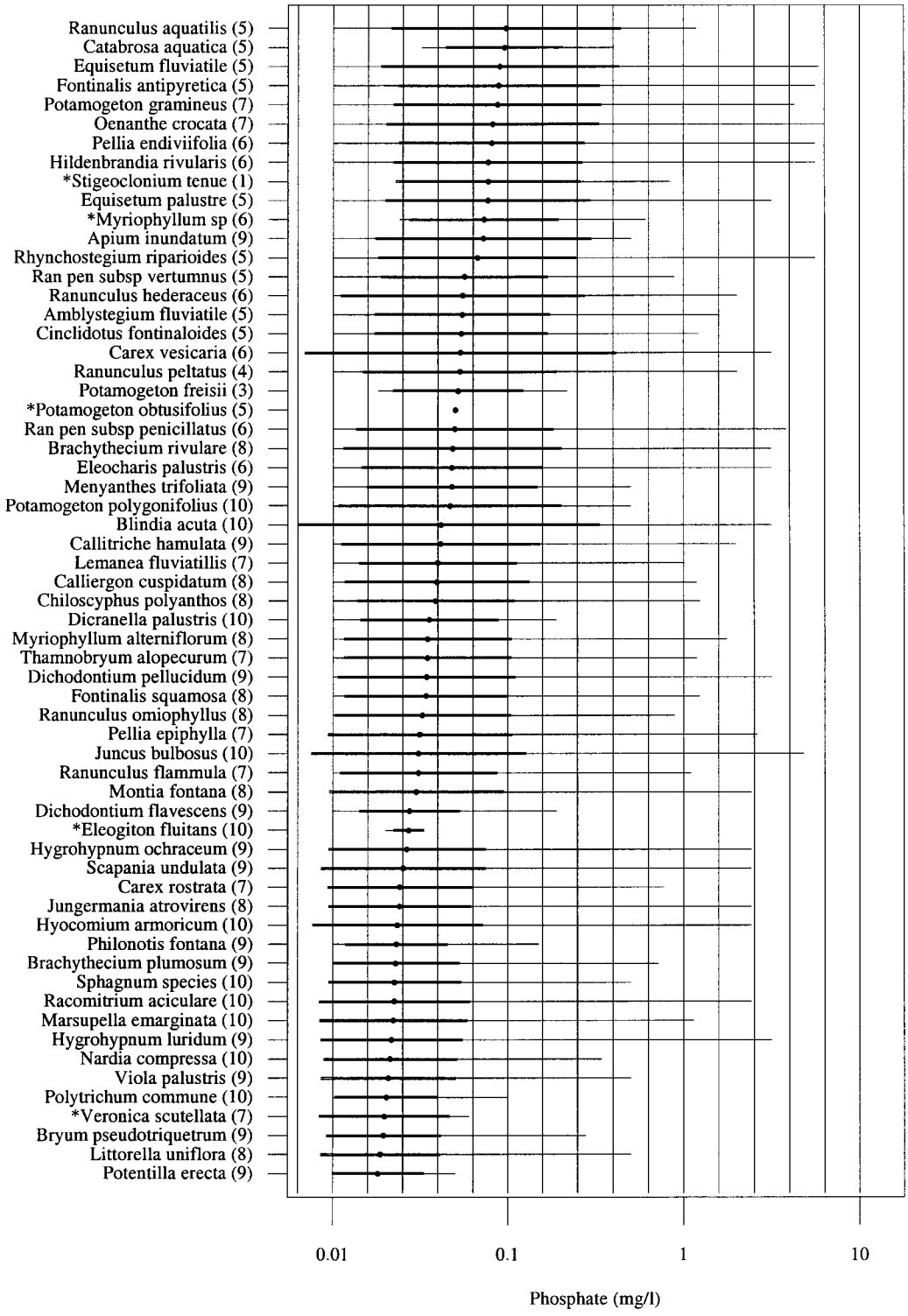


Figure 18a. Species occurrence ordered by phosphate-mean against the logarithm of the phosphate concentration within 5 km (summary data). I: low-ranking species. Mean phosphate concentration given as a dot, with a thick line giving plus/minus one standard deviation (SD) and a thin line giving minimum/maximum. Where no SD or minimum/maximum is shown, this indicates that one or more records for the species, but only a single phosphate datum. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets (all years). Macrophyte data matched with phosphate data from either the same site or from a site within 5 km. STR values given in brackets after the species name.



* Species with fewer than ten occurrences

Figure 18b. Species occurrence ordered by phosphate-mean against the logarithm of the phosphate concentration within 5 km (summary data). II: high-scoring species. Mean phosphate concentration given as a dot, with a thick line giving plus/minus one standard deviation (SD) and a thin line giving minimum/maximum. Where no SD or minimum/ maximum is shown, this indicates that one or more records for the species, but only a single phosphate datum. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets (all years). Macrophyte data matched with phosphate data either from the same site or from a site within 5 km. STR values given in brackets after the species name.

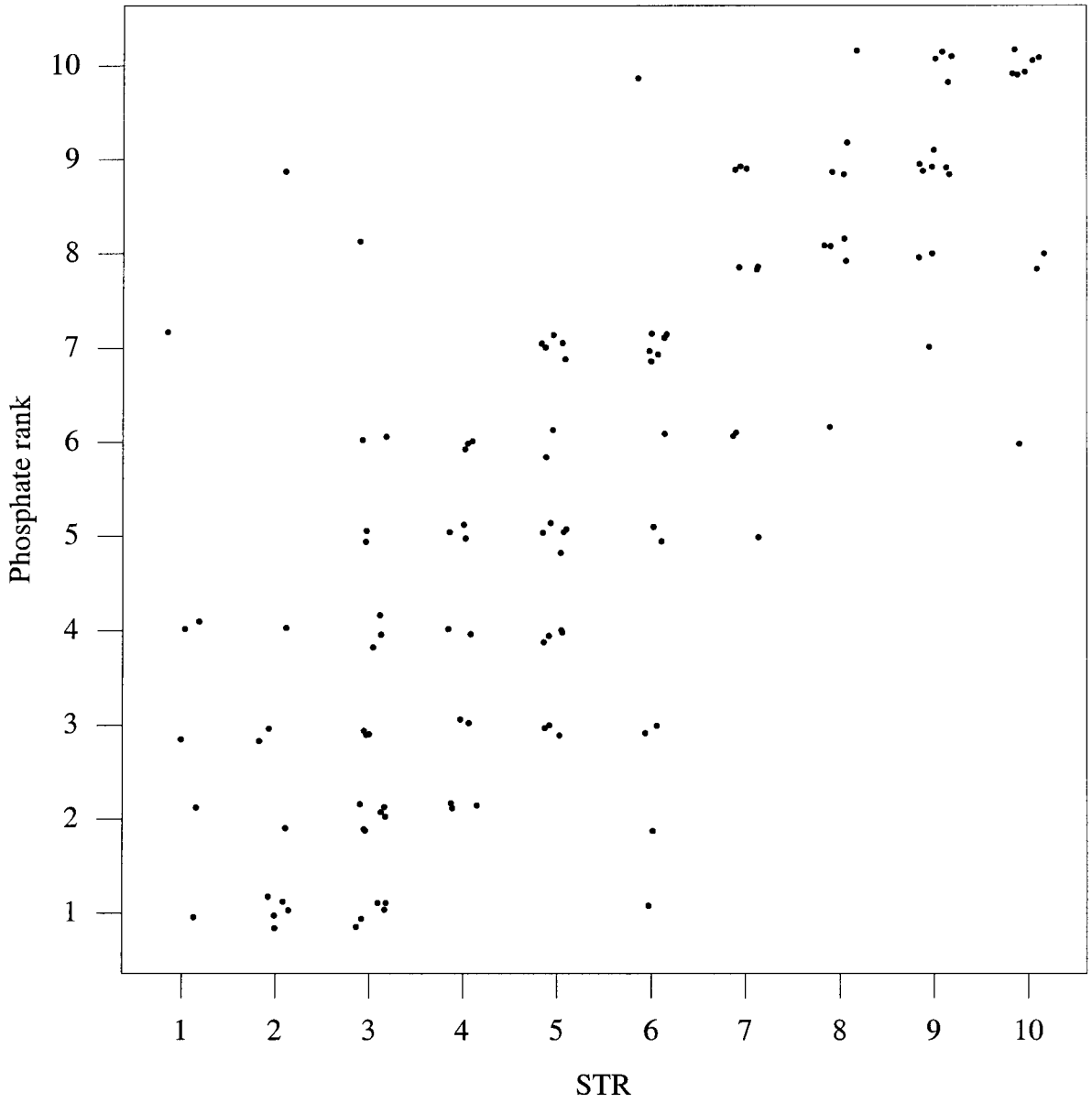


Figure 19. Relationship between STR and the 'phosphate rank'. Phosphate rank derived from EA, IFE, DoE/IRTU and Conservation Rivers datasets (Conservation Rivers data for post-1985 surveys only). Macrophyte data matched with phosphate data either from the same site or from a site within 1 km. Species then ranked in order of mean phosphate concentration at which they occur, and divided into ten scoring groups, each group comprising the same number of species. Outliers are listed in Table 4. A small random movement has been added to the X- and Y-axis positions to separate points and hence to improve clarity.

3.4.2 STRs present at different phosphate concentrations

The frequency of occurrence of the various STR groups, analysed over a range of phosphate concentrations (Figures 20a-c), confirms the findings outlined above and provides some indication of the structure of macrophyte communities at different trophic states.

Unlike the 'ideal' conceptual model introduced in 2.2.1, a moderate range of STRs occurs at all concentrations. There are noticeable differences, however, between the shape of the STR distribution when comparing the two extremes of the phosphate range. At very low concentrations (for example, < 0.03 mg l^{-1}) the distribution is skewed markedly to the right, with a high frequency of STR 7-10 species and with STR 1-4 species present but at lower frequency. In contrast, at high concentrations (for example, > 1.0 mg l^{-1}), the distribution is skewed markedly to the left, with STR 7-10 species very unlikely to occur and STR 1-5 species being frequent. In between these two extremes, there is a gradual transition in the general shape of the distribution. This transition is obscured to some degree, however, by the frequent occurrence of STR 3 and/or STR 5 species, and the relatively infrequency of STR 2 species, across the phosphate range.

The relative frequency of STR 3 and 5 species in the rivers surveyed (cp Figure 22) is mirrored in the list of MTR-scoring taxa (the MTR checklist). Unlike the 'ideal' conceptual model, there is a bias in the checklist towards STR 3 and 5, both when all scoring species are considered and when only highlighted species are considered (Figure 21). Although this has the potential to impart a corresponding bias of the resulting MTR towards scores of 30-50, this is difficult to test. The checklist was devised to include species tolerant of a range of physical and chemical conditions in each STR and may reflect the natural distribution of species. Species of STR 3 and 5 are the most commonly recorded and although they are present in most bands of MTR, STR 3 is rare above 65 and STR 5 rare below 15 (Figure 22). It is considered, however, that any effect of a bias in the MTR checklist on the MTR recorded at a site, is likely to be small in magnitude compared to the influence of other factors influencing the MTR. This applies also to the bias in the proportion of highlighted to non-highlighted species in each STR group, which is broadly similar across STRs except for the STR 7 group which contains an abnormally high proportion of non-highlighted species and the fewest number of highlighted species.

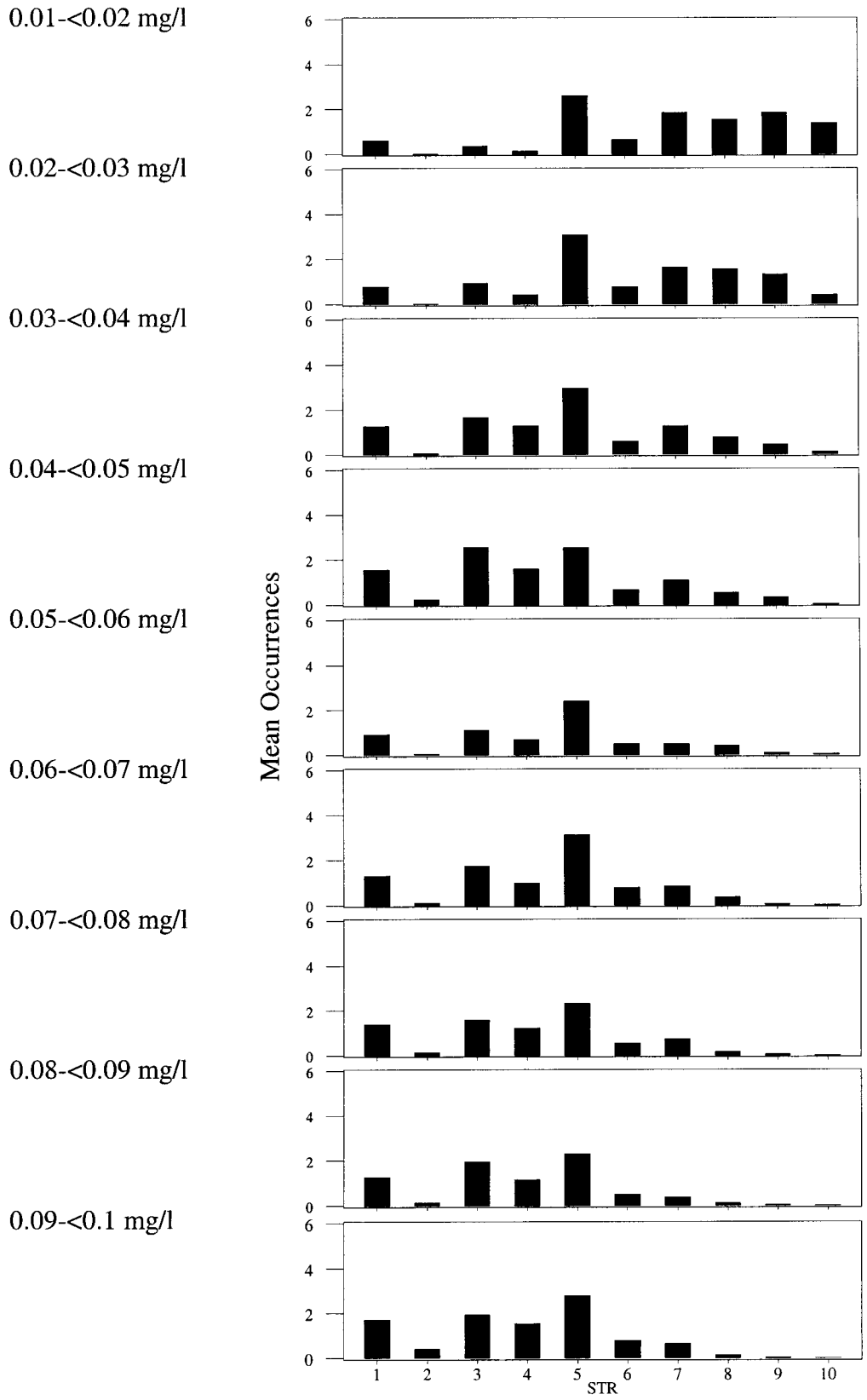


Figure 20a. Mean number of occurrences of species in each STR group, at phosphate concentrations of less than 0.1mg/l. Data presented in 0.01 mg/l band-widths, from 0.01 mg/l phosphate to 0.1 mg/l. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets (Conservation Rivers data for post-1985 surveys only). Macrophyte data matched with phosphate data either at the same site or from a site within 1 km.

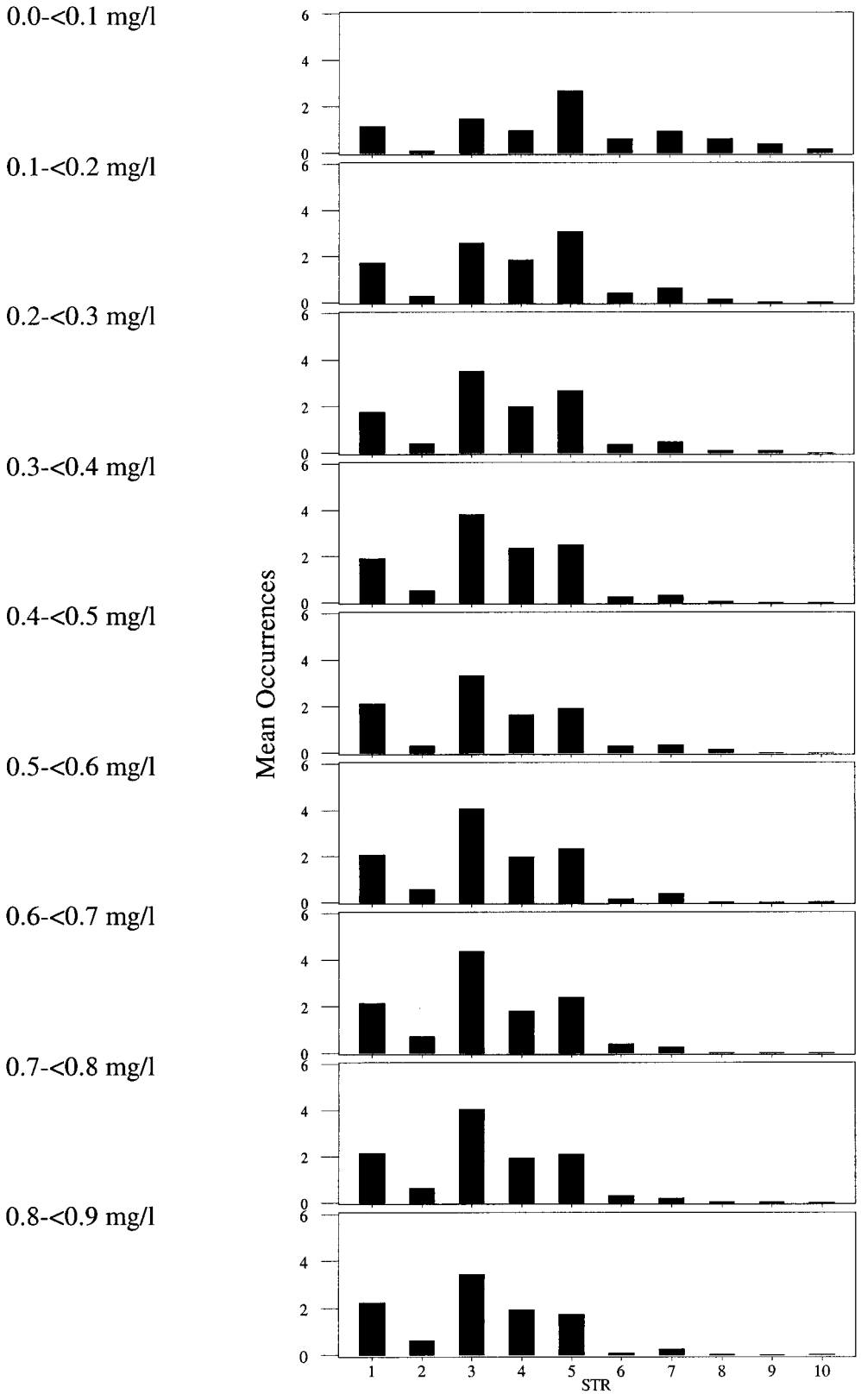


Figure 20b. Mean number of occurrences of species in each STR group, at phosphate concentrations of less than 0.9mg/l. Data presented in 0.1 mg/l band-widths, from 0.0 mg/l phosphate to 0.9 mg/l. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets (Conservation Rivers data for post-1985 surveys only). Macrophyte data matched with phosphate data either at the same site or from a site within 1 km.

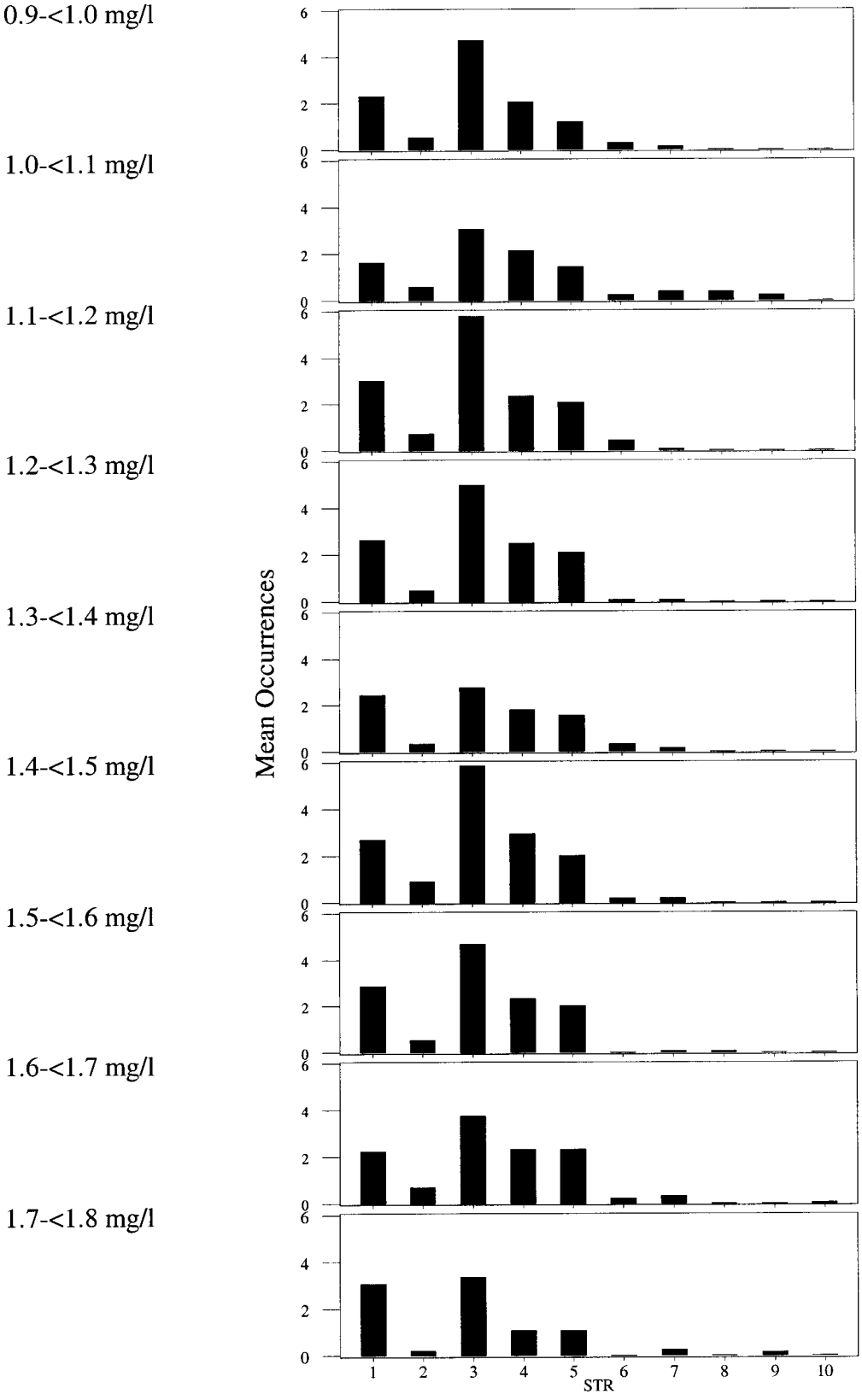


Figure 20c. Mean number of occurrences of species in each STR group, at phosphate concentrations between 0.9 and 1.8 mg/l. Data presented in 0.1 mg/l band-widths, from 0.9 mg/l phosphate to 1.8 mg/l. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets (Conservation Rivers data for post-1985 surveys only). Macrophyte data matched with phosphate data either at the same site or from a site within 1 km.

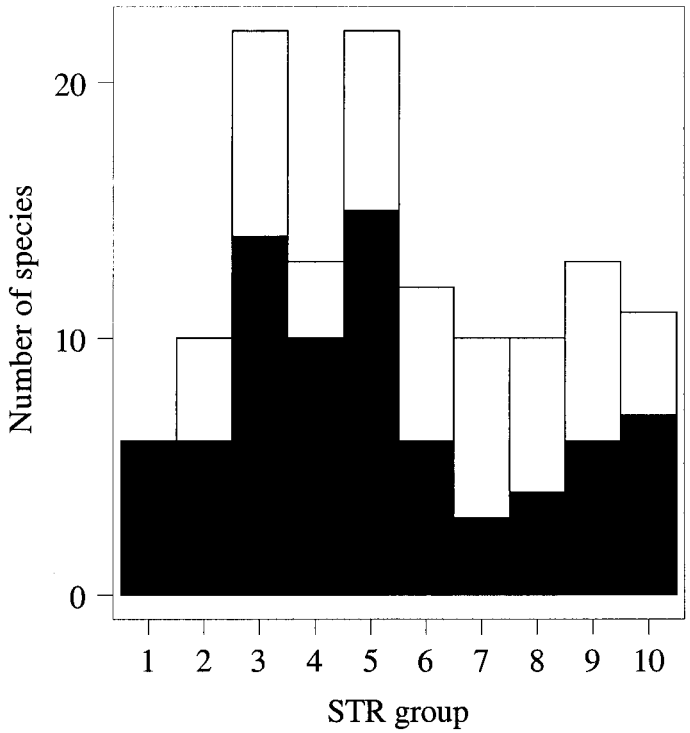


Figure 21. Number of highlighted species (black) and non-highlighted species (white) in each STR group of the MTR scoring taxa list.

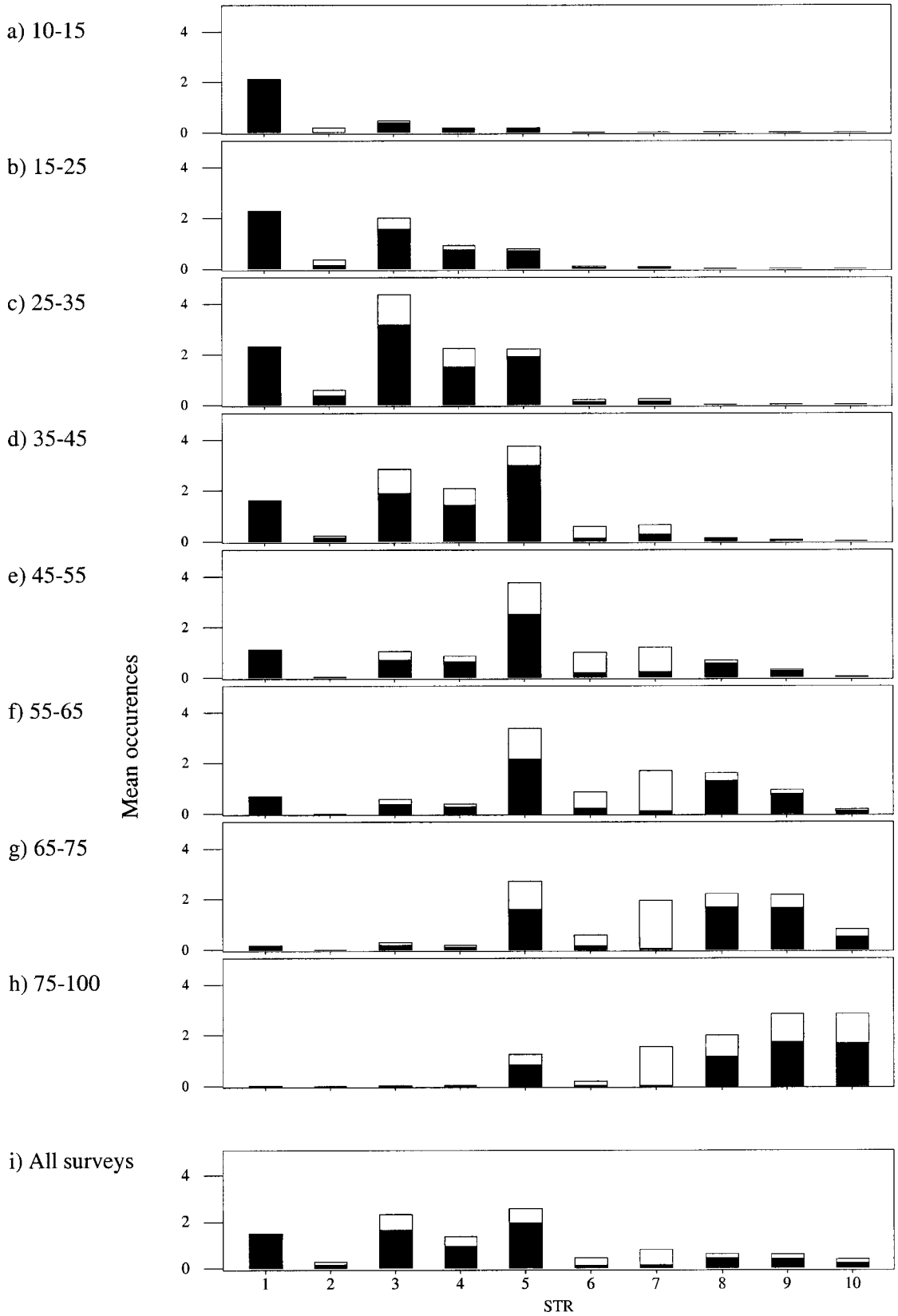


Figure 22. Mean number of occurrences of species in each STR group, at sites with an MTR in the ranges (a) 10-15, (b) 15-25, (c) 25-35, (d) 35-45, (e) 45-55, (f) 55-65, (g) 65-75, (h) 75-100, (i) all surveys (MTR 10-100). Frequency of non-highlighted species shown in white and of highlighted species shown in black. All datasets included.

3.4.3 Conclusions

1. No scoring species was found which occurred either within a sufficiently restricted range of phosphate concentration, or with a sufficiently strong relationship with phosphate concentration, to enable its use as a 'key' species, the presence or abundance of which may be useful for interpretation purposes. 'Key' associations were not examined.

2. In broad terms the STRs were found to represent a spectrum of tolerances to phosphate enrichment although incorporating a moderate degree of variability. High-scoring species were generally not found at high phosphate concentrations and low-scoring species were either cosmopolitan or found only rarely or at low abundance at low phosphate concentrations.

3. Assignment of STRs to species based on the data collected during this study, would result in some changes to STR ratings. At present, however, there is insufficient evidence to recommend substantive changes to the STR assigned to any species, with the exception of *Stigeoclonium tenue* which should be removed from the scoring list. Further research is recommended on the growth-phosphate relationship of some species to confirm the STRs. These include, but are not restricted to, *Callitriche hamulata* and *Juncus bulbosus* (see Tables 3 & 4). Additional analysis, taking cover into account, may prove useful.

4. At any one phosphate concentration, a wide range of STRs may occur. The balance of species present, however, may be disturbed by nutrient enrichment. At low phosphate concentrations, the macrophyte community is likely to be composed both of species sensitive to phosphate enrichment and those which are more cosmopolitan. Communities at high phosphate concentrations are likely to lack sensitive high-scoring species and are instead dominated by tolerant or cosmopolitan low-scoring species. This should be reflected in the MTR score (see 3.6).

5. The distribution of STRs within the MTR scoring list is biased towards STRs of 3 and 5. Although it is recognised that this may lend a corresponding bias towards MTR scores in the range 30-50, it is likely that the magnitude of this effect is small compared to the influence of other factors influencing the MTR. STR 3 and 5 species were the most commonly recorded species, with STR 2 species being surprisingly infrequent.

3.5 STR and nitrate

The summary statistics for species distribution in relation to nitrate concentration, showed a broad correlation between STR and nitrate concentration (Figure 23a & b and Figure 24). However, individual species were found to occur over a wide range of nitrate concentrations and there was much overlap between the ranges of different species. In general, STR was found to decrease as the logarithm of the mean nitrate concentration increased, but this relationship reached an apparent plateau above about 2–6 mg l^{-1}, with little apparent difference between species in the STR groups 4 to 1.

On an individual species basis, it is worth noting that *Ranunculus fluitans* has an STR of 7 but was only recorded at concentrations greater than 1 mg l^{-1} N and more frequently occurred at levels greater than 4 mg l^{-1}.

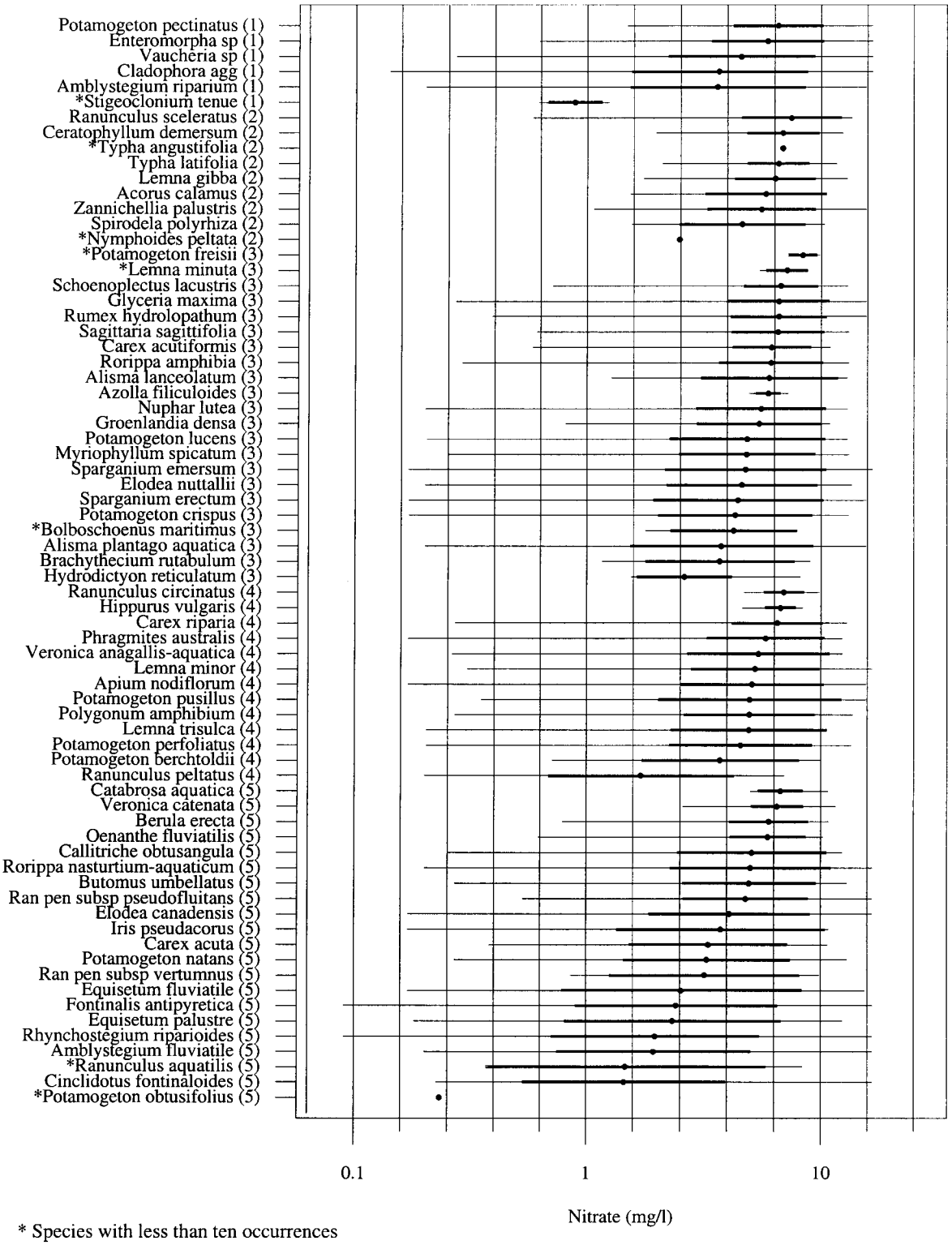
As with the STR–phosphate relationship, there are several 'outlier' species. Some of these outliers had less than ten occurrences and so confidence in the data was low. Some outliers may be indicative of enrichment by nitrate rather than phosphate. To help confirm the species for which a change in STR may be justified, a new set of rankings was derived based on the data collected during this study. Species were ordered according to the mean annual nitrate concentration at which they occurred (Figure 25a & b), then divided into ten equal groups and the species in each group assigned a 'nitrate rank'. Several species were assigned a different rank to the STR (Figure 26), some of these being markedly different (Table 5). The new rankings would need further detailed examination, requiring specific chemical data, before any firm recommendations of changes to STR could be made. It may also be useful to test the addition of a weighting factor to the mean nitrate concentration based on the cover of the species (as suggested above for phosphate).

Conclusions

There was a broad, negative correlation between STR and the logarithm of nitrate concentration, although not as strong as the STR–phosphate relationship. A change in STR may be justified for some species, but this would require further analysis before firm recommendations could be made. As with phosphate, there were no obvious species which either occurred only within a very restricted range of nitrate concentration, or with a sufficiently strong relationship with nitrate concentration, to enable its use as a 'key' species for purposes of interpretation. However, some species may be useful for distinguishing between nitrate and phosphate enrichment.

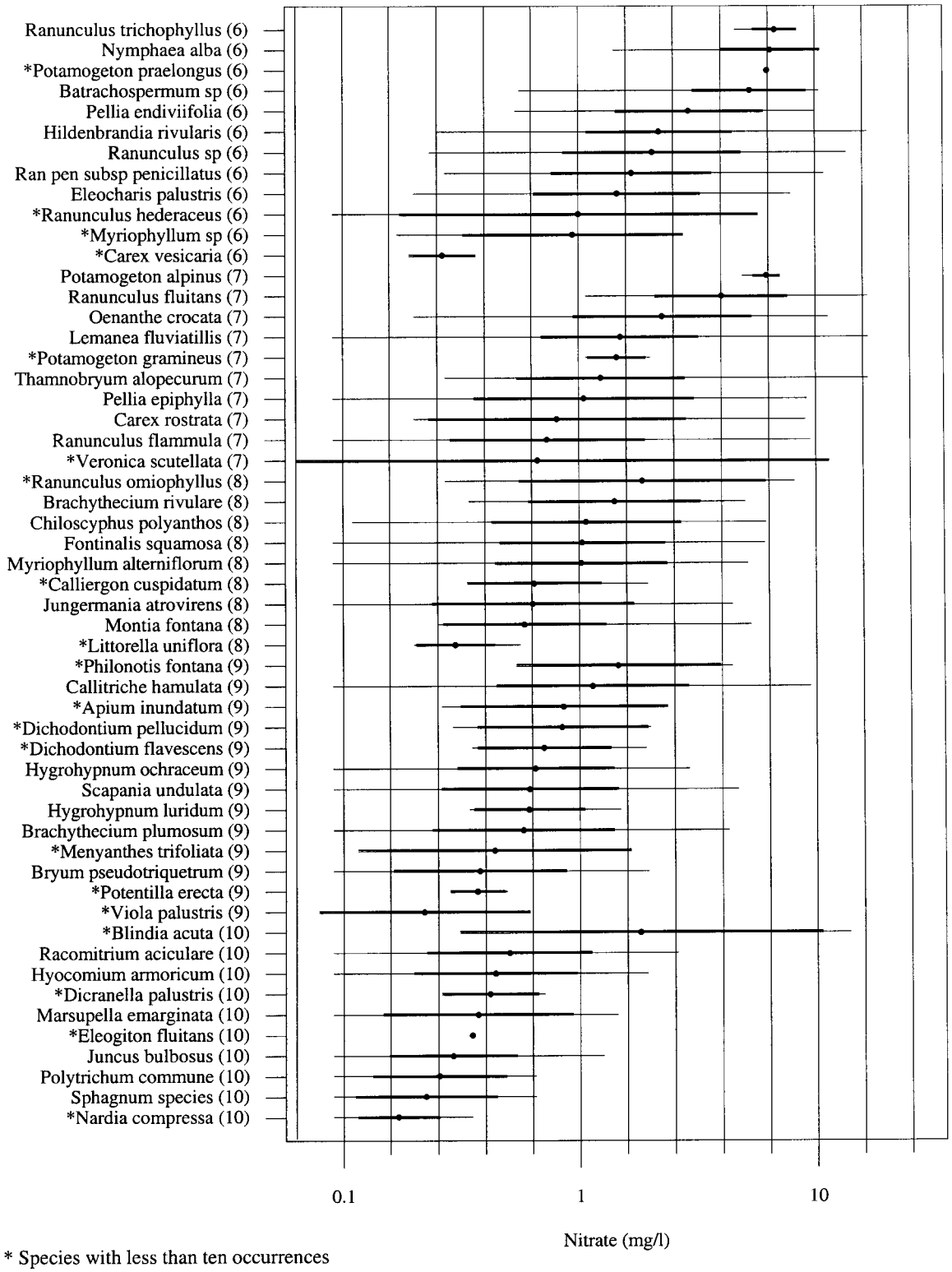
Table 5. List of species for which 'nitrate rank' differs significantly from STR. (>) indicates that the 'nitrate rank' is higher than the STR for that species; (<) indicates that the nitrate rank is lower than the STR.

Species	numbers of occurrences	STR	'Nitrate ranking'	Species present at sites with predominantly low phosphate but high nitrate
Stigeoclonium tenue	5	1	> 8	
Cladophora agg.	937	1	> 5	
Amblystegium riparium	312	1	> 5	
Vaucheria	480	1	> 4	
Nymphoides peltata	1	2	> 6	
Hydrodictyon reticulatum	31	3	> 6	
Ranunculus peltata	25	4	> 7	
Ranunculus circinatus	34	4	< 1	*
Hippuris vulgaris	39	4	< 1	*
Potamogeton obtusifolius	1	5	> 9	
Oenanthe fluviatilis	102	5	< 2	*
Veronica catenata	81	5	< 2	*
Berula erecta	120	5	< 2	*
Catabrosa aquatica	18	5	< 1	*
Carex vesicaria	4	6	> 9	
Batrachospermum	49	6	< 3	*
Potamogeton praelongus	2	6	< 2	
Nymphaea alba	14	6	< 2	
Ranunculus trichophyllus	10	6	< 1	
Potamogeton alpinus	6	7	< 2	*
Blindia acuta	3	10	< 7	



* Species with less than ten occurrences

Figure 23a. Species occurrence against the logarithm of the nitrate concentration within 1 km (summary data). I: low-STR species, ranked in STR order. Mean nitrate concentration given as a dot, with a thick line giving plus/minus one standard deviation (SD) and a thin line giving minimum/maximum. Where no SD or minimum/maximum is shown, this indicates that one or more records for the species, but only a single nitrate datum. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets (Conservation Rivers data for post-1985 surveys only). Macrophyte data matched with nitrate data either from the same site or from a site within 1 km. STR values given in brackets after the species name.



* Species with less than ten occurrences

Figure 23b. Species occurrence against the logarithm of the nitrate concentration within 1 km (summary data). II: high-STR species, ranked in STR order. Mean nitrate concentration given as a dot, with a thick line giving plus/minus one standard deviation (SD) and a thin line giving minimum/maximum. Where no SD or minimum/maximum is shown, this indicates that one or more records for the species, but only a single nitrate datum. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets (Conservation Rivers data for post-1985 surveys only). Macrophyte data matched with nitrate data either from the same site or from a site within 1 km. STR values given in brackets after the species name.

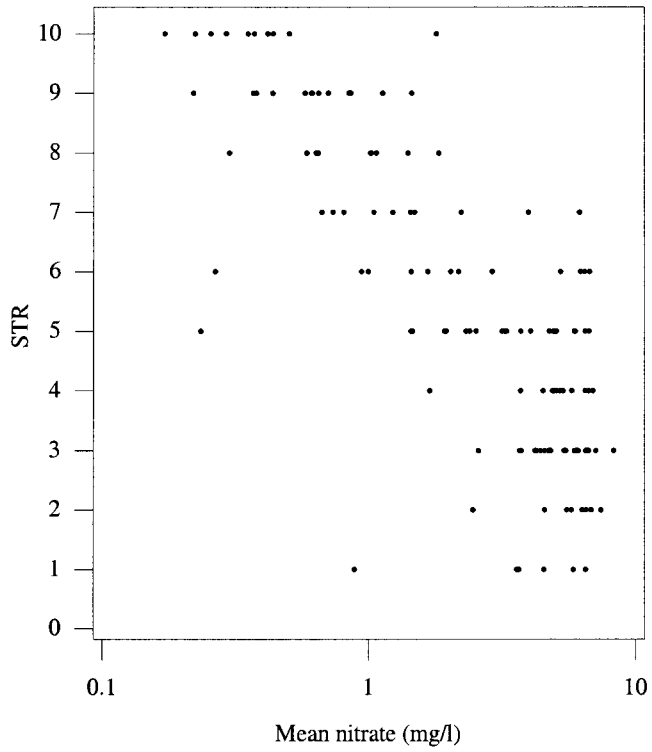


Figure 24. Relationship between STR and the logarithm of the mean nitrate concentration recorded for individual species. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets for 1985-97. Macrophyte data matched with nitrate data either from the same site or from a site within 1 km. A small random movement has been added to the Y-axis position to separate points and hence to improve clarity.

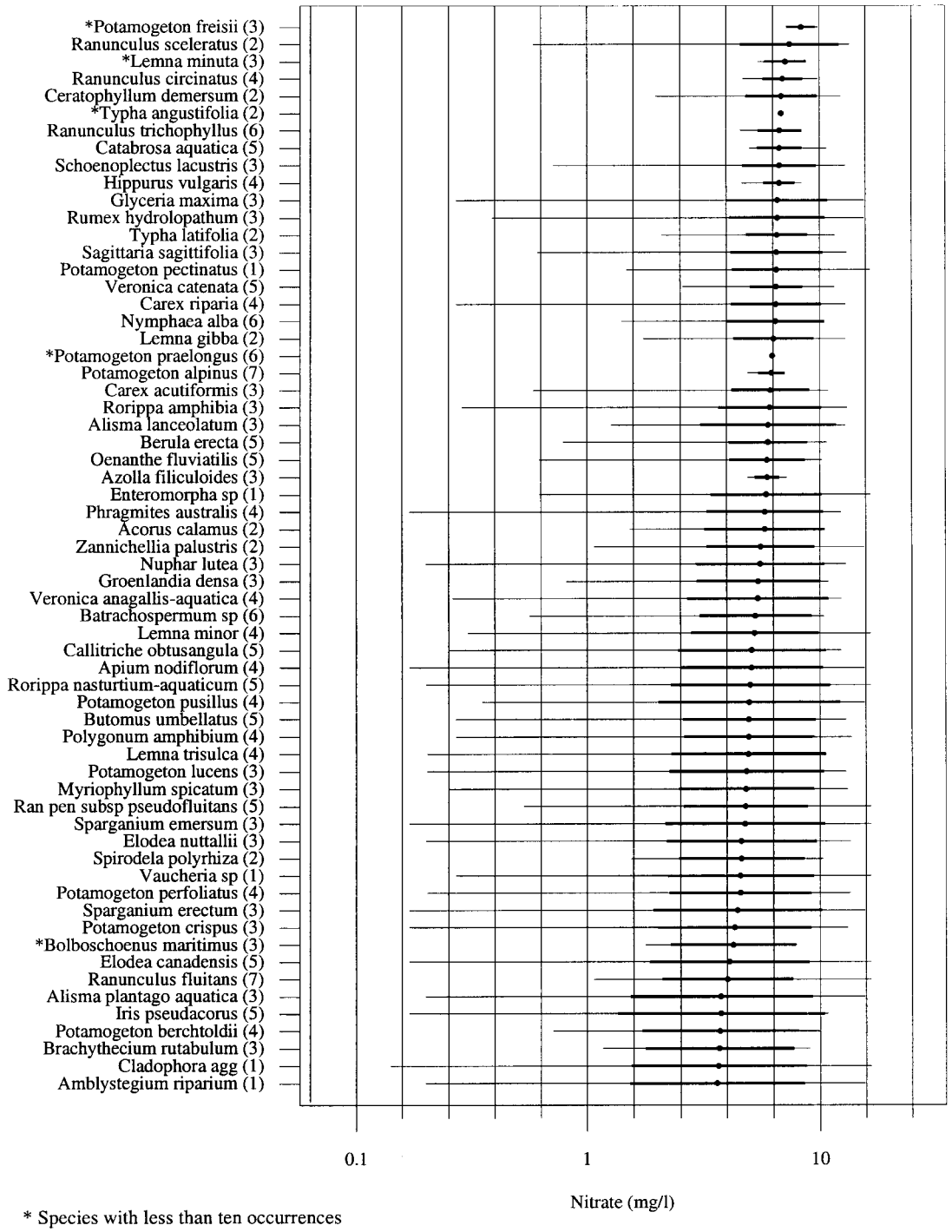


Figure 25a. Species occurrence against the logarithm of the nitrate concentration within 1 km, ranked in order of decreasing mean nitrate concentration. I: species with a high mean nitrate concentration. Mean nitrate concentration given as a dot, with a thick line giving plus/minus one standard deviation (SD) and a thin line giving minimum/maximum. Where no SD or minimum/maximum is shown, this indicates that one or more records for the species, but only a single nitrate datum. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets (Conservation Rivers data for post-1985 surveys only). Macrophyte data matched with nitrate data either from the same site or from a site within 1 km. STR values given in brackets after the species name.

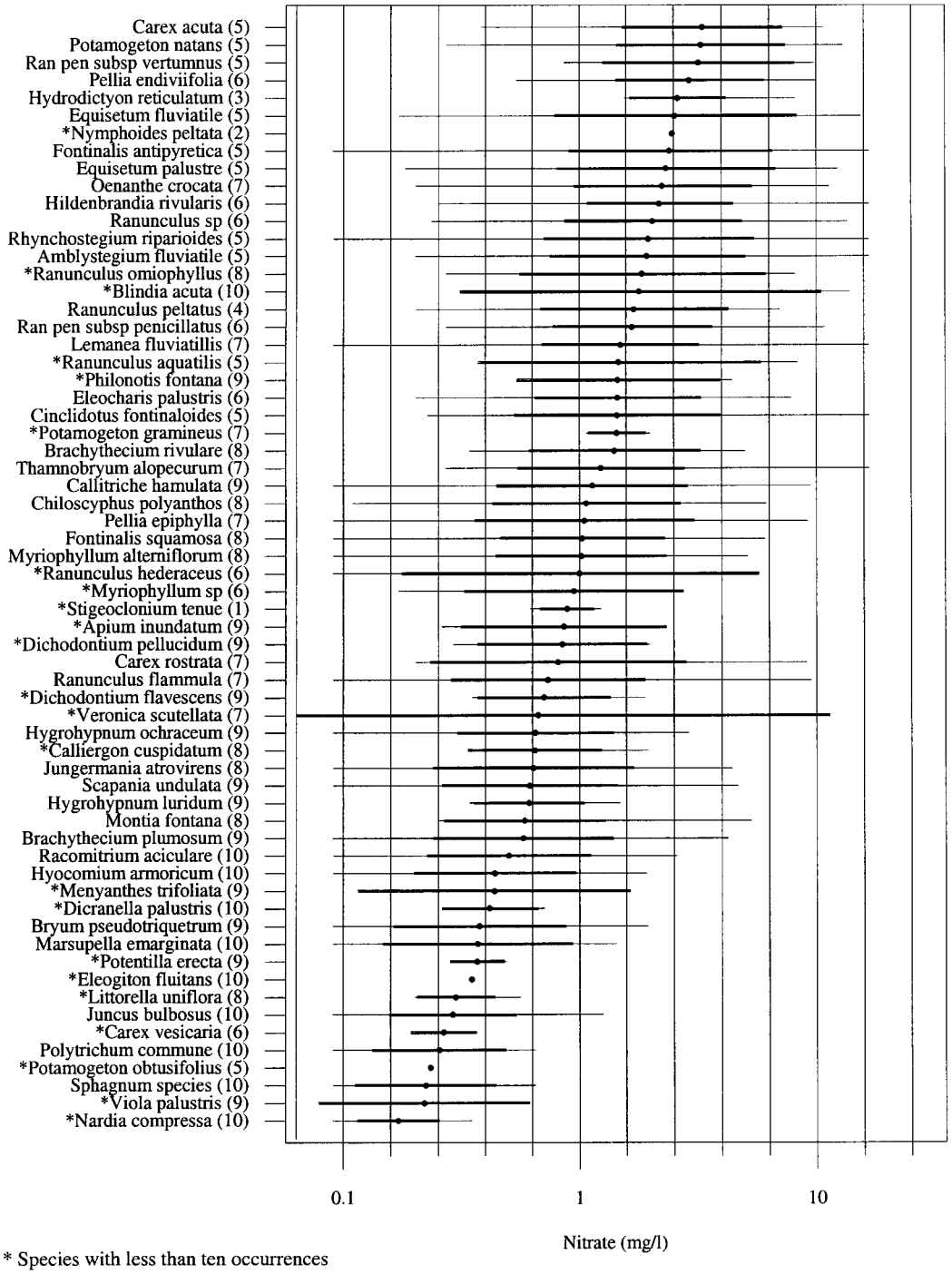


Figure 25b. Species occurrence against the logarithm of the nitrate concentration within 1 km, ranked in order of decreasing mean nitrate concentration. I: species with a low mean nitrate concentration. Mean nitrate concentration given as a dot, with a thick line giving plus/minus one standard deviation (SD) and a thin line giving minimum/maximum. Where no SD or minimum/maximum is shown, this indicates that one or more records for the species, but only a single nitrate datum. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets (Conservation Rivers data for post-1985 surveys only). Macrophyte data matched with nitrate data either from the same site or from a site within 1 km. STR values given in brackets after the species name.

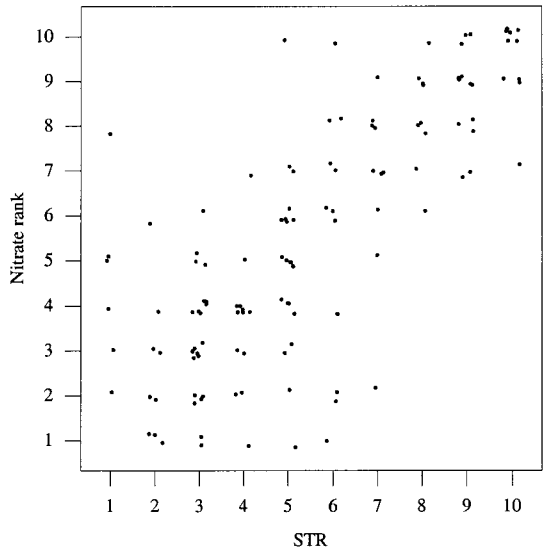


Figure 26. Relationship between STR and the 'nitrate rank'. Nitrate rank derived from EA, IFE, DoE/IRTU and Conservation Rivers datasets (Conservation Rivers data for post-1985 surveys only). Macrophyte data matched with nitrate data either from the same site or from a site within 1 km. Species then ranked in order of mean nitrate concentration at which they occur, and divided into ten scoring groups, each group comprising the same number of species. Obvious outliers are listed in Table 5. A small random movement has been added to the X- and Y-axis positions to separate points and hence to improve clarity.

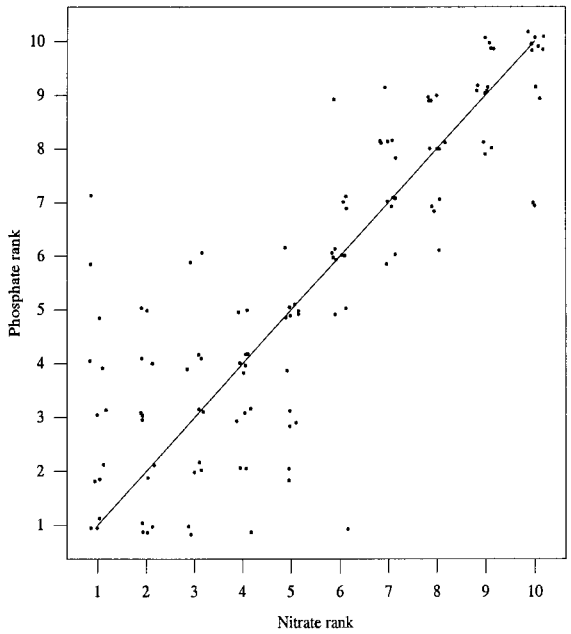


Figure 27. Relationship between 'phosphate rank' and 'nitrate rank'. Nitrate ranks derived as for Figure 26. Phosphate ranks derived from EA, IFE, DoE/IRTU and Conservation Rivers datasets (Conservation Rivers data for post-1985 surveys only). Macrophyte data matched with phosphate data either from the same site or from a site within 1 km. Species then ranked in order of mean phosphate concentration at which they occur, and divided into ten scoring groups, each group comprising the same number of species. A small random movement has been added to the X- and Y-axis positions to separate points and hence to improve clarity.

3.6 MTR and phosphate

3.6.1 MTR - phosphate relationship

The relationship between MTR and annual mean phosphate concentration (Figure 28a) shows that MTR declines with increasing phosphate concentration up to about 1 mg l^{-1}, above which MTRs are generally low with no clear pattern apparent. A linear regression between MTR and phosphate concentration is significant (P = 0.01), but only explains 14% of the variation. There is, however, a stronger correlation between MTR and the logarithm of phosphate, this explaining 30% of the variation (Figure 28b). The relationship between MTR and the logarithm of phosphate is not constant across the range of phosphate concentrations recorded. The relationship is strongest for phosphate concentrations of less than 0.5 mg l^{-1}, and poorest for phosphate concentrations above 1.0 mg l^{-1}.

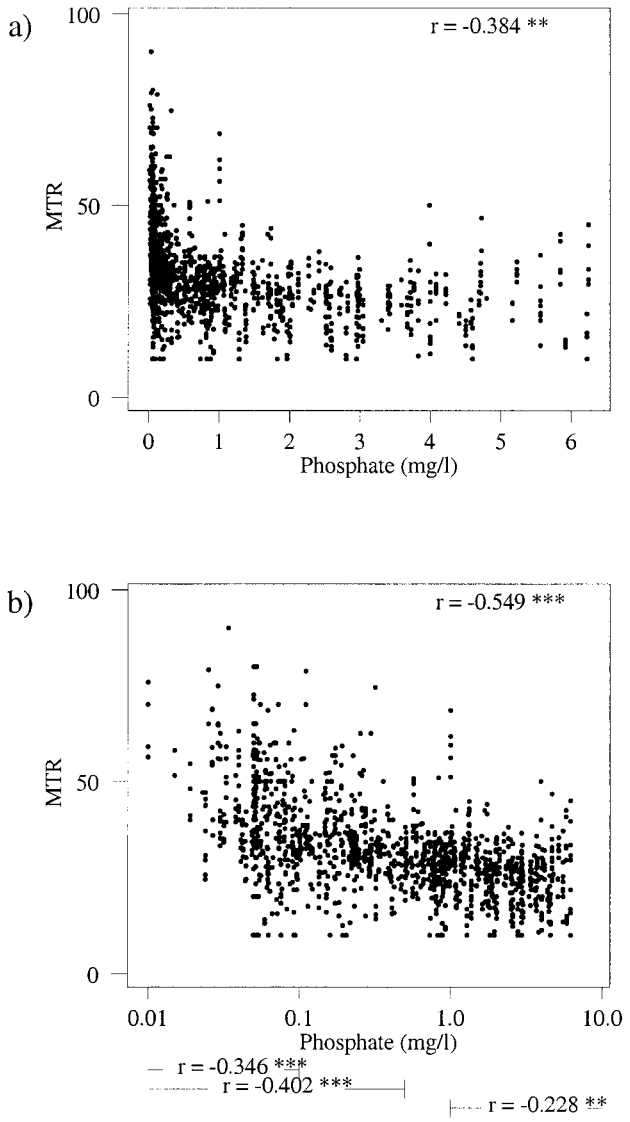


Figure 28. Relationship between MTR and (a) phosphate concentration and (b) the logarithm of phosphate concentration, for EA, IFE, DoE/IRTU, RHS and Conservation Rivers datasets. (Conservation Rivers data for post-1985 surveys only, matched to phosphate concentrations within 1 km). Correlation coefficients for sections of the logarithmic relationship are given below (b). ** highly significant P = 0.01. *** very highly significant P = 0.001.

It is recognised that the presence of plant species may be more strongly influenced by the maximum phosphate concentration occurring within the growth season, rather than the mean concentrations. As data on seasonal phosphate maxima were not available within the scope of this project, an approximation was made using the statistical upper estimate of the variance of the annual mean from the monthly samples. This estimate (the annual mean phosphate concentration plus twice its standard deviation) was correlated with the MTR, but very similar patterns were found: highly significant negative correlations for both normal ($r = -0.353^{***}$) and logarithmic relationships ($r = -0.588^{***}$). The optimal type of statistical normalisation of the data was not determined.

3.6.2 Surveys not conforming to the expected MTR-phosphate relationship

Surveys with a high phosphate concentration and a relatively high MTR score were investigated to determine why they did not comply with the overall and 'expected' trend of decreasing MTR with increasing phosphate concentration. The most common causes of high MTR scores on these surveys was the presence of species with an STR of 4 or more, occurring in abundance. These species included: *Ranunculus fluitans*, *Potamogeton natans*, *Rorippa nasturtium-aquaticum* and *Berula erecta*. The latter three species all have an STR of 5, but *R. fluitans* has an STR of 7 and so would not be expected at high phosphate concentrations. Assuming it has been correctly identified in these surveys, further analysis may thus be required to confirm the STR assigned to *R. fluitans*, especially when compared to the relationship with nitrate concentration (only found above 1 mg l^{-1} nitrate, see 3.5).

Another general cause of high MTR at high phosphate concentration, may be the presence of species which may not reflect the water quality at the site. For example, high-scoring emergent species such as *Oenanthe crocata* (STR 7), may be growing more on the bank than in the water channel. Atypical conditions (such as washout of plants) and surveyor error (mis-identification or missing species) may also give rise to unexpected results. For example, this may explain the unusually high MTR recorded in one set of four surveys, when a low scoring species was absent in one survey but recorded in three other surveys at the same site. One site of particular note as an outlier is the River Erme at Ivybridge, Devon, where many species of high scoring mosses were still recorded downstream of a WWTW despite the large discrete increase in phosphate (1.0 mg l^{-1}).

3.6.3 Analysis of subsets of data

The MTR-phosphate relationship was re-examined for sub-sets of the dataset. The purpose of this was two-fold: to establish whether a particular bias in one (or more) of the data sources was unduly skewing the overall results; and to determine whether the relationship between MTR and phosphate is stronger in certain geographical areas.

The clearest relationship between MTR and phosphate concentration was for the Conservation Rivers database (Figure 29). This database comprises surveys from rivers throughout Great Britain, selected as potentially being of regional or national conservation interest and surveyed at various sites along their lengths. These rivers represent a broad spectrum of quality and physical character, without a bias in phosphate concentration as indicated by its wide range (Figure 29). There is a significant correlation between MTR and phosphate both when data are used for all surveys with matched GQA phosphate data from within 1 km (Figure 29a) and when this dataset is restricted to only those surveys completed after 1985 (Figure 29b). There is no indication that the difference in time between the date of

the macrophyte survey and the date of the chemical sample date had any apparent effect on the relationship between MTR and phosphate concentration, since both correlation coefficients are highly significant.

The relationship between MTR and phosphate concentration would appear to be stronger in England and Wales than in Northern Ireland (Figures 30a-d). The datasets are not fully comparable, however, as the limits of detection (or minima) vary from 0.024 mg l^{-1} in the former to 0.05 mg l^{-1} in Northern Ireland. In addition, no phosphate concentrations above 1 mg l^{-1} were recorded in Northern Ireland, this presumably being a reflection both of existing phosphate removal in N. Ireland but also the different sampling strategy to England and Wales, ie not upstream and downstream of QDs.

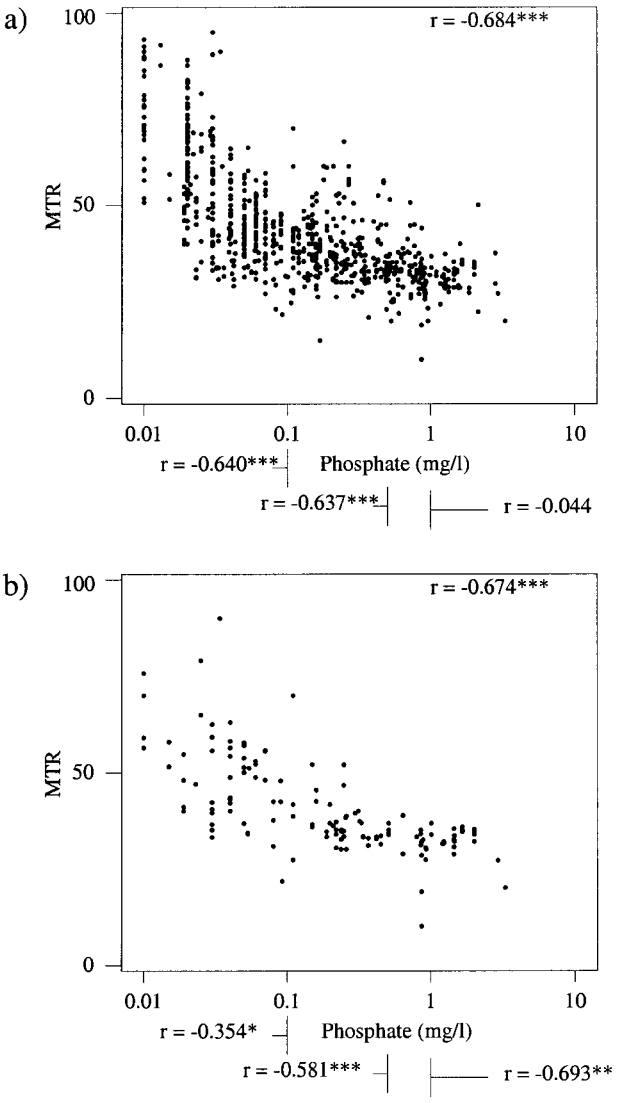


Figure 29. Relationship between MTR and the logarithm of soluble phosphate concentration, derived from Conservation Rivers data for England and Wales: (a) all surveys (b) surveys completed after 1985. Macrophyte data matched with phosphate data either from the same site or from a site within 1 km on the same river.

Further division of the data for England and Wales into Agency regions shows best correlations in Midlands and Thames regions, with the lowest concentrations of phosphate in Wales (data not shown). Again, however, the datasets are not comparable and so no firm inferences can be drawn as to the comparative usefulness of the MTR in different regions. The data do confirm the different MTR and phosphate ranges recorded in different regions.

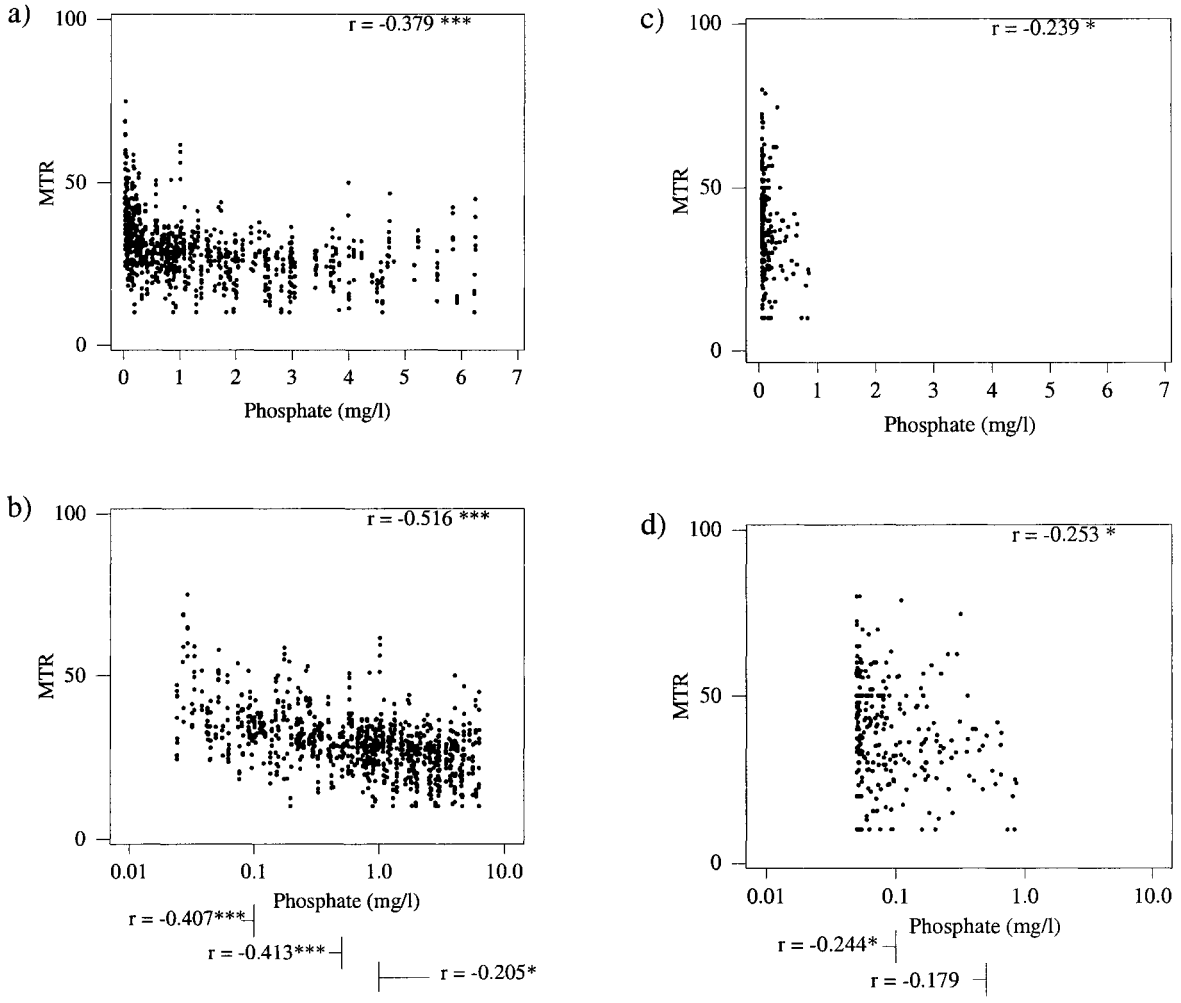


Figure 30. Relationship between (a) MTR and phosphate, and (b) MTR and the logarithm of phosphate concentration, derived from EA and IFE data for England and Wales; and between (c) MTR and phosphate, and (d) MTR and the logarithm of phosphate concentration, derived from DoE/IRTU data for Northern Ireland. Note that phosphate data for (c) and (d) have a lower limit of detection of 0.05 mg l^{-1}.

3.6.4 Conclusions

MTR is significantly correlated with phosphate concentration and especially with the logarithm of phosphate concentration. The correlation is stronger at concentrations below 1.0 mg l^{-1} and even more so at concentrations below 0.5 mg l^{-1}. Although differences in the strength of this relationship may occur between geographical areas, there is insufficient evidence to test this.

3.7 MTR and nitrate

The correlation between MTR and nitrate concentration (Figure 31) is as significant as the correlation between MTR and phosphate. In contrast, however, the conversion of nitrate to a logarithm did not improve the correlation as occurred with phosphate (cp Figures 28 and 31). The minimum nitrate level recorded of 0.11 mg l^{-1} is unlikely by itself to limit plant growth; although most of the lower nitrate concentrations (< 0.5 mg l^{-1}) are in N. Ireland, there is a scattering throughout Britain. The correlation, or cross correlation, of phosphate and nitrate was significant but lower than either of those for phosphate or nitrate with MTR, showing that there is considerable variation in the nitrate to phosphate ratio, and that the MTR-to-phosphate and MTR-to-nitrate relationships vary independently.

There is no clear relationship between the nitrate:phosphate ratio and MTR (Figure 32a & b). The majority of ratios lie in the range from 5:1 to 50:1, with many centred around a ratio of 10:1. These characteristics are consistent with those considered to be typical for aquatic plants (cp Kelly & Whitton 1993 for algae). Ratios outside this range were recorded, but would have a marked effect on the plant community only if the absolute concentrations of nutrients were limiting to plant growth. For example, a N:P ratio of 0.35 was recorded at a site downstream of Hailsham South WWTW on the White Dyke, East Sussex, where the phosphate concentration was 5.85 mg l^{-1} and the nitrate was only 2.06 mg l^{-1}; but this level of nitrate is more than sufficient for maximum growth.

The correlation of the mean concentration of phosphate and nitrate for individual species does not confirm the expected strong relationship at the normally accepted ratio of 10:1 for N:P but there is, however, a parallel relationship at a higher ratio for much of the range (with an intercept on the graph at about 0.2 mg l^{-1}, or the normal detection limits, Figure 33). There are several outliers, of which three in particular may have a greater phosphate tolerance (*Juncus bulbosus*, *Ranunculus hederaceus* and *Nymphoides peltata*), whereas four others may have a greater tolerance to nitrate (*Philonotis fontana, Ranunculus vertumnus, Potamogeton freisii* and *Catabrosa aquatica*, cp species with asterisks in Tables 4 & 5). The distribution of these species may merit further attention, to investigate whether they could be useful indicators to distinguish between phosphate and nitrate enrichment. The capacity to distinguish may be crucial in demonstrating whether reduction measures of a specific nutrient would be beneficial.

Conclusions

An acceptable correlation exists between MTR and nitrate concentration; not merely a product of the N:P ratio but as a similar but independent relationship. Although the level of enrichment attributable to nitrate as opposed to phosphate cannot be determined using MTR at this stage, some species potentially may be useful in making this distinction (further work is needed to confirm this). The minimum nitrate concentration recorded (0.11 mg l^{-1}), however, is unlikely by itself to be limiting to plant growth and so in most cases phosphate is likely to the limiting factor.

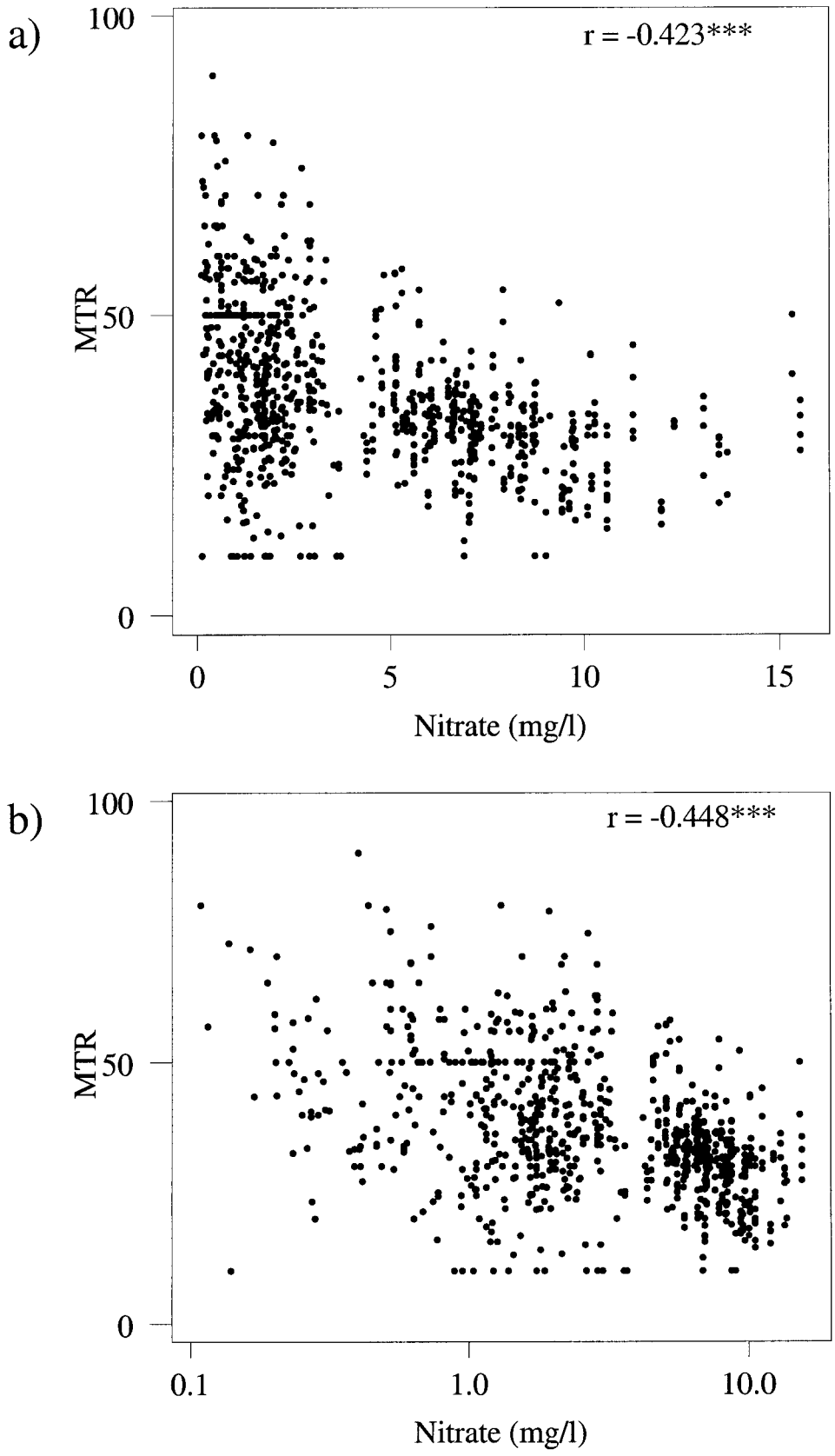


Figure 31. Relationship between MTR and (a) nitrate concentration and (b) the logarithm of nitrate concentration. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets (Conservation Rivers data for post-1985 surveys only). Macrophyte data matched with nitrate data from either the same site or from a site within 1 km.

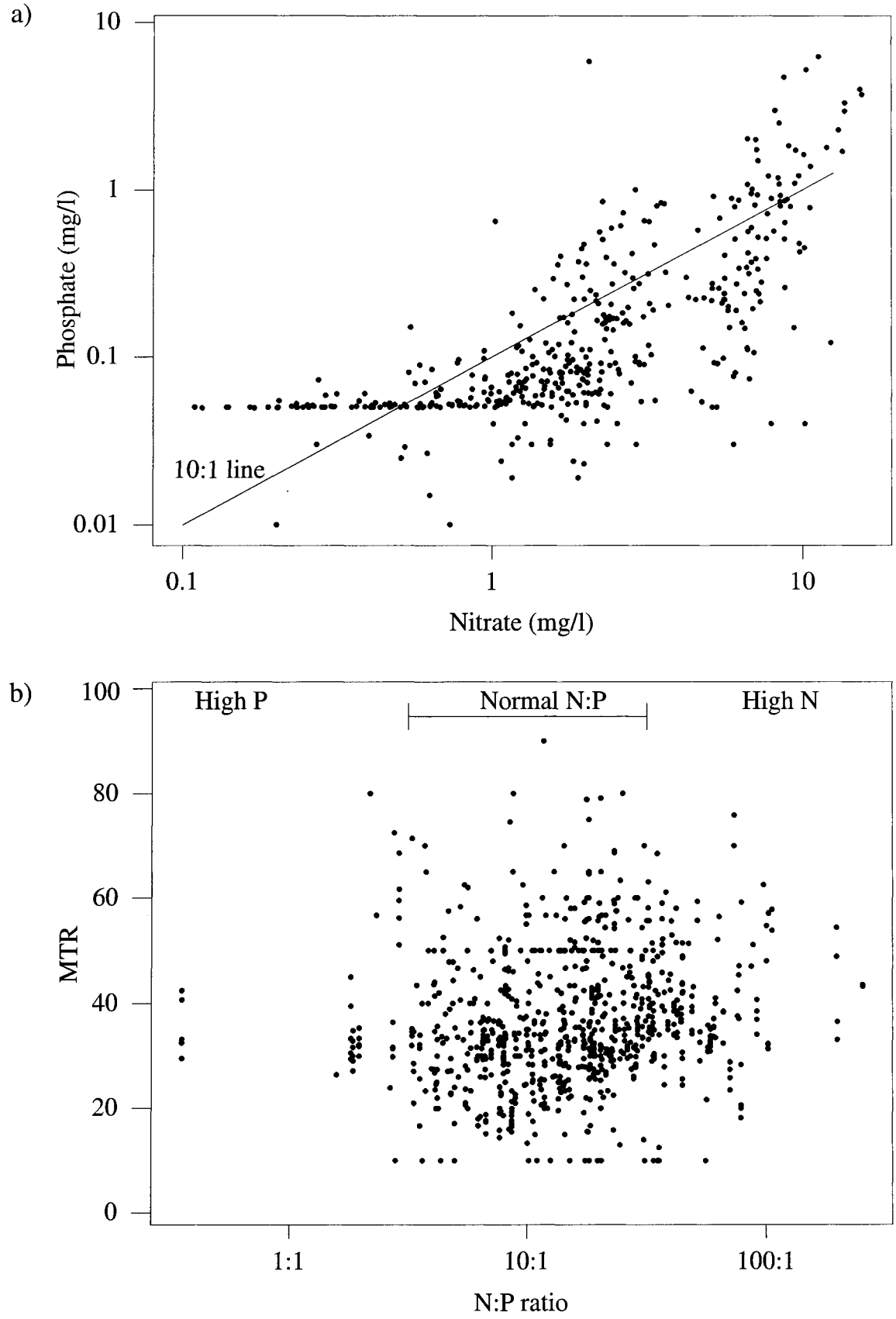


Figure 32. Relationship between (a) nitrate and phosphate concentration and (b) MTR and the ratio of nitrate to phosphate (N:P ratio) on a logarithmic scale. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets (Conservation Rivers data for post-1985 surveys only). Macrophyte data matched with nitrate or phosphate data from either the same site or from a site within 1 km.

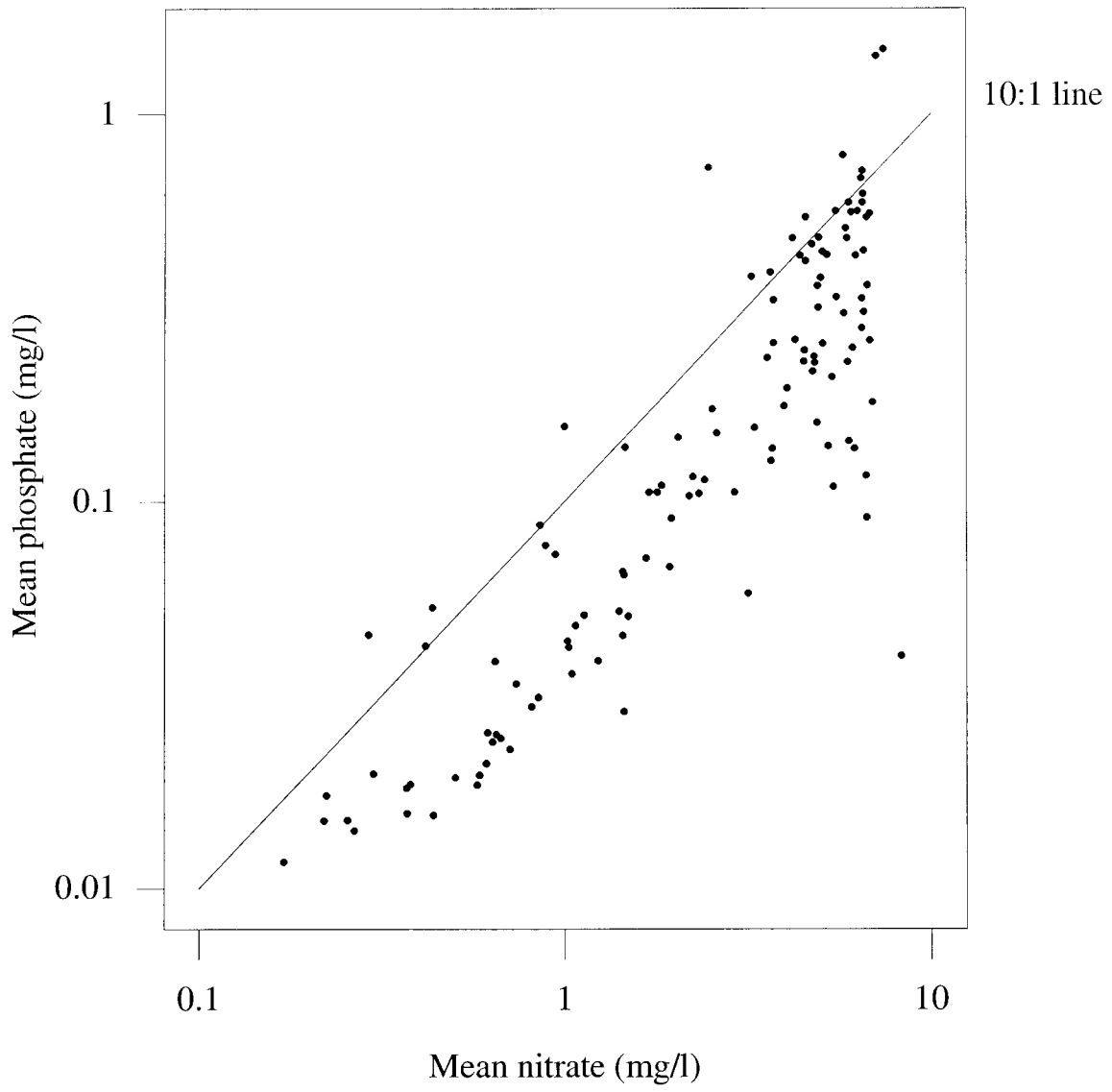


Figure 33. Relationship between the mean phosphate concentration and the mean nitrate concentration at which species were found to occur. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets (Conservation Rivers data for post-1985 surveys only). Macrophyte data matched with nitrate and phosphate data from either the same site or from a site within 1 km.

3.8 Determining change in nutrient status

3.8.1 Downstream changes in nutrient status

To test whether the general relationship between MTR and nutrient status would allow downstream changes in trophic status to be assessed, MTR, phosphate and nitrate data for individual rivers were examined at two scales: (i) localised changes downstream of a point-source input such as a QD; (ii) changes along the length of a river.

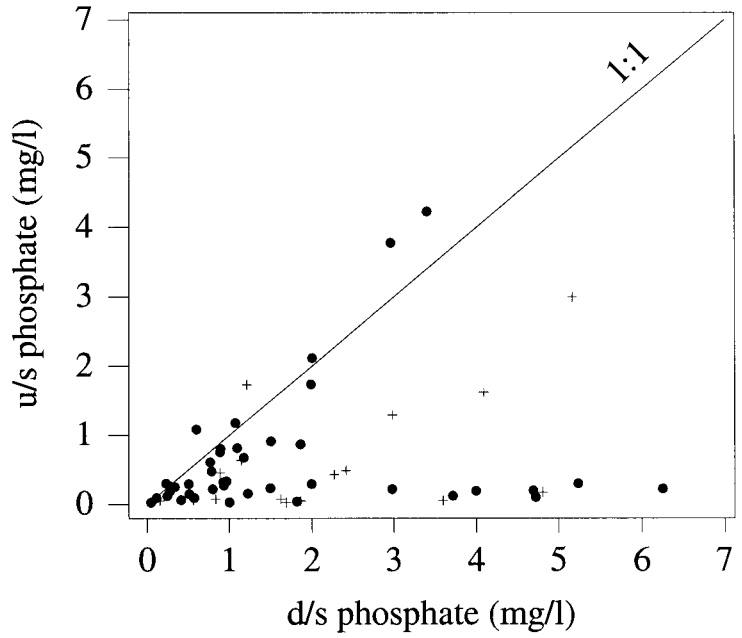
Localised changes

Phosphate and nitrate concentrations downstream of QDs were found to be generally similar or higher than the corresponding upstream site (Figure 34a & b). However, there was a less consistent downstream decrease in MTR score, regardless of the physical comparability of the paired sites. Only 59% of the 651 pairs of sites showed a decrease in MTR (Figure 35). Several of these decreases were significantly large, however, and demonstrated a definite change in trophic status. Although these paired sites were predominantly in the MTR 20–50 range, the upstream-downstream differences were similar across the whole MTR range recorded (MTR 10–80).

When the percentage downstream change in phosphate concentration is related to the percentage downstream change in MTR score, the expected decrease in MTR when phosphate decreases at the downstream site is shown on some surveys, but there are many exceptions to this (Figure 36a). There are even more exceptions when a similar analysis is undertaken for nitrate concentration (Figure 36b).

Data were limited not only because pairs of sites were required, but also because only about half of these pairs had phosphate-monitoring sites both upstream and downstream with data available and which were considered, by Agency data suppliers, to be sufficiently close. The proximity of the reported chemical sampling position to MTR sites for the above pairs, was confirmed from maps.

a)



b)

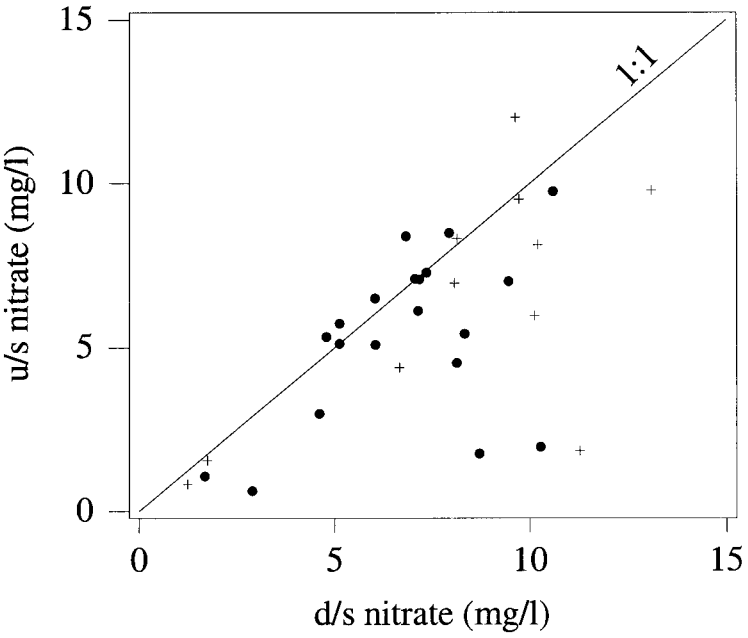


Figure 34. Relationship between concentrations of (a) phosphate and (b) nitrate, upstream (u/s) and downstream (d/s) of qualifying discharges. Key: (•) paired sites that are physically comparable (>50% similar: suffix of confidence of I or II). (+) paired sites that are physically dissimilar (<50% similar: suffix of confidence of III). Data from EA and IFE surveys. The diagonal line indicates the 1:1 ratio, where there is no difference between upstream and downstream concentrations.

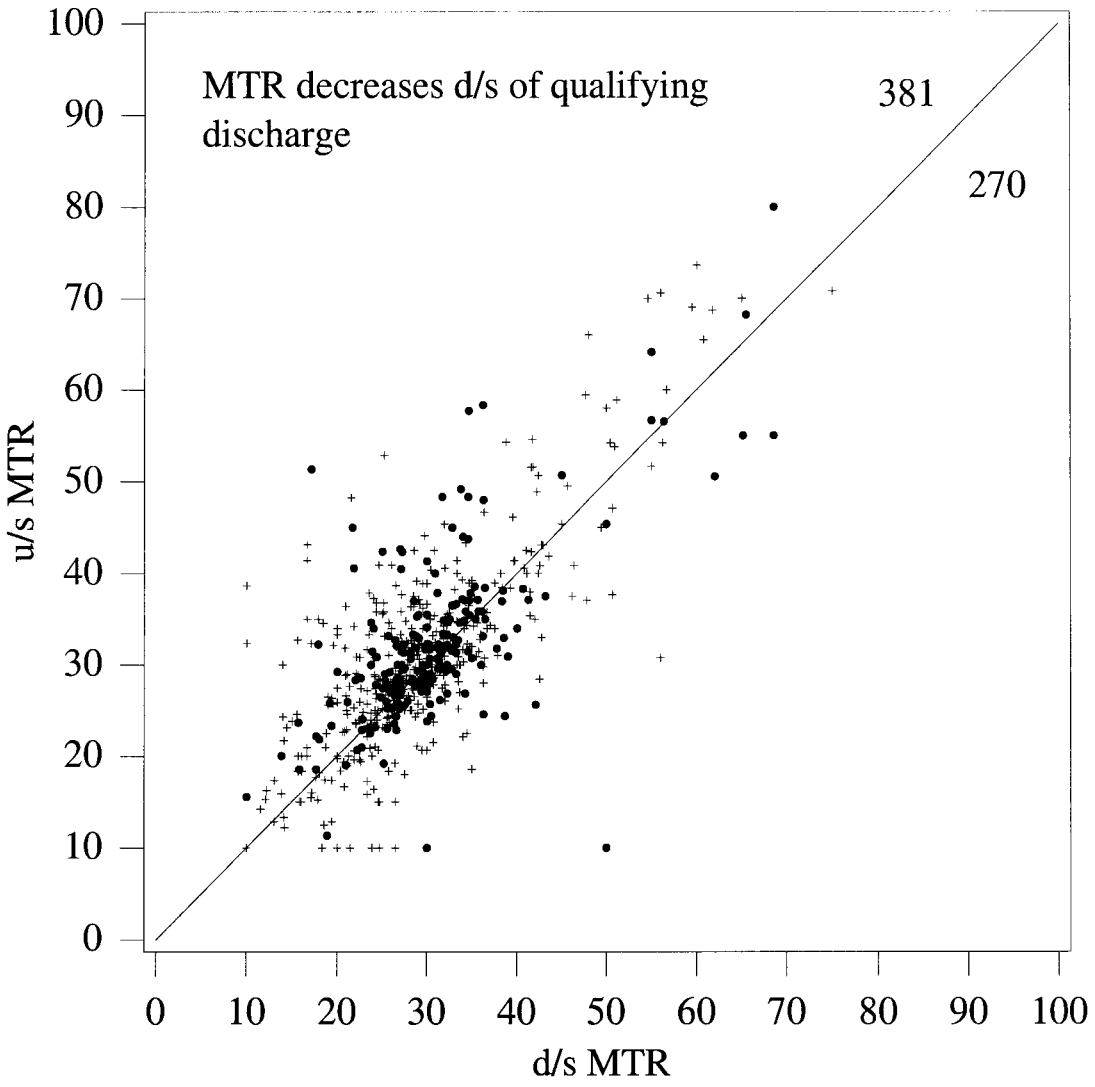
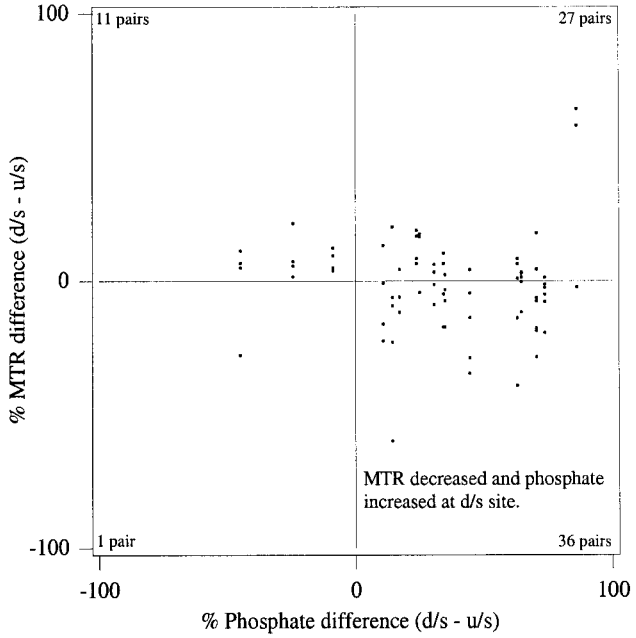


Figure 35. Relationship between MTR upstream (u/s) and downstream (d/s) of qualifying discharges. Key: (•) paired sites that are physically comparable (>50% similar: suffix of confidence of I or II). (+) paired sites that are physically dissimilar (<50% similar: suffix of confidence of III). Data from EA and IFE surveys. The diagonal line indicates the 1:1 ratio, where there is no difference between upstream and downstream MTR scores. The number of pairs of sites lying either side of this line are shown.

a)



b)

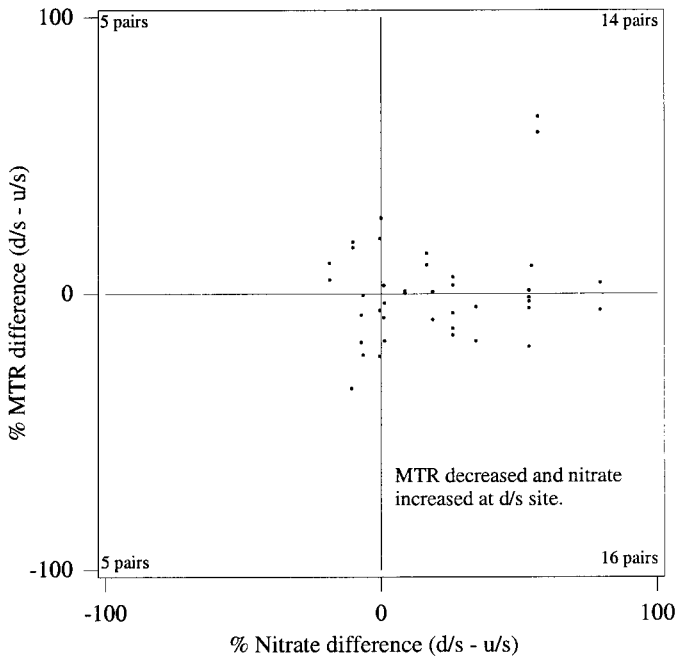


Figure 36. Percentage change in MTR score against the percentage change in (a) phosphate and (b) nitrate concentration, for paired sites upstream (u/s) and downstream (d/s) of qualifying discharges. Data only included for paired sites that are physically comparable (>50% similar: suffix of confidence of I or II). Data from EA and IFE surveys.

Changes along the length of a river

Downstream trends in MTR along the length of a river were examined for the rivers Avon (Hampshire), Eden (Cumbria), Ribble (Lancashire) and Welland (Leicestershire/ Lincolnshire). Data were extracted from both the Conservation Rivers database, relating to surveys undertaken in the 1970s or 1980s, and from the surveys undertaken in the 1990s by the Agency and IFE (Figure 37). Although there is some variation between the MTR values recorded from the two datasets (Conservation Rivers and Agency/IFE), this is not always the case and where it does exist, may be only as great as the variation within the individual datasets or may be a reflection of different site locations and sampling strategies. It was thus considered feasible to use the combination of the two datasets to indicate general downstream trends in MTR, while still recognising that temporal changes may have occurred.

The results presented indicate that a similar trend of MTR should not be expected to occur along the length of different rivers. The rivers Eden and Ribble both show some decrease in MTR within the first 20 km from the source when calculated from data of the 1970s and 1980s (but with some evidence of lower MTRs in recent years). In contrast, the Avon and Welland show little mean difference in MTR along almost their entire length (with no evidence of a recent decline in MTR). Although small upstream–downstream changes in MTR do occur at QDs, levels soon seem to return to the average level or to decrease only slightly downstream. This downstream consistency in MTR in the Avon and Welland may reflect the importance of the effects of diffuse nutrient inputs and of nutrient cycling within the river.

The difference in the downstream MTR pattern in the Avon and Welland compared to the Eden and Ribble, may be attributable to their differing habitat and geological characteristics, as well as their overall nutrient status. The Eden and Ribble both arise at about 570m on predominantly carboniferous limestone before flowing over millstone grit or mudstones and sandstones in their more downstream channels. The more lowland Welland and Avon both arise on Liasses and flow over Permian or Triassic sandstones and mudstones (although there is a downstream area of clay on the Welland). Water samples during the spring and summer in the mid 1990s, show that the rivers Eden and Ribble are relatively low in phosphate (<0.15 mg l^{-1} maximum) and nitrate, whereas the Welland and Avon have higher concentrations and may reach around 2.2 mg l^{-1} P or 10 mg l^{-1} N at maximum.

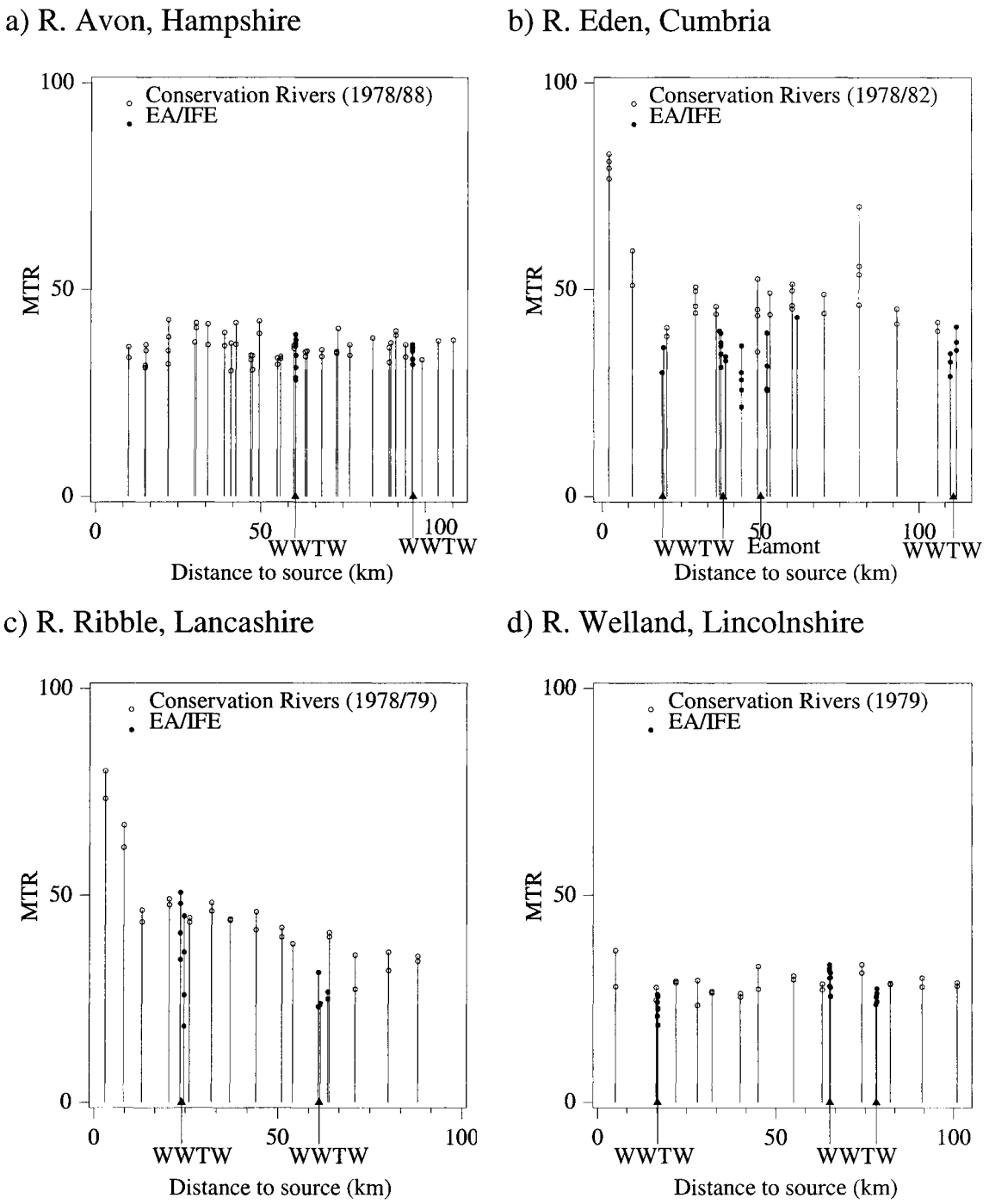


Figure 37. Downstream trends in MTR from surveys in the 1970s or 1980s from the Conservation Rivers dataset (○), and in the 1990s from EA/IFE surveys (●), along (a) River Avon, Hampshire, (b) River Eden, Cumbria, (c) River Ribble, Lancashire and (d) River Welland, Lincolnshire/Leicestershire. Position of wastewater treatment works (WWTWs) are marked (▲). Note also the effect of the junction with the River Eamont on the River Eden.

3.8.2 Temporal changes in nutrient status and differences between rivers

Although important, it was not possible to evaluate the performance of the MTR at assessing changes in nutrient status at the same site over a period of time as no adequate data are currently available. Thus only broad inferences can be made based on the overall relationship between the growth of individual species, STRs, MTR and nutrient status. It is recommended, however, that MTR data continue to be gathered together with chemical data, particularly at sites where phosphate is to be removed from the QD.

Although the MTR system was designed specifically to detect downstream changes in trophic status at QDs, the potential for expanding its application to the detection of differences between rivers, was also considered in this project. This involved analysis of factors other than nutrient status which may influence the MTR (Chapter 4) and consideration of the interpretation of MTR results (see 5.4).

3.8.3 Conclusions

1. A general pattern of decreasing MTR downstream was often seen below QDs, although MTR may also vary along the length of a river in relation to diffuse nutrient inputs.

2. The performance of the MTR at assessing changes along the length of rivers cannot be evaluated until further data on diffuse inputs and physico-chemical parameters become available for analysis. To allow for this evaluation, it is recommended that MTR monitoring of sites continue, particularly the sampling of intermediate sites between existing MTR sites and the systematic surveys of whole river systems. This should include both macrophyte and chemical sampling.

3. The performance of the MTR at assessing temporal changes in trophic status cannot, similarly, be evaluated until adequate time-series data have been gathered. To allow this evaluation, it is recommended that MTR monitoring of sites continue, particularly where phosphate reduction measures are due to commence.

3.9 National applicability

A standard assessment method must be applicable on a national basis. In terms of individual scoring species, natural regional variations in plant species were found to occur within the British Isles, as would be expected due to habitat and geological differences, amongst other factors. The distribution ranges recorded for most of the MTR-scoring species (Appendix 5) are broadly comparable to those published by Hill et al (1991) and by Preston and Croft (1997), although generally with fewer occurrences due to the smaller size of the dataset and the targeted nature of sampling strategies. This indicates that the ranges recorded in the MTR surveys are representative of the natural distribution ranges of the plant species.

The distribution of the following species differ in MTR surveys compared to those recorded by Hill et al (1991) and by Preston and Croft (1997):

- *Jungermannia atrovirens* - a few more records in north east Scotland and an occurrence in the New Forest, Hampshire, compared to published distribution;
- *Amblystegium fluviatile* - new records for Northern Ireland, East Anglia, Kent, Hampshire, Dorset, Somerset and more occurrences in Scotland;
- *Amblystegium riparium* - more records for Northern Ireland than in published distribution;
- *Dichodontium flavescens* - more records in Wales, Scotland, Yorkshire and N. Devon;
- *Fontinalis squamosa* - more records for N. Ireland and occurrences in Somerset;
- *Alisma lanceolatum* - a few more records for Wales;
- *Callitriche obtusangula* - more records for Scotland;
- *Lemna minuta* - more records particularly for the Trent catchment;
- *Ranunculus flammula* - distribution restricted to north and west of UK whereas published distribution is nation-wide;
- *Ranunculus penicillatus* subsp. *penicillatus* - some more records from Midlands and East of England than in the published distribution.

Regardless of the variations in the distributions of individual species, the national applicability of the MTR rests more on the geographical distribution of STR groups (Figure 38). The comparative distribution of STR groups is related to the expected distribution of water quality, with the exception of STR 2, whose distribution is particularly restricted compared to that of STR 3 and 1. The comparative absence of STR 2 species in Scotland, and to a lesser extent in north-west England, Wales and south-west England, may be a reflection of a bias within the STR 2 group towards species which prefer low energy river habitats. As each STR group should ideally include species representing a range of environmental conditions, this apparent bias should be investigated to refine the methodology and enhance its national applicability, particularly in Scotland.

The potential assignment of regional weightings to the STRs of individual species, was not addressed in detail as it was considered that a national system was more desirable than regional ones. The original choice of STR values and species was designed to include species found over a range of different geographical regions and water composition, and the species and STRs chosen have not been proven to be incorrect. Regional comparisons of the relationship between MTR and phosphate concentration were made but were inconclusive due to differences in phosphate ranges and other physico-chemical characteristics in different geographical areas (see 3.6.3).

Conclusion

MTR is applicable to use across the UK. Future work to refine the methodology should include further analysis of the distribution of STR 1–3 species in Scotland, and the possible need to select additional STR 2 species for high energy river habitats.

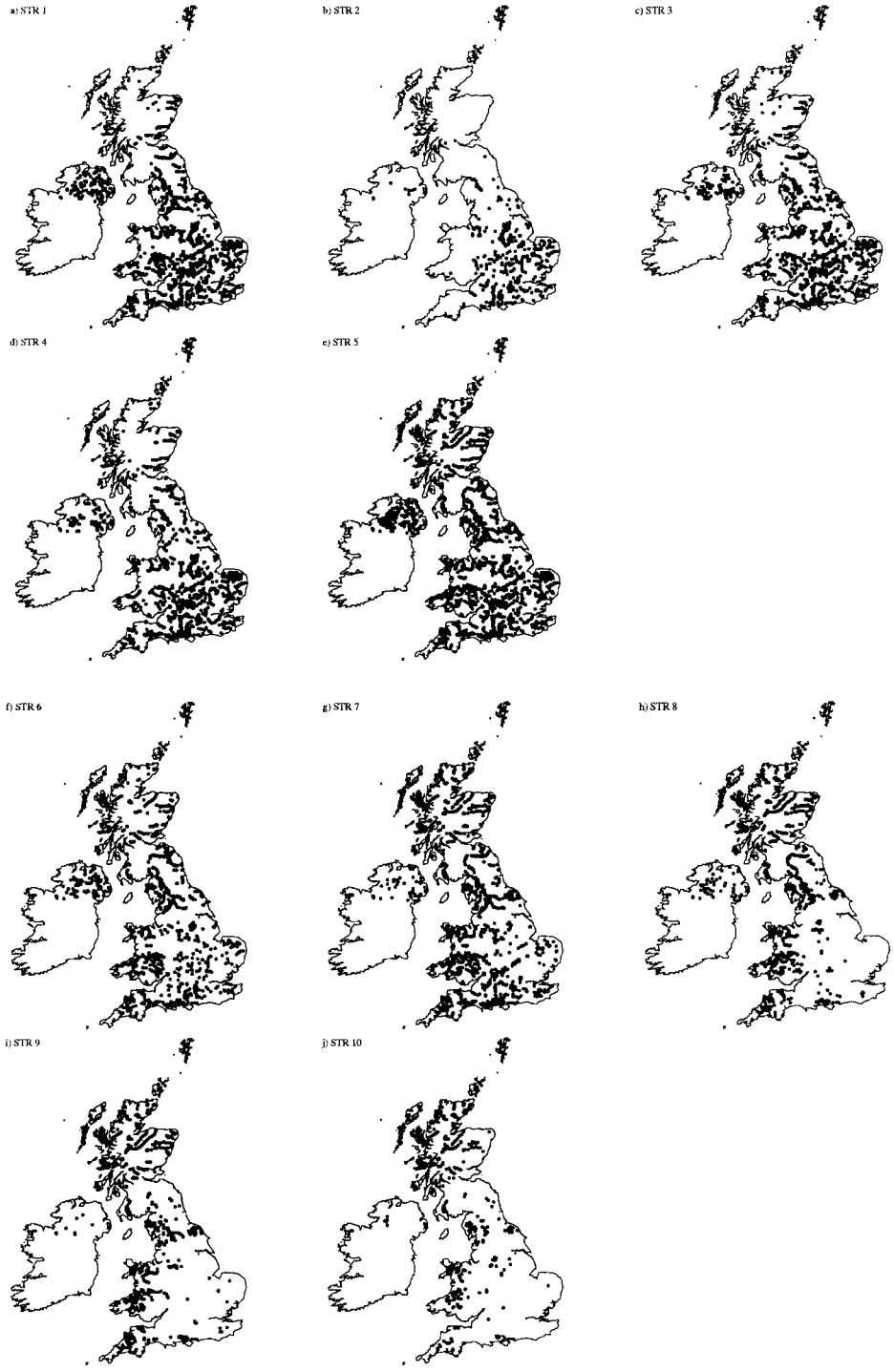


Figure 38. Distribution of STR groups 1-10 in England, Wales, Scotland and Northern Ireland for all datasets.

3.10 Species diversity and phosphate

The relationship between species diversity and phosphate concentration was examined to determine whether diversity could be used as a supplementary parameter to facilitate interpretation of MTR in terms of trophic status. Species diversity is one element of the 'balance of organisms' which may potentially be disturbed by eutrophication.

The relationship between the number of scoring species (hereafter termed the *diversity* for convenience) and mean annual phosphate concentration may be better described as an 'envelope' relationship with a maximum threshold or upper limit, rather than as a linear relationship (Figure 39a). The nature of this relationship indicates that although the phosphate concentration may determine to some extent the maximum diversity possible at a site, this potential may not be realised in many cases. Factors other than phosphate concentration which may limit the richness of the community structure include organic enrichment, silting, extreme shade, or the presence of trace metals. Changes in flow, for example due to global warming, may also influence the species richness.

In general, however, there is a decrease in the upper limit to diversity as phosphate increases (Figure 39a). If a logarithmic scale is used for annual mean phosphate concentration, then the expanded lower end of the scale, though there are less data, shows that there may be a slight decrease in this limit with decreasing phosphate concentration at concentrations of < 0.05 mg l^{-1} (Figure 39b). This is not unexpected and may be related either to an insufficiency of nutrients or the absence of a suitable type of habitat for the growth of scoring plants. High energy streams on rocks of low solubility, dominated by bryophytes which may not have all been assigned STR scores, may fall into this low-diversity low-nutrient category.

Of the eleven sites with more than 25 scoring species present and phosphate data available, nine are Conservation Rivers' surveys on the River Ouse between St. Neots and Huntingdon, Cambridgeshire, completed in 1991/92. The MTRs for these nine sites range from 28.4 to 35.0, however, illustrating that a high diversity does not always coincide with a high MTR.

Conclusions

On the basis of these results it is recommended that species diversity should only be used with extreme caution as a supplementary measure of the community response to trophic status. The following points should be borne in mind:

- a low or intermediate diversity may be recorded at any phosphate concentration;
- a high diversity is unlikely to be recorded at either very high concentrations or very low concentrations of phosphate;
- at any phosphate concentration, the 'envelope' shape of the relationship between diversity and phosphate concentration means that a wide range of diversities may be recorded.

Diversity should only be used to support the interpretation of MTR results in those situations where both a downstream change in diversity, or a temporal change at the same site, is very marked and the influence of factors other than a change in trophic status is deemed insignificant.

The relationship between the number of highlighted species and phosphate concentration is discussed later (4.6.4) in relation to the use of this parameter as a measure of confidence in the MTR.

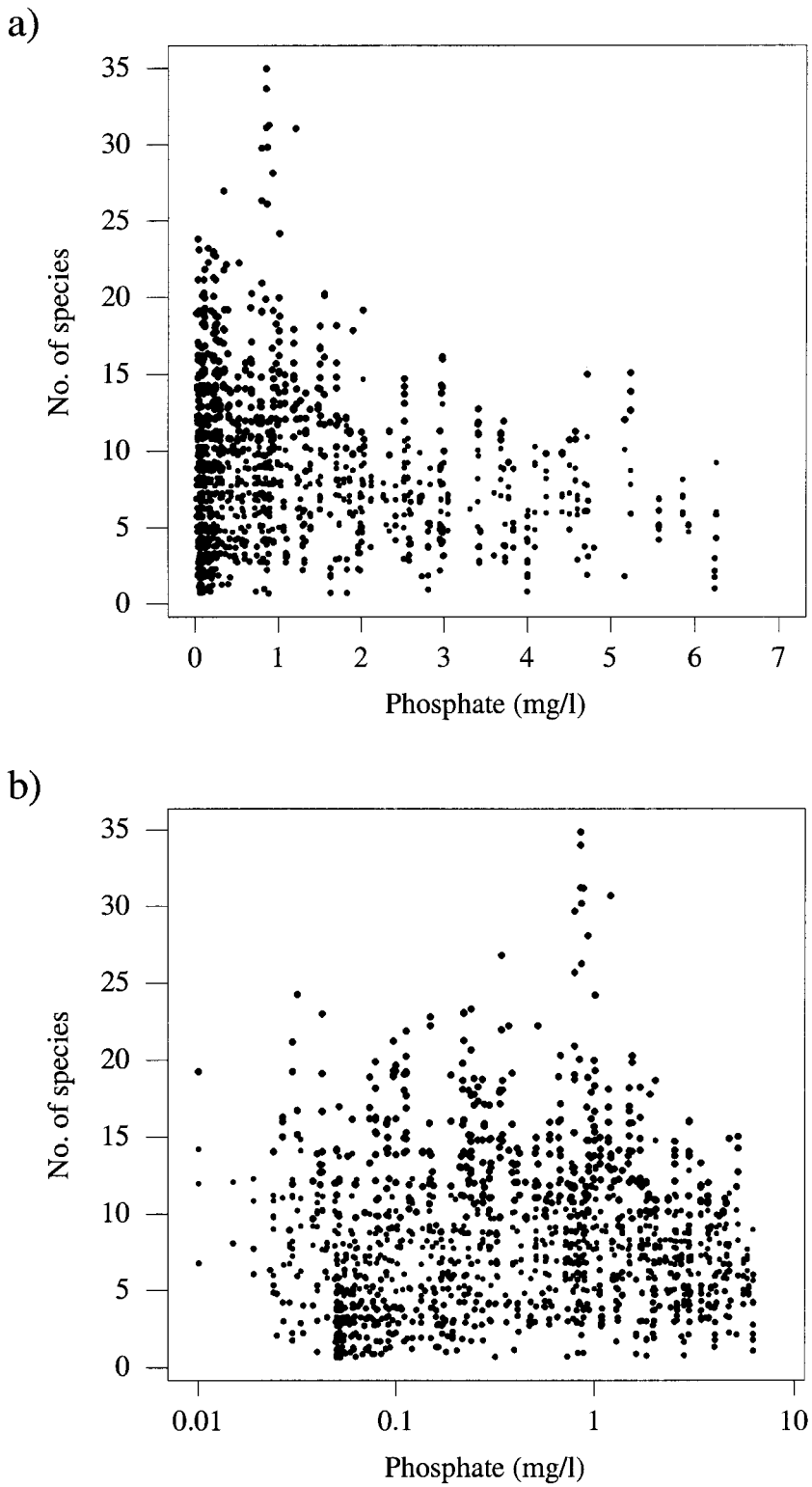


Figure 39. Relationship between number of scoring species and (a) phosphate and (b) the logarithm of phosphate concentration. Data from EA. IFE, DoE/IRTU and Conservation Rivers datasets (Conservation Rivers data from post 1985 surveys only). Macrophyte data matched with phosphate data either from the same site or from a site within 1 km. A small random movement has been added in relation to the Y-axis to separate points and hence to improve clarity. The high values centred on 1 mg/l and >25 species, relate to the Great Ouse River (see text).

3.11 Overall percentage cover and phosphate

The overall percentage cover of macrophytes includes both scoring and non-scoring species. The relationship between this overall cover and phosphate concentration was examined to determine whether the former could be used as a supplementary parameter to facilitate interpretation of MTR in terms of trophic status.

There is no obvious relationship between the overall percentage cover at a site and the phosphate concentration (Figure 40). Both high and low cover were recorded at both high and low phosphate concentrations. Variation in cover can be due to seasonal growth cycles and to the influence of factors other than trophic status, especially the degree of shade and water flow regime. The cover of floating plants, in particular, can be variable, and further variation may occur as a result of washout of plant material following high flows.

Conclusions

Despite increased plant growth being integral to eutrophication as defined by the UWWTD and Agency's Eutrophication Strategy, it would appear that this relates more to certain individual species (as shown in Figure 12 & 13) than to the overall cover of macrophytes at a site. Overall percentage cover, by itself, is of little use in terms of assessing trophic status. It is still recommended that overall percentage cover is recorded, however, for the following three reasons.

1. In many cases it acts as a further check on the percentage cover values given to individual scoring species and non-scoring species.
2. In some cases, nutrient enrichment may cause a change in overall percentage cover, and if so, is worthy of note.
3. Excessive growth of macrophytes, resulting in very high overall percentage cover values, can in itself result in problems to users of rivers, giving rise to complaints, and may impact on other river management functions. Where such complaints or impacts arise, it is important to establish whether the excessive growth is due to nutrient enrichment, or to other factors. Where it is due to nutrient enrichment, then this clearly constitutes an 'undesirable disturbance' in UWWTD terms.

Whether nutrient enrichment has caused the recorded change in overall percentage cover can be assessed by analysing the corresponding MTR and species list. Where there is a change in the overall percentage cover but no significant change in MTR, then the change in overall percentage cover is unlikely to be a symptom of a change in trophic status. Where a marked change in both overall percentage cover and MTR is recorded, and the changes to both can be attributed to the same key species, then the change in overall cover is likely to be a symptom of a change in trophic status. The most notable example of this relates to situations where the key species is *Cladophora.* If an increase in total percentage cover and a decrease in MTR is due mainly to an increase in percentage cover of *Cladophora*, then it may be deduced that the increase in overall percentage cover is a manifestation of eutrophication. The same may be true for *Enteromorpha*, with respect to nitrate enrichment. For some species, however, an increase in percentage cover may occur as nutrient concentrations decrease (Figure 12 & 13).

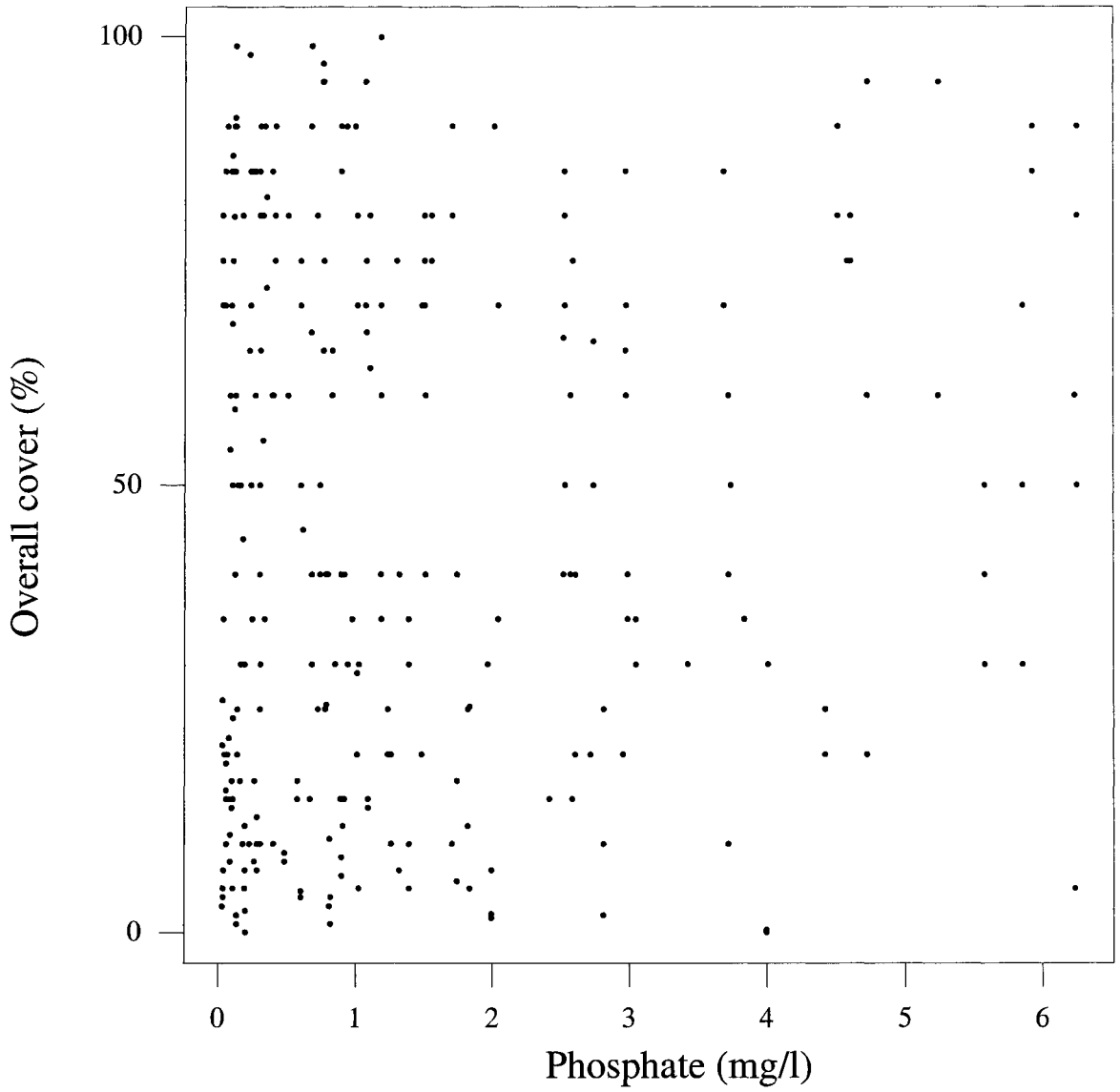


Figure 40. Relationship between overall percentage cover and phosphate concentration. Data from EA 1996 and DoE/IRTU datasets.

3.12 MTR and nutrient status: overall conclusions

In general, the performance of the MTR at assessing nutrient status, was found to be encouraging.

1. The aquatic macrophyte flora showed a spectrum of tolerances to nutrient enrichment. Although most species occurred over a broad nutrient range, differences in species' preferences were apparent. Species occurrence and/or abundance (percentage cover) increased with nutrient concentration for some species, but decreased for others.
2. The STRs were generally found to represent a spectrum of tolerances to nutrient enrichment, although further research is recommended to confirm the STRs for a few species.
3. MTR was found to decline with increasing phosphate and nitrate concentration and to be more significantly correlated with the logarithm of phosphate concentration, particularly at concentrations below 1 mg l^{-1} P or 10 mg l^{-1} N but even more at concentrations less than 0.5 mg l^{-1} P or 5 mg l^{-1} N.
4. Although the MTR cannot be used to distinguish between enrichment by phosphate and nitrate, the presence or abundance of certain individual species may potentially be useful in this respect. Phosphate is likely to the limiting factor to plant growth in most cases.
5. A general pattern of decreasing MTR downstream was often seen at QDs, although MTR may also vary along the length of a river in relation to diffuse nutrient inputs.
6. It was not possible to evaluate the performance of the MTR at assessing temporal changes in nutrient status, due to the current lack of adequate long-term data.
7. MTR is applicable to use across the UK. The distribution of STR 2 species may merit further investigation to improve the national applicability of the method.
8. In some cases, species diversity and/or overall percentage cover, may provide useful supplementary information to MTR results, although these parameters should not be used alone as indicators of trophic status.

This evidence supports the use of the MTR to assess trophic status, but implies that it may be most useful at detecting eutrophication impacts when the concentrations upstream of (or prior to) the nutrient input, are less than 1 mg l^{-1} phosphate-P or 10 mg l^{-1} nitrate-N. The methodology detects the symptoms of eutrophication as defined by the UWWTD and Eutrophication Strategy: increased abundance of species tolerant of nutrient enrichment results in a reduction in MTR score, reflecting a disturbance to the 'balance' of macrophyte species present.

4 VARIABILITY AND ERROR

4.1 Introduction

There are several potential sources of variability and/or error in the MTR methodology. They can be categorised as:

- the inherent variability of the method;
- inter-surveyor variation;
- natural background variation.

Together, these sources of variability determine the robustness of the MTR methodology as a tool to assess trophic status. If the level of variability can be defined, then confidence limits can be assigned to MTR scores. The various sources of variability are thus examined below, followed by an evaluation of the potential for confidence limits, and measures which can be taken to improve the quality of survey data.

4.2 General methods of assessing variability

The range and sources of variability were initially examined by resurveying MTR sites and comparing the results with the primary surveys. UWWTD sites were selected for resurvey if MTR scores had changed by greater than 50%, whether the comparable surveys were carried out in the same year or in different years. Of the sites selected 22 were resurveyed within 2 weeks of the comparable survey (hence, fulfilling the criteria for audit surveys). A further eleven surveys were carried out within two weeks to one month of the primary survey and as such have been discounted from the analysis but used with another 80 sites surveyed by the IFE in 1996 as comparison sites to help in interpretation of general errors in quality as necessary (ie they overlap with Agency surveys at the same sites in the same survey season). Agency surveys undertaken in 1996 were also subject to internal audit. Ten percent of IFE surveys were subjected to internal audit resurvey of the same site by other surveyors on the same day or within a few days.

The results were analysed and those showing marked differences were examined on a case-by-case basis to establish possible causes of the differences. The possible sources of inconsistency investigated were: difficulties in interpretation of the methodology; difficulties in application of the methodology (eg difficulties in taxonomy and in deciding which specimens are 'in' and 'out' of the channel); or inability to meet the re-survey criteria (eg errors in the survey location). A marked difference in MTR was taken as being a numerical difference of 3 for the purpose of this analysis, as this approximately equated to the median difference between primary and audit surveys (see 4.4). In addition, analysis of large same-year and year-to-year differences in MTR results at the same site where possible 'substitution' of difficult taxa may have taken place, was carried out.

These data, results and conclusions are incorporated in the appropriate sections on variability and variation.

4.3 Inherent variability - the repeatability of the MTR method

The level of inherent variability in the method determines its repeatability as defined in 2.2.3. Ideally, it would be evaluated by analysis of data from surveys where the same surveyor undertakes a repeat survey of a site within a short space of time. It would be difficult to obtain truly objective results, however, as the surveyor is likely to retain a memory of what species were found at a site, and possibly even where they were found within a survey length. Although such surveys and analysis were not undertaken for the present study, it can be reasonably assumed that the inherent variability of the method is relatively low. The main justification for this assumption is that all the survey area is surveyed: no sub-sample is taken as a representative of the whole, and so there is no sample error to consider as such. The only variability is likely to be human error in terms of the estimation of percentage cover of macrophyte species, mis-identification of the plant species or mis-application of the method. This can only be reduced by training and by rigorous and consistent application of one of the recommended means of estimating the area covered by plants.

The degree of variation in MTR that is likely to occur due to variation in the estimates of percentage cover, or to misidentification of plant species, was determined by a mathematical exercise. A hypothetical species list was constructed which used the rounded mid-range MTR of 50 and ten species. All species in the test list scored an STR of 5, but were 'present' at different abundances: one dominant plant species at cover value 8, one co-dominant at cover value 5, and eight minor species each of cover value 2. One of the STR or cover values was than changed at random to an alternative adjacent value (eg one species from STR 5 to 6, or from cover value 2 to 1), and the MTR recalculated. This process was repeated many times using different combinations of random changes: changes of ± 1 STR for up to five species; changes of ± 1 cover value for up to five species; and, changes in both STR and cover value for up to five species. This gave a hypothetical range of variations from ± 1.2 MTR units (range of ± 2.8 MTR units) when using one random alternative STR of +1 or -1 to any one species, to ± 2.5 (range of ±7.5) MTR units when using five random variations of alternative cover and species. Varying the cover of species (by one class) with the same STR does not make any difference but varying the STR value of the dominant species may make a significant change in MTR.

Conclusion

The MTR methodology is assumed to be repeatable, provided the surveyor is careful, fully trained and estimates the percentage cover of plants in a rigorous and consistent manner.

4.4 Inter-surveyor variation - the reproducibility of the MTR method

The level of inter-surveyor variation determines the reproducibility of the methodology, as defined in 2.2.3. It can be evaluated by analysis of results (46) from quality assurance audit surveys, where repeat surveys are undertaken by a different surveyor(s) within two weeks of the primary survey. The word 'difference' is used in preference to 'error' as a resurvey is as likely to be different from the 'real' situation as the initial survey.

The results from audit surveys showed that 52% differed from the primary survey by less than 3 MTR units and 66% by less than 4 units. In percentage terms, 46% of audit surveys differed by less than 10% of the primary survey MTR and 68% by less than 15% (Figure 41a&b). In normal statistical terms, the distribution was insignificantly different from normal having a mean difference in MTR of 0.9 with 95% confidence limits of ± 13; differences when analysed as an absolute difference ('one-sided') confirmed this range (Figure 41c&d). The 48% of audit surveys showing a 'marked difference' (see 4.2) differed due to one or more factors, including: differences in the precise location of the survey length; differences between surveyors' recording of sparsely distributed species; differences in estimation of species-cover; differences in identification of *Ranunculus* species; and/or the (presumed) washout of species from the site (eg algae such as *Enteromorpha* or *Cladophora).* All but the last factor can be deemed to be inter-surveyor variation and elements of the reproducibility of the MTR. The latter factor, washout of species, may occur and be a factor in inter-surveyor differences but is more an element of the natural temporal variation in the MTR (4.5.2). Although not audit surveys, analysis of surveys carried out within the same year but at different times of the season (same-year surveys: see 4.5.2), revealed another potential source of inter-surveyor variation: the mis-identification of moss species.

To qualify as an audit survey, a repeat survey must be carried out within two weeks of the primary survey. Given this short space of time, it is unlikely that the differences noted between primary and audit surveys were due to a real change in the trophic status of the water. This is for two reasons: it is unlikely that the underlying phosphate concentration (not peak concentration) will have changed significantly in the period between the primary and the audit survey; and even if it had, it is considered unlikely that the macrophyte community would respond so quickly (although this may not be true for species such as *Cladophora).*

Conclusions

The audit results, although based on a relatively small sample of surveys, indicate that the MTR system is not precisely reproducible, but variation is low with half of surveys differing by less than 3 MTR units or about 10% MTR and two thirds differing by less than 4 MTR units (or 15% MTR). The main sources of inter-surveyor variation are the estimation of macrophyte cover, the ability of surveyors to find species with only a sparse cover, and the mis-identification of *Ranunculus* and moss species. All can be reduced by appropriate training and quality assurance measures.

Error in determining the precise location of survey length, although not evaluated in the analysis of audit survey data, is another potential source of inter-surveyor variation. It may be minimised by improved location maps indicating more permanent structures, such as fences or hedge lines, plus useful detail such as parking and access (this can be incorporated into the main survey form: see example in Appendix 4 as used in IFE surveying for this project).

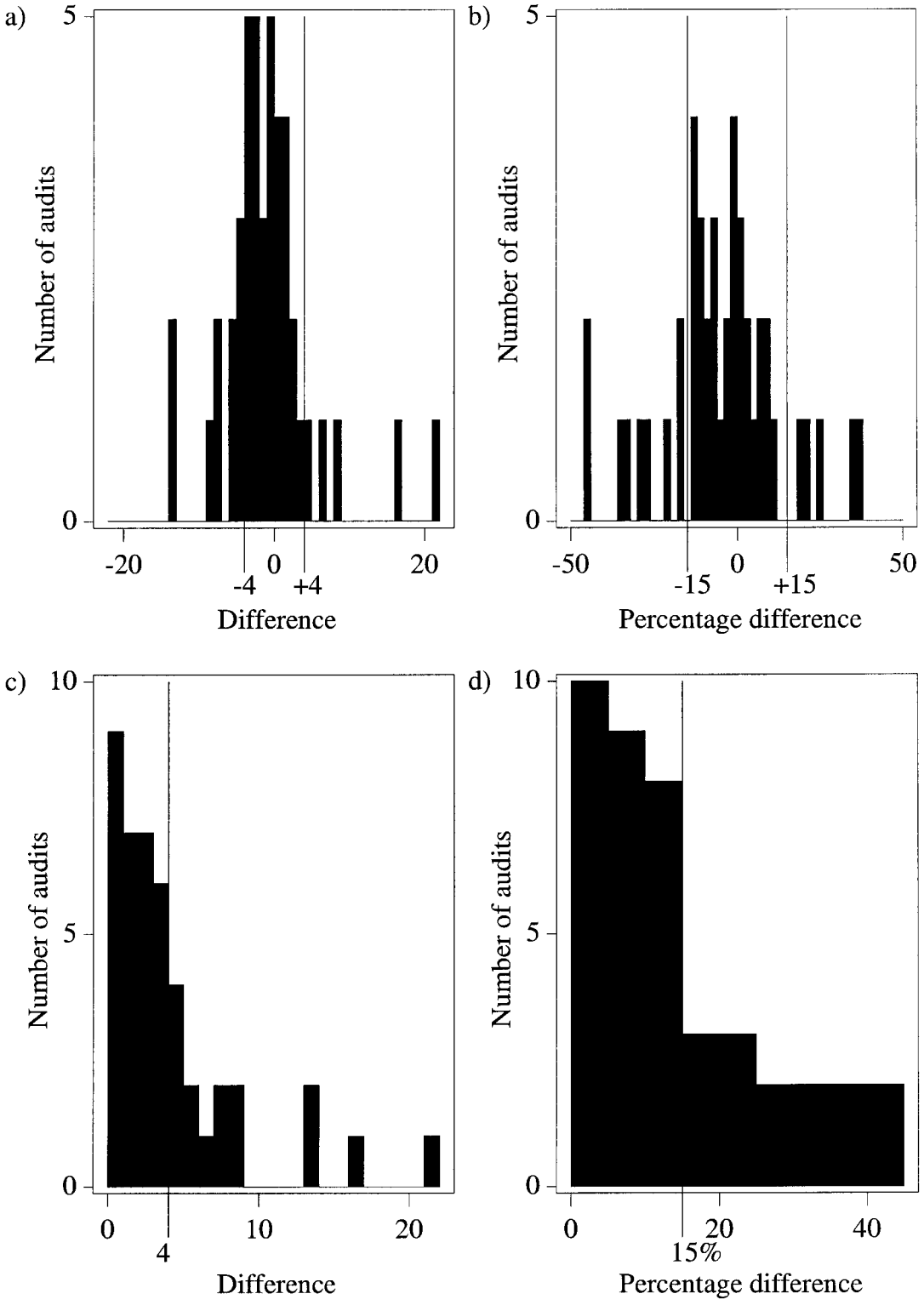


Figure 41. The distribution of differences in MTR between primary and audit surveys carried out in 1996 a) as MTR units (mean -0.9 ± SD of 6.2) and b) as a percentage (mean –4.7 ± SD of 17.8). The distribution of absolute (ie one-sided) differences in MTR, between primary and audit surveys c) as MTR units and d) as a percentage. The vertical lines represent the proposed level for the limit of confidence in the survey. Data are not all presented as the axes are slightly shortened for clarity. Data from EA and IFE.

4.5 Natural background variation

4.5.1 Introduction

Natural background variation may be manifest on either a temporal or spatial scale. Examples on a temporal scale may include variation during the survey season or between years, either as a result of the natural growth patterns of plants and/or as a result of temporal perturbations in the plants' environment. Examples on a spatial scale may include variation between sites as a result of differences in either the physical and/or chemical characteristics of the sites, and/or downstream variation along rivers as a result of natural colonisation and growth patterns of plants. The level of variability arising from natural background variation is not easy to evaluate, however, nor is it easy to separate the influence of the different sources of variability.

4.5.2 Temporal variation

Variation in the MTR during the survey season was examined by deriving the frequency distribution of the percentage difference in MTR value recorded at surveys undertaken at the same site, in the same year, but at different times within the survey season ('same-year' differences, Figure 42). The same-year difference from early to late in the survey season is, on average, an increase of +7.5% or 0.66 ± 14.5 MTR units over the range from 10-73.6. This increase is extremely unlikely to reflect any real improvement in trophic status as the time between surveys (~110 days) is considered too short for the plant community to respond. The most likely explanation is due to differences in the relative cover of species of differing STR as a result of differing intra-seasonal growth patterns and not species change. In some cases, however, wash-out of species from the site may occur, especially after a temporal perturbation such as a period of high flow or a spate (see presentation of audit survey results above, under discussion of reproducibility). Although MTR changed between surveys in the same season, no statistically significant increase or decrease could be established between the change and the number of days between surveys.

To help set the variation described above for same-year differences into context, this variation is compared with the corresponding distribution of percentage differences between all the physically comparable sites (ie with a suffix of confidence of either I or II) upstream and downstream of QDs, surveyed at the same time within the survey season. The two distributions were found to be statistically similar. The same-year mean difference of 7.5% is greater than the upstream-downstream mean difference of 2.6% (or 1.59 ± 14.2 MTR units in the range of 10-80 at upstream sites and 10-75 at downstream sites); but the variances are similar. The distribution of the upstream-downstream differences, however, are slightly skewed to one-side compared to the almost perfect normal distribution of the same-year differences, demonstrating that there is likely to be a difference between MTRs upstream and downstream of QDs.

Natural, background variation may also occur from year to year. The level of this scale of variation was difficult to evaluate, however, as differences between survey years were found mainly to relate to surveyor error: either high MTRs in 1994, due mostly to differences in identification of *Ranunculus* species which were not required to be determined to species in 1994; or, the number of species recorded per survey which increased, although not significantly, from an average of 7 in 1994 to 8.7 in 1996. This increase in species recorded was presumably due to the greater experience of surveyors, the greater awareness of areas of

possible error, and possibly the changes to the standard species listed on the recording form, although the total number of species recorded in UWWTD surveys decreased from 199 in 1994 to 164 in 1996. Reduced MTR scores during the period may reflect real changes in water quality or may be related to low flow conditions in 1995 and 1996.

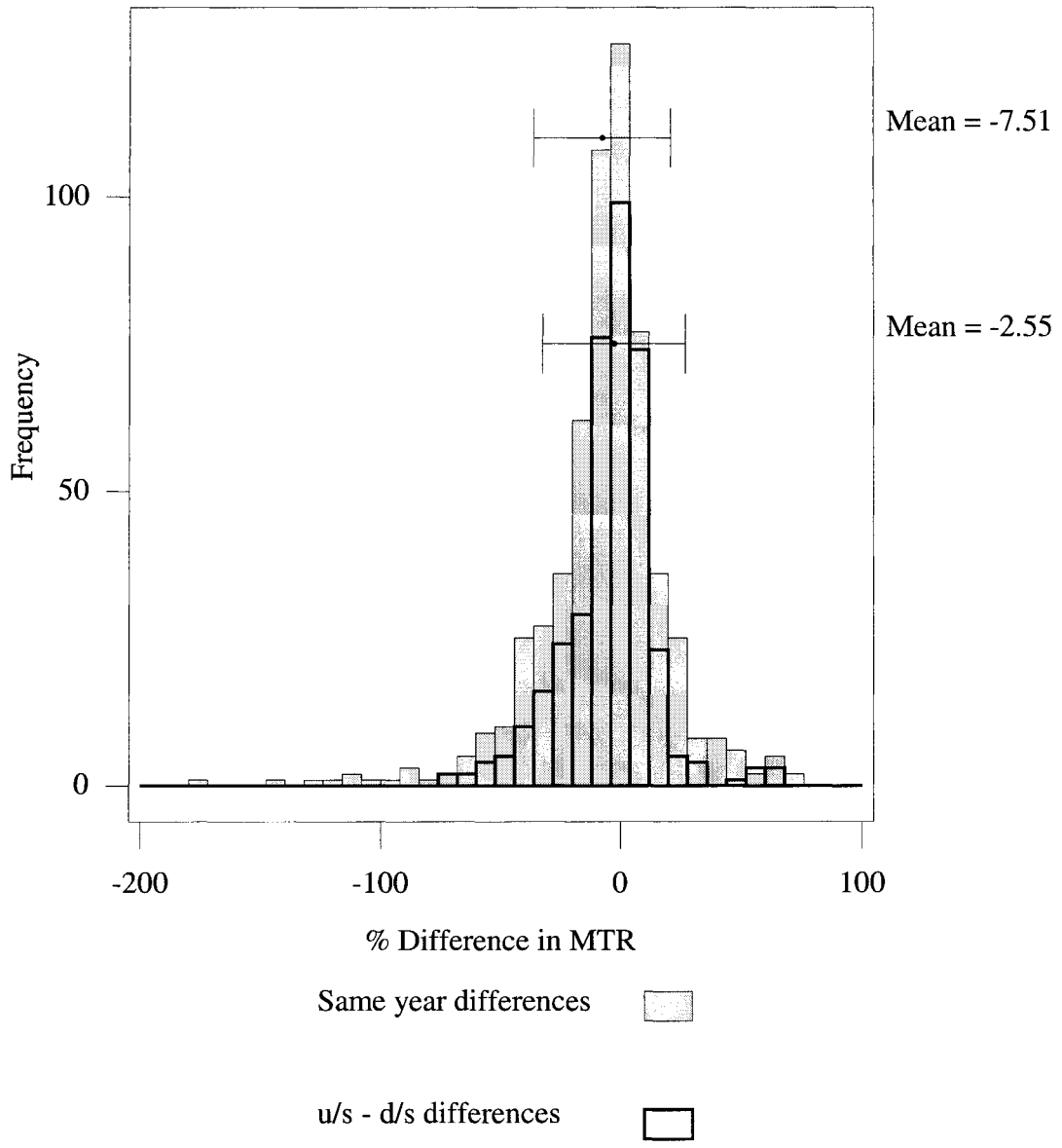


Figure 42. Frequency distribution of the percentage difference between MTR scores, contrasting same-year differences (early - late season) and upstream-to-downstream differences at sites with good physical comparability (ie suffix of confidence of I or II). The population means (.) and standard deviations (|—|) for each frequency distribution are given. Data from EA & IFE datasets.

4.5.3 Sensitivity of the MTR to the physical characteristics of the site

The sensitivity of the MTR to the physical characteristics of the site was evaluated by examining the relationship between the MTR and a variety of physical factors for which sufficient data were available (Appendix 2). The analysis provides an insight into the factors which may influence the performance of the MTR at assessing trophic status.

Significant standard correlations were found between MTR and physical variables (bold type in Table 6). Factors shown to be correlated with MTR, to varying degrees, included: mean depth, cross-sectional area, flow category, mean substrate size (as expressed by phi), slope, altitude of source, and solid geology (Figure 43). Cautious interpretation of results is required not only because of the limitations described below, but also because of co-correlations. For example, MTR is negatively correlated with phi and positively correlated with altitude. Thus, as larger phi numbers mean smaller substrates, the tentative conclusion may be reached that MTR is greater at sites with larger substrates and likely to be lower at sites with finer substrates. Results from the related RHS project demonstrate that upland sites frequently have larger substrates, but also that lowland sites are more often modified and have finer substrates (Raven et al 1998). Modification or site altitude may therefore affect MTR rather than substrate size. Similarly, MTR is less in deeper rivers, but these rivers are also more likely to be in the urban, lowland areas with higher levels of nutrient enrichment.

Little correlation was found between MTR and shade (Figure 44), although the pre-selection criteria for sites (to avoid shaded sites) means that it is not possible to analyse fully the influence of shade on the performance of MTR. The altitude of the site, the river width or the distance from source also show little correlation. The latter seems consistent with the lack of downstream trends in MTR particularly apparent on two of the rivers selected for analysis in section 2.3.2 (Figure 37, see 3.8.1).

In further analysis, solid or deep geology was classified into eight types, using a combination of sub-groups from those formerly used by the Nature Conservancy Council in plant classification systems. Sub-division of the phosphate and MTR data by these groups improved the strength of the relationship between MTR and phosphate for sites on types of hard limestone geology, implying that the MTR may perform better at assessing trophic status at such sites (Table 7).

The validity of this analysis was limited by the incomplete nature of the various datasets and the non-random nature of the sampling strategies. The database was complete with respect to the Agency and IFE datasets, but not for the other datasets (Table 1).

Table 6. Pearson correlation coefficients between MTR and some physical factors for which sufficient data were available for analysis. Parameters are those for which data were available for >50% of sites (Appendix 2). It was noted that there is a significant correlation between eastings and northings and MTR score, with highest scores in the west and north of the UK.

	Physical factor	Correlation coefficient	Dataset		
			EA/IFE	DoE/IRTU	CR.
MTR with	Mean Width	-0.061	•		•
	Mean Depth	**-0.286** **	•		•
	Cross Sectional Area	**-0.201** *	•		•
	Mean substrate size (phi)	**-0.374** ***	•		•
	% Un-shaded	-0.069	•		
	Site Altitude	0.093	•		
	Slope	**0.241** *	•		
	Altitude of source	**0.307** **	•		
	Distance to source	0.021	•		
	Flow category	**0.206** *	•		
	Solid geology coded (classifications below)	**-0.225** *	•		
	NGR Easting	**-0.519** ***	•	•	•
	NGR Northing	**0.420** ***	•	•	•

Confidence levels: * > 95%; ** 99%; *** 99.9%.

Table 7. Correlation coefficient of MTR with the logarithm of phosphate concentration, in subsets of geology. Data from EA, IFE and Conservation Rivers datasets (CR data were matched with chemical data at sites within 5 km).

NCC Geology Code					
3	4	5	6	7	8
Clay	Shale	Sand	Chalk	Hard Limestone	Hard Rocks
-0.493 ***	**-0.502** ***	**-0.537** ***	**-0.530** ***	**-0.815** ***	**-0.637** ***

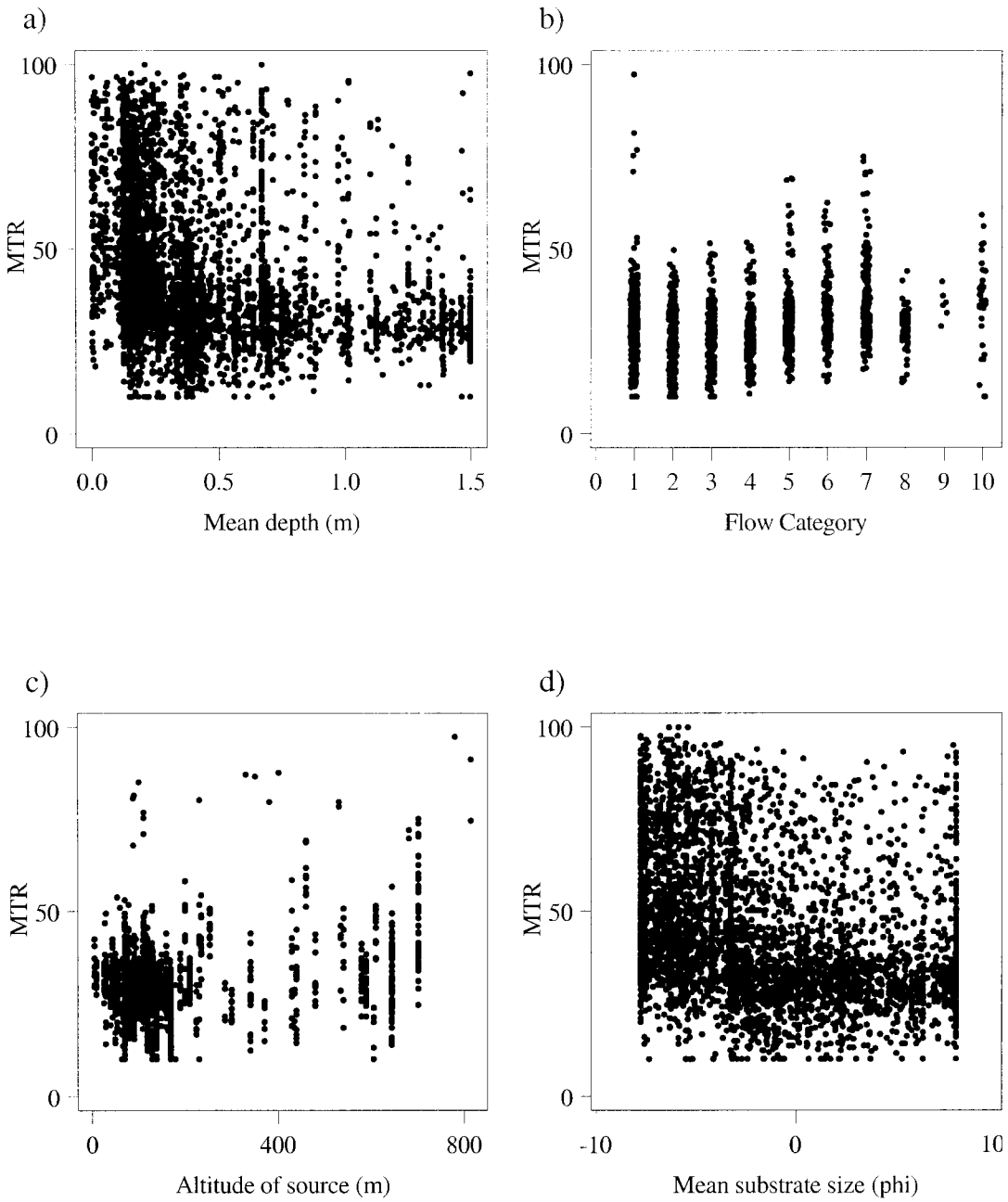


Figure 43. The relationship between MTR and a) estimated mean depth in metres, b) flow category, c) altitude of the source in metres and d) mean substrate size on phi scale (see Appendix 2) for EA, IFE and Conservation Rivers data. A small random movement has been added to the X- and y-axis position to separate points in order to aid clarity.

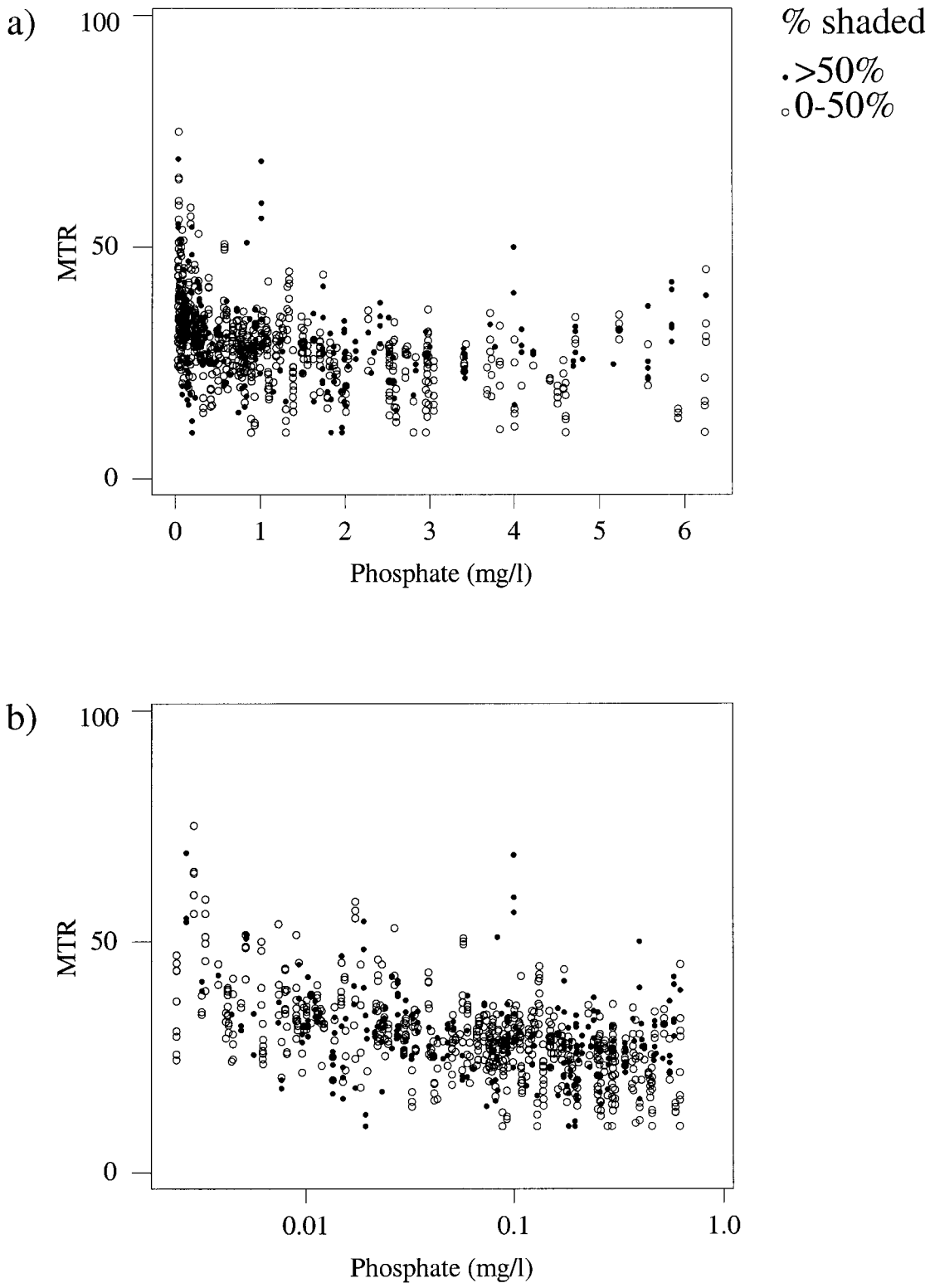


Figure 44. The relationship between MTR and the concentration of a) phosphate and b) its logarithm, for the range of shade values (see key) recorded at EA, IFE and Conservation Rivers survey sites. Macrophyte data matched with phosphate data either from the same site or from a site within 1 km. Conservation Rivers surveys for the years post-1985.

4.5.4 Sensitivity of the MTR to the chemical characteristics of the site

The sensitivity of the MTR to the chemical characteristics of the site was evaluated by examining the relationship between the MTR and a variety of chemical determinands for which sufficient data were available. Interpretation of the results was limited by the incompleteness of the various datasets and possible co-correlation between factors, as with the analysis of physical characteristics.

The results show that MTR is correlated most strongly with the concentration of those nutrients associated with trophic status - phosphate and nitrate (or their logarithms: Table 8); the use of the logarithm of phosphate particularly enhances the relationship. This supports the use of the MTR as a tool to assess trophic status. There is evidence, however, that other chemical determinands may influence the MTR to a lesser degree. No significant correlation was found between MTR and ammonia or suspended solids.

Table 8. Pearson correlation coefficients between MTR and the chemical determinands for which sufficient data were available. EA, IFE and Conservation Rivers survey sites. MTR data matched with chemical data from the same site or from a site within 1km. Conservation Rivers for the years post-1985.

	Chemical determinand	Correlation coefficient	Dataset		
			EA/IFE	IRTU	Cons
MTR with	Phosphate	**-0.384** ***	•	•	•
	Log. of phosphate	**-0.549** ***	•	•	•
	Nitrate	**-0.423** ***	•	•	•
	Log. of nitrate	**-0.448** ***	•	•	•
	pH	-.180	•	•	
	Alkalinity	**-0.405** ***	•	•	
	Conductivity	**-0.294** **	•		
	Chloride	**-0.315** **	•	•	
	Ammonia	-0.178	•		
	BOD	**-0.272** **	•		
	Suspended solids	-0.016	•		

Confidence levels: ** > 99%; *** > 99.9%.
Alkalinity and conductivity, 0.355**

4.6 Confidence Limits

4.6.1 Introduction

At this stage of the method development, it is not feasible to assign statistical confidence limits to an MTR score. Sources of human error, whether they be inter-surveyor variation or mis-application of the method, can be reduced by training and appropriate quality assurance measures (see section 4.7). In recognition of the various sources of natural background variation, however, Holmes (1996) proposed the use of three suffixes of confidence: relating to the confidence in the survey, the confidence in the comparability between survey sites, and the confidence in the MTR score.

4.6.2 Assigning a measure of confidence in the survey

This measure of confidence is assigned according to how accurately the surveyor feels the survey reflects the typical situation which would prevail at the site (Holmes 1996; Environment Agency 1996). Examples given for situations where the survey may have been hampered and perhaps rendered meaningless, include: recent river management, extreme flooding events, poor survey conditions, water clarity, excessive blanketing algae of floating vegetation obscuring the view or smothering other vegetation. Surveyors are asked to score on a scale of A to C the degree to which such factors may have distorted their survey findings:

A - data not affected, or any effect limited to less than 25% of the site;
B - the accuracy of records in 25-50% of the site influenced to a considerable degree;
C - the accuracy of records in >50% of the site influenced to a considerable degree.

Surveyors are asked to note the factor(s) which potentially distorted the accuracy of the survey.

Although the effect of such events cannot be demonstrated in the present study, the use of this measure of confidence is considered to be valid and helpful. As currently described, however, it can lead to inconsistent application between surveyors. This arises, firstly, from the possible mis-application of 'typical' to mean typical of the reach (ie typical in a spatial context) rather than the intended meaning of typical for the site (ie in a temporal context). The second area of inconsistency relates to the two distinct sources of distortion of survey findings: events which may have caused recent, real changes to the macrophyte community so that it does not reflect the prevailing conditions; and events which reduce the accuracy of the survey. River management operations and flood events fall into the former category; poor survey conditions and plants obscuring other plants fall into the latter. Both categories are included in the measure of confidence. Inconsistency may be reduced by training and by re-wording of the guidance on how to use this measure of confidence.

4.6.3 Assigning a measure of confidence of comparability between pairs of survey sites

When undertaking surveys to assess downstream changes in trophic status, surveyors are currently required to assign a measure of confidence in the physical comparability of the survey lengths being compared (Holmes 1996, Environment Agency 1996). They are asked to identify, on a scale of I to III, how comparable the sites are:

I - if substrata, velocity, depth and degree of shade are broadly comparable for more than 75% of the site;
II - if comparability is limited to 50-75%;
III - if less than 50% is comparable.

Given the potential influence of the physical habitat (see 4.5.3), it is recommended that such a measure of confidence for the physical comparability of survey lengths is retained in the methodology. The factors to be considered when assigning this measure of confidence should be: **depth, substrata, habitats (flow features), shading,** (as in Holmes (1996); **and in** addition, **width, water clarity** and **bed stability**. As the evidence for the present study suggests that some of these factors, singly or in some combinations, may have a stronger influence on MTR than others, it may help interpretation of results if the surveyor notes on the field sheet the factor(s) which are not comparable between sites. Extra care must be exercised in choosing sampling sites to minimise the physical differences: for example, a single factor difference, such as the mismatch of silty and cobble dominated-sites, may have an influence on the MTR.

A change is recommended to the precise means of assigning the three confidence levels. At present, surveyors must estimate the proportion of a survey length which is similar for all the physical factors being assessed. In practice, this is difficult to apply, which reduces its usefulness and may be a source of inter-surveyor variation. It is also not consistent with the method chosen for the comparable suffix of confidence for DQI surveys (Kelly 1996b). In the latter, surveyors must assess each physical factor separately, judge whether or not it is similar between the two survey lengths, and then assign the measure of confidence according to the number of factors which are similar. This is considered to be easier to apply in practice, and hence should lead to greater accuracy and less inter-surveyor variation. As it is not known which of these two approaches provides the most useful measure of confidence in ecological terms, it is recommended that the approach used for MTR surveys be changed in line with that used for DQI surveys.

The new approach would thus require the surveyor to consider the following seven physical characteristics - width, depth, substrata, habitats, shading, water clarity and bed stability - and assign one of the following categories:

I - 5 or more of the physical characteristics are similar for more than 75% of the site for each pair of survey lengths;
II - 3 or 4 of these characteristics are similar for more than 75% of the site for each pair of survey lengths;
III - 2 or less of these characteristics are similar for more than 75% of the site for each pair of survey lengths.

4.6.4 Assigning a measure of confidence in the MTR - the number of highlighted species

Prior to this study, surveyors were required to assign a measure of confidence with which the MTR data can be considered, according to the number of 'highlighted' species recorded (Holmes 1996, Environment Agency 1996). One of the following categories is assigned as a 'suffix of confidence':

a - > 8 highlighted taxa are present;
b - 5-8 highlighted taxa are present;
c - < 5 highlighted taxa are present.

'Highlighted' species are those scoring species which were considered during the development of the MTR, to be more reliable indicators of trophic status. The rationale for using the number of highlighted species as a measure of confidence in the MTR score is that the greater the number of reliable indicators species present, the greater is the likelihood of increased reliability of the resulting MTR. For the number of highlighted species to be recommended as a measure of confidence this premise of reliability must be confirmed, the MTR-phosphate relationship shown to be the same regardless of the suffix of confidence based on highlighted species and the three suffixes shown to be independent of the MTR score.

Restricting the relationship between phosphate concentration and MTR by calculating MTR solely using highlighted species present rather than from all scoring species, does not increase the significance of the correlation (Figure 45, cp. 3.6.1 & Figure 28). When MTRs are separated into three groups according to the number of highlighted species and a separate linear regression line fitted simultaneously to each of the three groups and compared with the MTR-phosphate relationship shown for all sites (Figure 28), the proportion of explained variation in MTR at all sites increases, but only by 0.9% from 30.2% to 31.1%. This indicates that, in practical terms, the number of highlighted species present does not affect the general relationship between MTR and phosphate. However, it may improve the precision of this relationship, as the variability around the mean increases with greater numbers of highlighted species. The residual standard deviation is 18.9 MTR units when < 5 species are present, 12.1 with 5-8 species and 7.8 for > 8 species (Figure 46a, b, c respectively). This increase in precision could simply, or mostly, be due to the total number of scoring species present rather than their highlighted status.

There is a similar inconclusive relationship between STR and phosphate concentration when divided into highlighted and non-highlighted species (Figure 47a&b cp Figure 16).

The relationship between the number of scoring species and phosphate concentration, already demonstrated in Figure 39, is mirrored when considering only highlighted species (Figure 48). When the number of scoring species are categorised according to the three suffixes of confidence (<5, 5-8, >8), the data points in each of the three categories occur across the whole phosphate range with no constant pattern, indicating that the categories are independent of phosphate concentration (Figure 49a-c).

The number of highlighted species may, however, not be *fully* independent of the MTR score (Figure 50d). The greatest numbers of highlighted species are found predominantly at sites with MTR values between 25 and 50, but, whilst decreasing at higher and lower MTR, they are still well represented across the majority of the MTR range. This pattern may be a reflection, in part, of the relationship between the total number of highlighted species and the phosphate concentration (Figure 48). However, the restriction on numbers of highlighted species at very high MTRs may also be an artefact of the MTR derivation itself as there are a restricted number of species assigned the higher STRs (for example, only seven highlighted STR 10 species). This means that the theoretical maximum attainable MTR, although 100 when there are 11 or fewer species present, decreases with increasing number of species, to a maximum attainable of 96 when 30 species are present. The mean MTR recorded at sites in each of the three categories used in the suffix of confidence (sites with <5, 5-8, >8 highlighted species) were not found to be significantly different (Figure 50 a-c).

In conclusion, there is little evidence that highlighted species are more reliable indicators of the trophic status, although the MTR score may be more reliable when more highlighted species are present probably because of the greater total number of scoring species. The number of highlighted species is not completely independent from the MTR score: low numbers can be found at any MTR score, but high numbers of highlighted species (> 8) are less likely to be found at either very low or very high MTR scores, eg scores less than 20 or more than 90. It is, however, recommended that the use of this suffix of confidence be continued as an interim measure, noting that that the achievement of a suffix of 'a' or 'b' will certainly lend confidence to the results; but achievement of a 'c' suffix may not necessarily mean that the result is inaccurate. Emphasis should always be placed on obtaining information from as many sources as possible and on drawing conclusions using the balance of evidence available.

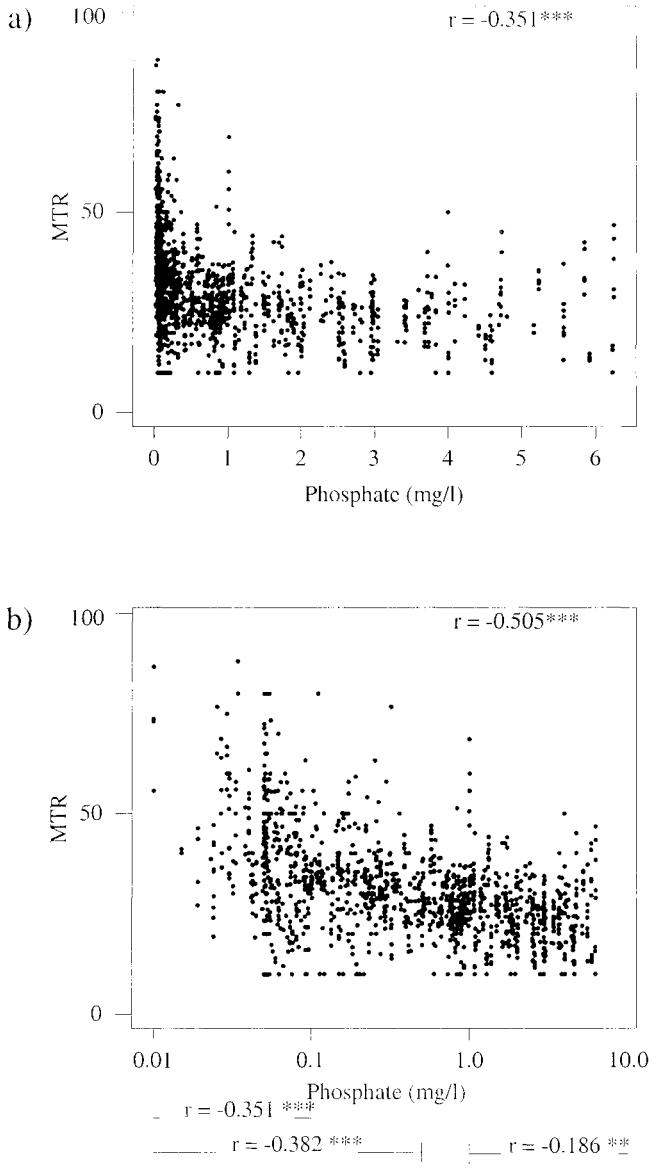


Figure 45. Relationship between MTR and the concentration of a) phosphate and b) its logarithm, calculated using only the highlighted species. Data for EA, IFE, DoE/IRTU and Conservation Rivers. Macrophyte data matched with phosphate data either from the same site or from a site within 1km.

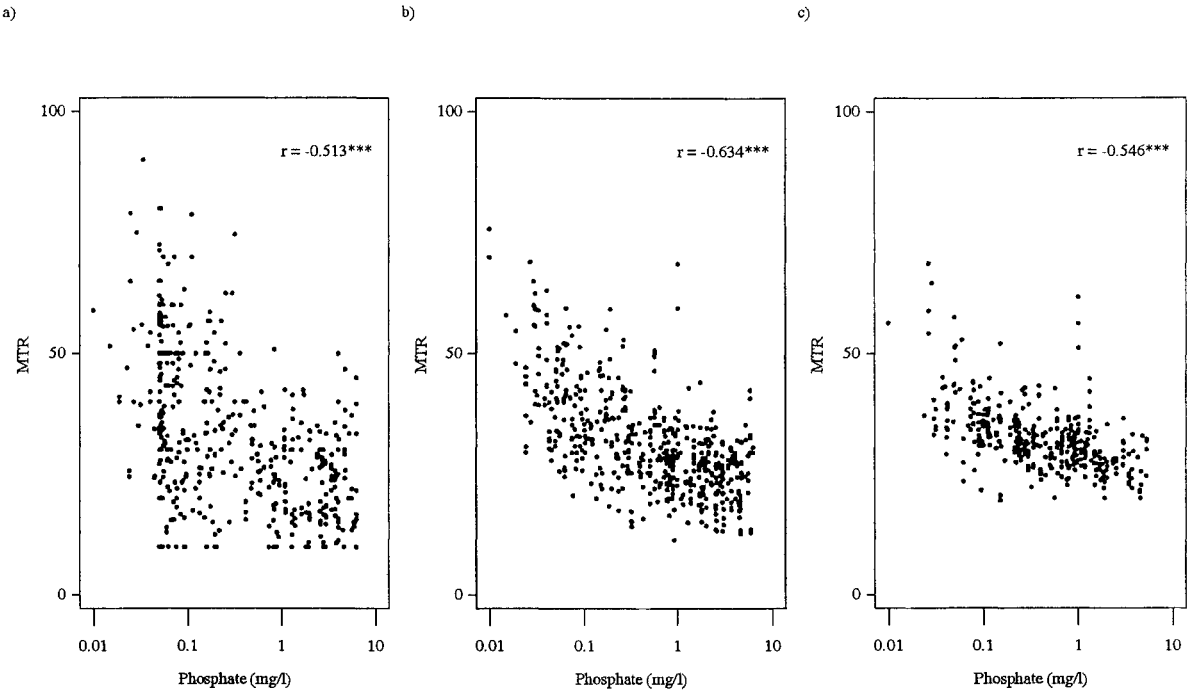


Figure 46. Relationship between MTR and the logarithm of phosphate concentration for sites with a) less than 5, b) 5-8 and c) >8 highlighted species. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets post 1985. MTR data matched with phosphate data either from the same site or from a site within 1km.

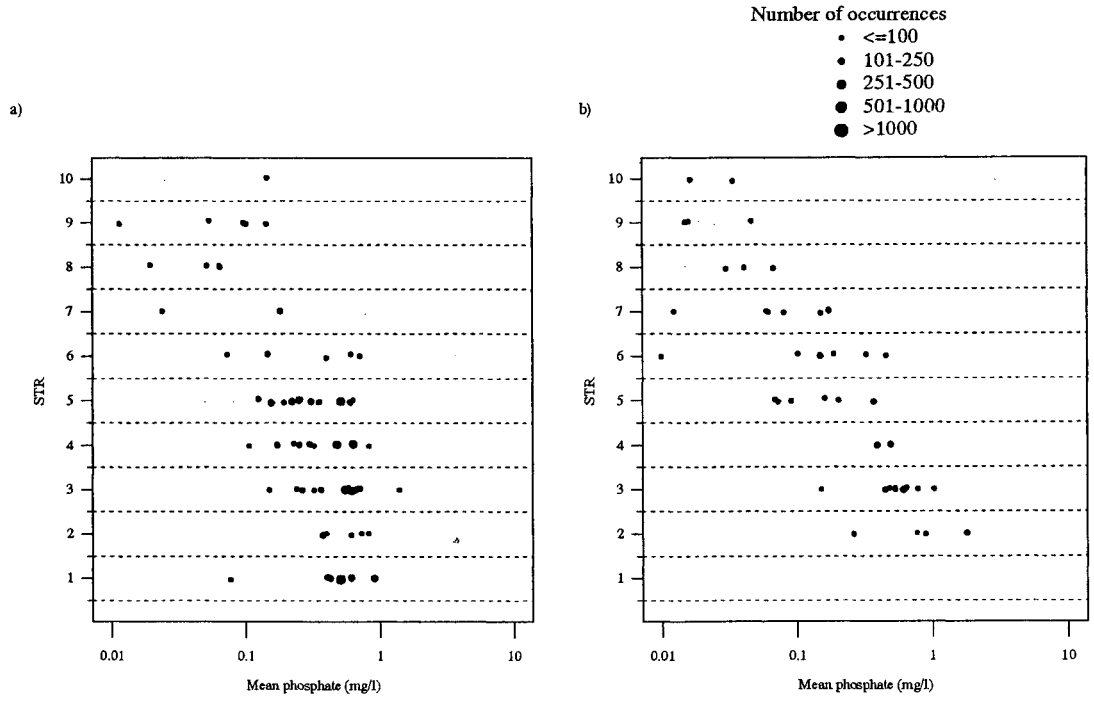


Figure 47. Relationship between STR for a) highlighted and b) non highlighted species and the logarithm of the mean phosphate concentration. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets post-1985. Macrophyte data matched with phosphate data either from the same site or from a site within 1km. A small random movement has been added to the Y-axis position to separate points and hence to improve clarity.

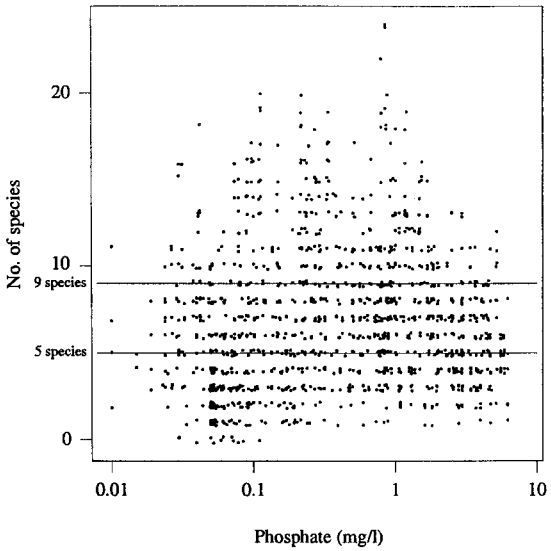


Figure 48. Relationship between the logarithm of phosphate concentration and the numbers of highlighted species in surveys. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets post 1985. Macrophyte data were matched with phosphate either from the same site or from a site within 1 km. A small random movement has been added to the x- and y-axis position to separate points in order to improve clarity.

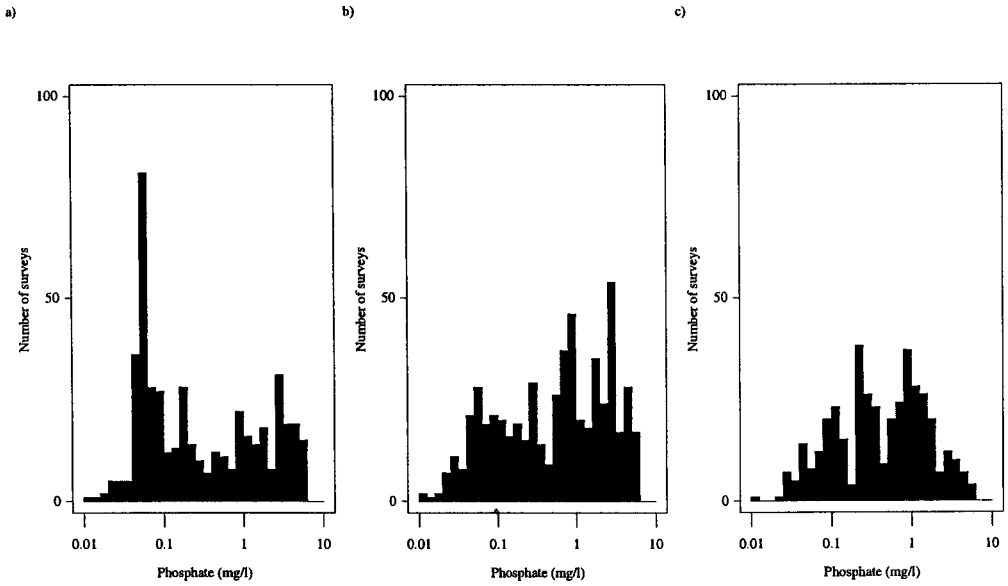


Figure 49. Relationship between logarithm of the phosphate concentration and the number of surveys with a) less than 5, b) 5-8 and c) >8 highlighted species. Data from EA, IFE, DoE/IRTU and Conservation Rivers datasets post 1985. Macrophyte data were matched with phosphate either from the same site or from a site within 1 km.

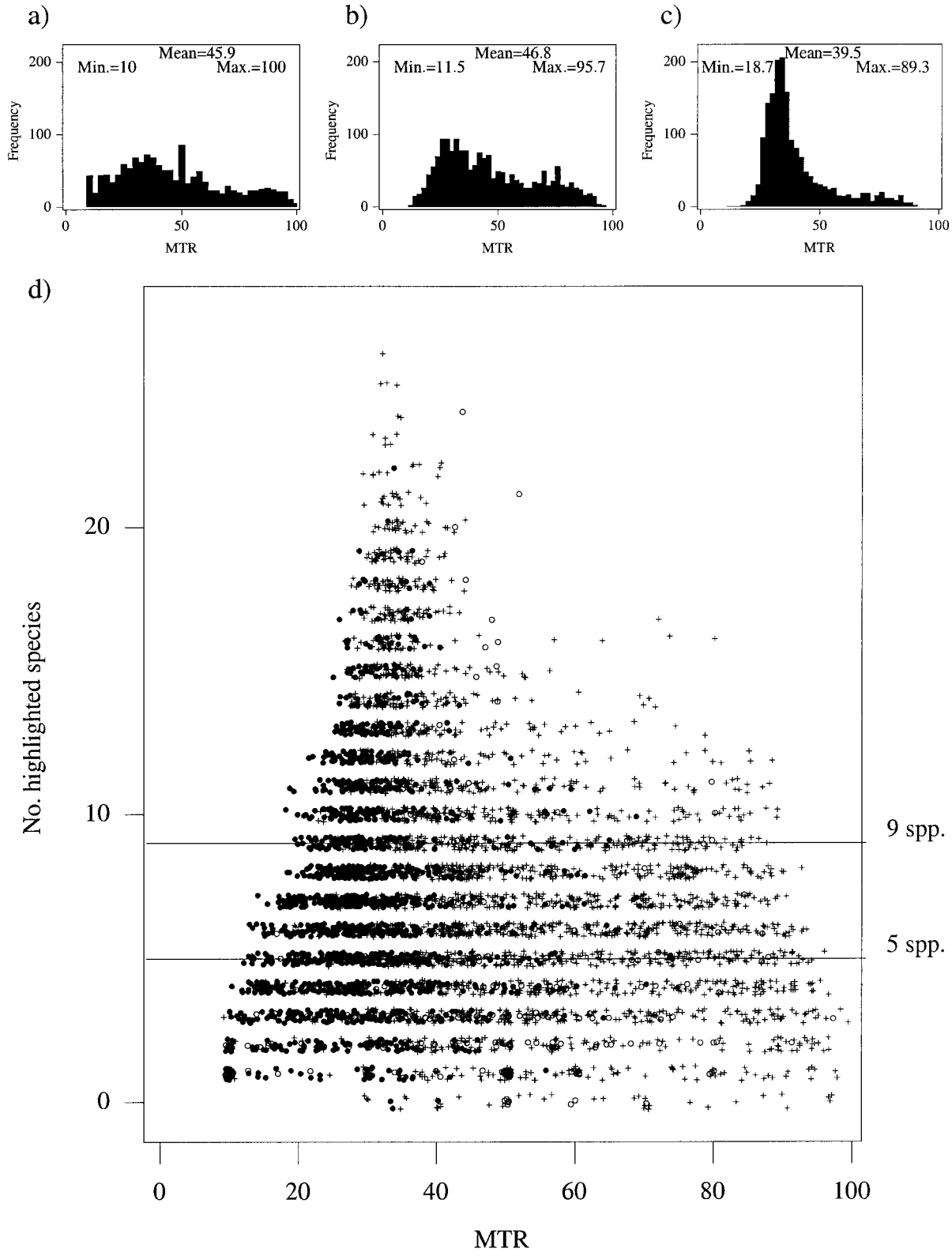


Figure 50. Relationship between MTR score and the frequency of survey sites with a) < 5, b) 5-8 and c) >8 highlighted species. d) Relationship between MTR and the number of highlighted species for EA (•), Conservation Rivers (+) and other datasets (o). A small random movement has been added to the position x- and y-axis of (d) to separate point in order to improve clarity.

4.7 Measures to improve the quality of MTR data

4.7.1 Introduction

It is important that the maximum possible confidence can be placed in the accuracy, or 'quality' of survey results. This is normally achieved by application of a quality control procedure, the aim of which is to minimise unavoidable errors in carrying out the survey methodology; set quality targets and determine whether these are being met; and provide a means for restoring quality if targets are not met. Random checks are made on a part of the survey process and statistical limits of variation used to extrapolate what proportion of the total number of surveys will fail the quality standards set. There are several assumptions within this quality control procedure:

- the process is standardised (it is repeatable, with defined statistical limits of variation);
- all products are produced by the same 'machine' or by 'operatives' working in exactly the same conditions (it is reproducible, with defined statistical limits of variation);
- the quality of the 'product' can be assessed against predetermined criteria for success or failure.

It has already been shown (4.3 & 4.4) that the MTR survey methodology is repeatable, and that its reproducibility depends on the training of surveyors and the rigorous and consistent application of the method. Statistical limits of variation for repeatability and reproducibility are not, however, available at present. Furthermore, although criteria for success or failure can be set, it is difficult to derive a satisfactory means of determining whether these are being met, and of ensuring that the required quality is maintained. Most standard quality control systems which are used for other biological surveys and chemical analyses, such as control charts, ring sorts and resorting of samples, are not appropriate for macrophytes. Macrophyte surveys produce data directly from a field survey. No samples are taken, except for those required for identification or confirmation purposes. Re-surveys alone are probably not sufficient, as they occur after the surveys have been undertaken and real changes may have occurred in the interim. In addition, the de-limitation of the 'sample unit' available to the re-surveyor, ie the upstream and downstream limits of the survey length and the definition of the 'channel', is itself integral to the methodology and open to error.

Never-the-less, several aspects of macrophyte surveys can undergo quality assurance to a greater or lesser degree. These include:-

(a) operation of the method
(b) number of species
(c) abundance categories
(d) overall percentage cover
(e) identification
(f) database entry accuracy

In addition, survey length selection can be audited if required.

Quality assurance measures relating to many of the above, aimed at minimising errors, are already described as an integral part of the survey methodology (Environment Agency 1996). These should continue to be adhered to as a very minimum (see MTR User's Manual, Chapters 4 & 5). One new recommendation is that a minimum of double-staffing should be adopted as good practice, for the following reasons (see MTR User's Manual, 3.3.8):

- it allows consensus decisions to be made on estimations of percentage cover, macrophyte identification and survey length relocation, which should reduce 'inter-surveyor variation' and thus improve the quality of the survey data collected;
- it may improve the efficiency of the survey by allowing different tasks to be shared simultaneously;
- at some sites, safe working practices may require multiple staffing.

In addition to the integral measures referred to above, it is strongly recommended that surveyors are adequately trained and that re-surveys (audit surveys) are undertaken for quality assurance purposes. These measures are described in full in the MTR User's Manual, Chapter 7 (Holmes et al 1999), and are outlined only briefly below.

4.7.2 Training

Specific training in the following areas is recommended:

- training for new staff in macrophyte identification, the operation of the methodology and in data-handling;
- on-going training for MTR surveyors, including an annual refresher course, a minimum number of surveys to be undertaken each year, and advanced macrophyte identification courses for 'difficult' species such as *Juncus bulbosus*, some *Ranunculus*, fine-leaved *Potamogeton*, *Callitriche* and bryophyte species.

4.7.3 Re-surveys for quality assurance purposes (audit surveys)

Audit surveys are useful in monitoring consistency of performance between surveyors or survey teams, highlighting mis-application or mis-interpretation of the method, and thus providing an additional means to minimise errors. They require, however, a high resource investment and so the number undertaken should be minimised and be proportionate to the need for results to be of high quality.

Two audit protocols have been derived and are described in full in the MTR User's Manual (Chapter 7.3). Each protocol delivers a different level of specification in terms of quality assurance, and each requires a different resource investment. The choice of protocol is left to those managing the MTR surveys (whether this be at national, regional or local level), but may depend on the purpose of the survey programme, the resources available and the cost-benefit/value-for-money.

Once the audit surveys have been completed analysis of results involves establishing the reasons for differences between audit and 'primary' survey results, whether these can be deemed to be significant, and the likely reasons for the differences. It is suggested that action is triggered if any of the following criteria are met:

i) 3 or more species missed, recorded incorrectly or identified incorrectly (or 4 or more, if a total of 20 or more scoring species were recorded in the primary survey);
ii) 10% or more of Species Cover Values differ by 3 or more SCV units;
iii) 20% or more of Species Cover Values differ by 2 or more SCV units;
iv) a difference of more than 15 percent points in overall percentage cover;
v) a difference of more than 4 in the MTR (either direction).

These criteria are based partly on the current project results and partly on levels of 'error' or difference found in a small study of re-surveys in Anglian Region of the Agency (Environment Agency 1996c), which found that 87% of scoring taxa were recorded within one SCV and 94% within two SCV. Criterion (v) is set at a level greater than the median difference between audit and primary surveys analysed in this study and is assumed to be outside the normal deviation when sampled correctly by the same surveyor on different occasions. It is recommended that these criteria are reviewed in the future, when more audit data becomes available for analysis

The action triggered depends both on the reason for the difference in the survey results and on the level of audit protocol adopted for the survey programme. Guidance on appropriate action is given in the Manual (Table 2).

4.8 Variability and error: overall conclusions

1. The MTR is repeatable, but not necessarily reproducible. Both repeatability and reproducibility can be improved by provision of adequate training, correct application of the method and adoption of quality assurance measures.

2. Survey results can only be compared with confidence if the surveys are undertaken at the same time of the survey season.

3. Any change in MTR must be greater than 4 MTR units or 15% for it to be deemed as significant in terms of trophic status, this being twice the mean difference which may be expected from seasonal change in MTR and greater than the median difference which may be expected from the inter-surveyor variation.

4. When providing a collective dataset over a number of years, periods of atypical flow conditions should be avoided if possible.

5. It should be assumed that the size of the river, its slope, substrate size, underlying solid geology and the altitude of its source may influence the MTR, as well as chemical determinands other than phosphate. These effects should be taken into account when interpreting results, and care should be taken when selecting survey lengths to minimise the differences in these factors between sites being compared. Shaded areas should be avoided, if possible, when selecting survey lengths, but although efforts should be made to minimise differences in the amount of shade at sites being compared, the amount of shade is unlikely to have a strong influence on the MTR score.

6. Comparison of MTR scores at physically similar sites (ie with a rating of I or II) along the length of a river can be recommended at this stage although it may be difficult to find similar sites in some rivers. The validity of comparing MTR scores from physically similar sites in different river catchments has not yet been confirmed and so should be treated with caution. The comparison of MTR scores from physically dissimilar sites (with a rating of III), whether on the same or different catchment, is not recommended.

7. Further research is required to confirm the precise influence on MTR of depth, cross-section, flow, substrate size, slope, solid geology, and the altitude of the source. The future development of a predictive element to the MTR system is recommended to address this although additional data may also be required to fill in the gaps in the existing dataset. One such system 'plantpacs' (Plant Prediction and Classification System) is currently being scoped in a collaborative Agency/IFE project (R&D Project No. W1-017). 'Plantpacs' would be designed to use the environmental characteristics of a site to predict the macrophyte flora and corresponding MTR score to be expected in undisturbed conditions. Comparison with the observed flora and MTR score would then allow the degree of disturbance from the 'reference' conditions to be measured. This should give numerically comparable evaluations of eutrophication impact across different sites, rivers and catchments.

8. It is recommended that surveyors continue to assess confidence in the MTR survey, in the comparability of survey lengths and in the resulting MTR scores, by use of three suffixes of confidence. The way in which comparability between sites is assessed should be changed in line with the methodology for DQI surveys. Surveyors need to be provided with clear guidance on the use of these suffixes of confidence.

9. Measures should be adopted to improve the quality of MTR data. In addition to the integral measures undertaken as part of the survey method itself, surveyors should be adequately trained and re-surveys (audit surveys) should be undertaken for quality assurance purposes. The measures described in the MTR User's Manual should be followed.

5 PRACTICAL CONSIDERATIONS

5.1 Introduction

The recommendation of MTR as a routine application in a biological monitoring programme, must not only show that MTR performs well on a technical basis, but that it is also relatively easy to use and cost-effective. In addition, results need to be expressed in a way which is easily understood by non-biologists.

5.2 Ease of use

The MTR is considered to be a relatively easy method to use, provided training is undertaken. The method is relatively straightforward and comparable to other tasks performed by professional biologists. Practical difficulties with the methodology were discussed by MTR practitioners at the workshop held as part of this project in Lancaster in March 1996 (Newman et al 1997 a & b), and some minor amendments or clarifications to the methodology made, as appropriate (incorporated into the User's Manual, Holmes et al 1999). The successful use of the method is dependant on accurate identification and recording of species, and accurate estimation of percentage cover values. These will normally be improved by training and experience, and errors further reduced by multiple-staffing according to the recommendations for quality assurance (4.7.1). Resolution of difficulties with plant identification should be aided by the preparation of herbarium collections for later confirmation of taxonomic status.

Some potential simplifications of the method have been considered. One example would be to use a 5-point scale for estimating the percentage cover of individual species, rather than the current 9-point scale. To evaluate this, data from all sites which were originally surveyed using the 9-point scale were converted to the 5-point scale and the MTR re-calculated. There was no significant difference between the two scores (Figure 51), suggesting that the 9-point scale could be reduced without compromising the accuracy of the results. As it is considered, however, that the change would not deliver a significant reduction in time spent, it is recommended that the full 9-point scale be retained to maintain the maximum potential resolution between sites. This may be particularly important when distinguishing between sites which have few species, but where the species present are very abundant. This recommendation is consistent with the findings from the preliminary analysis undertaken by Holmes (1996).

Several other potential modifications or simplifications to the methodology have been proposed but have been rejected as, in general, they would result in a loss of in-built error checks and a greater dependability on 'one-off' correctness. For example, removing the more commoner and/or cosmopolitan species from the list of scoring taxa would reduce the number of species to record and hence simplify the method. It would also result, however, in a greater dependence on the reliable identification of the 'indicative' species retained on the species list. Simplification of the method by requiring plant identification at the genus rather than species level is not recommended as this would result in the loss of valuable ecological information. Different species of a genus often have narrower and differing ranges of environmental requirements. Restriction of the survey to abundant species only is also not recommended as sparsely-distributed species can have an influence on the MTR recorded (see 4.4).

Feedback from MTR practitioners at a training workshop held in September 1997, confirmed that confusion can arise when using the abbreviations SCV (species cover value) and CVS (cover value score), as they are so similar. However, as there are no viable alternatives, no change to these terms is recommended. Surveyors should take care to ensure that the correct terms are applied.

Most difficulties in applying the methodology relate to interpretation of results rather than the survey methodology itself. This requires an understanding of how aquatic plant communities respond to nutrients and how macrophyte communities are impacted by physical and other chemical factors. The interpretation of single MTR scores is perhaps the most difficult element of the process, requiring an in-depth knowledge of such impacts. The interpretation of downstream changes in trophic status and the significance of such changes is relatively straightforward when paired sites (eg upstream and downstream of a QD) are being compared, provided the guidance in the User's Manual is followed. Any difference of greater than 4 units or 15% is deemed to be significant. For uses other than monitoring of point-source discharges, interpretation of trophic status is difficult unless the MTR is put in a catchment and/or national context. This involves regular macrophyte surveys of catchments from source to sea, which enable diffuse pollution impacts and other factors affecting the MTR score to be elucidated. Results also need to be compared with those from other rivers of a similar type, for example, by reference to the average and top quality MTR scores recorded for different river community types (Holmes et al 1999, Appendix 3). These difficulties in interpretation should be significantly reduced in the future by the development of the 'plantpacs' predictive system currently being scoped by the Agency (see 4.8 & 7.3.2).

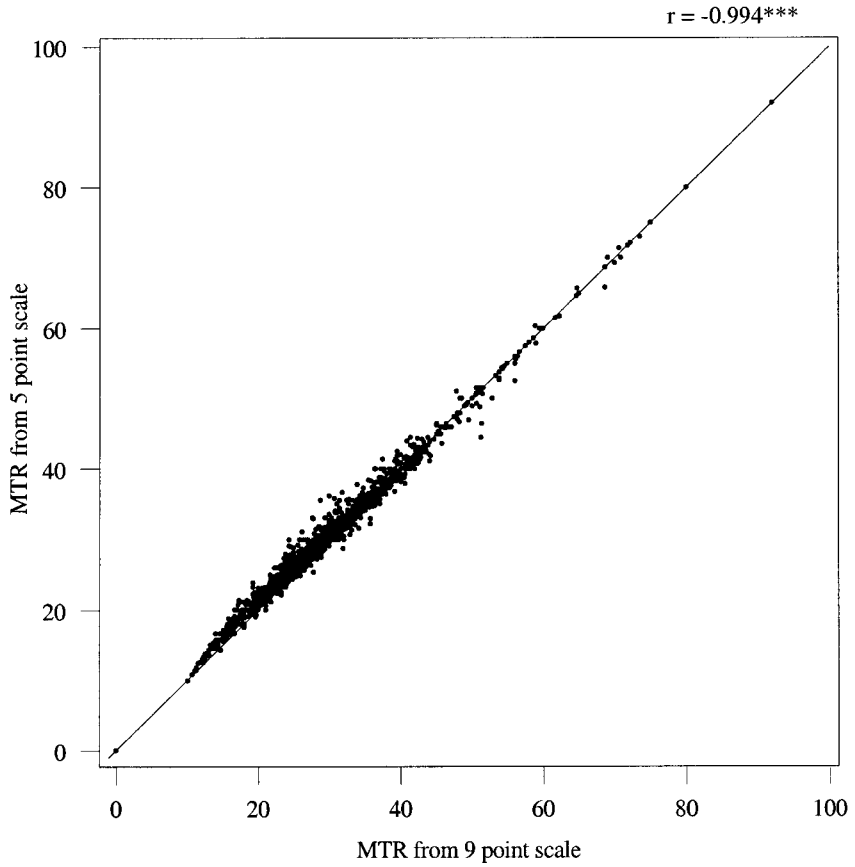


Figure 51. Relationship between the MTR score calculated on surveys completed using the 9 point cover scale and the MTR score at the same sites recalculated by back converting to the 5 point scale (EA, IFE and DoE/IRTU data).

5.3 Cost effectiveness

5.3.1 Introduction

To evaluate the cost effectiveness of the methodology, the following questions were considered.

1. Can the cost-effectiveness of the MTR methodology be improved?
2. How does the cost of an MTR survey compare with other biological methods of assessing trophic status?

5.3.2 Can the cost-effectiveness of the MTR be improved?

Two scenarios were considered:

- Can the cost of the methodology be decreased without reducing the accuracy of the judgement of trophic status? By what means can this be achieved?
- Can the accuracy of the results, in terms of judgement of trophic status, be increased by changing the way data are collected? By what means can this be achieved and at what cost? Does the increase in accuracy justify the increase in cost?

A reduction in cost could potentially be brought about by simplifying the method but it is considered that this would result in loss of valuable ecological information and so is not recommended.

The accuracy of the results could potentially be increased by increasing the survey length, as it is likely that a greater number of species will be recorded. Comparison of surveys completed over 100m and those completed on an overlapping 500m section, however, indicates that increasing survey length is unlikely to change the MTR, or the judgement of trophic status, although the number of species increases by an average of a fifth (Figures 52 & 53). This option is therefore not justified in terms of cost-effectiveness, and the validity of the 100m survey length is confirmed, this being easier and quicker to survey. This is consistent with the findings from the preliminary analysis undertaken by Holmes (1996). It should be noted that re-surveys undertaken for quality assurance purposes, demonstrate the importance of investing sufficient search time/effort within the 100m, to ensure that all scoring species are recorded (section 4.4). The option to undertake a 500m survey in addition, to obtain useful information about the stretch of river and to verify the interpretation of trophic status from the 100m survey, is left to the surveyor. Although the 'working' MTR calculated for a 500m survey will not be significantly different from that derived from the 100m survey in most cases, it should not be used for reporting purposes.

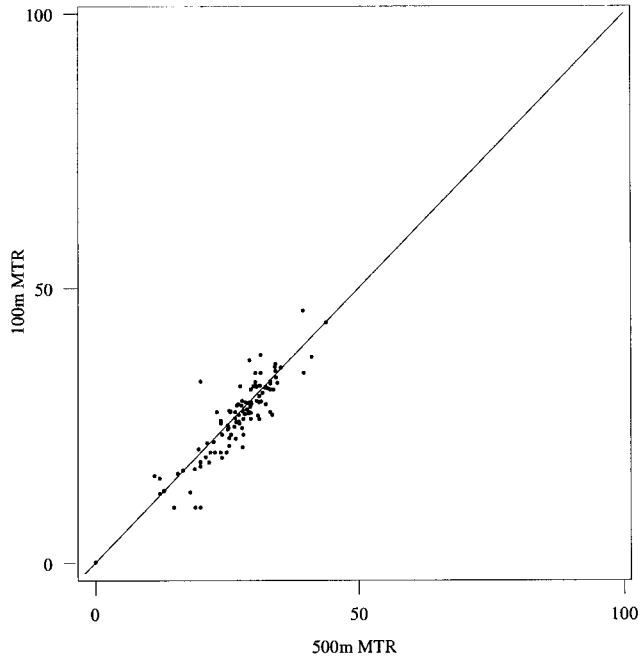


Figure 52. Relationship between MTR score from surveys completed over a 100m length and the MTR from the overlapping 500m section (EA data).

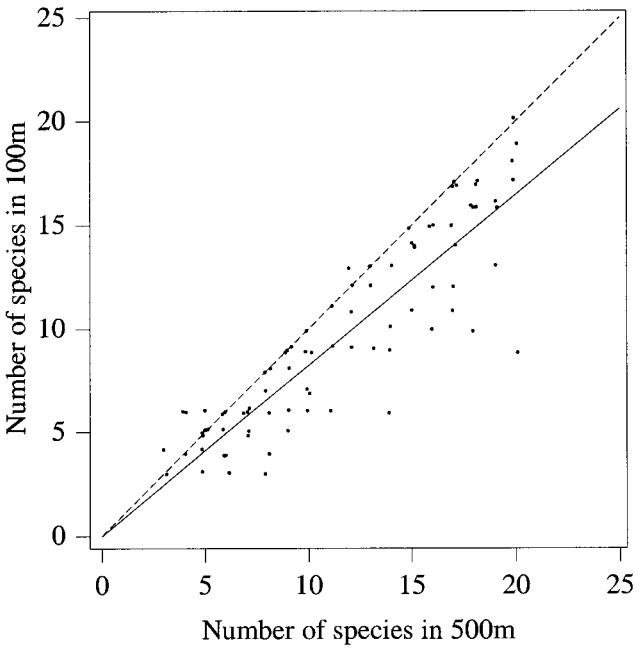


Figure 53. Relationship between the number of scoring species from surveys completed over a 100m length and those from the overlapping 500m section (solid line). EA data. The gradient of the solid line is 0.81 and is very highly significant (r^2 = 78%) and the dashed line is a 1:1 relationship. A small random movement has been added to the x- and y-axis position to separate points to aid clarity.

5.3.3 How does the cost of an MTR survey compare with other biological methods?

The time required to undertake an MTR survey - including planning, field survey, laboratory analysis, data input and data interpretation - is approximately one person-day per site, when a small number of surveys (~10) are undertaken by a team of two 'average' practitioners, within a reasonable travelling distance of their base. This is considered to be comparable to the time required for the only other biological method for trophic status assessment, the DQI (TDI). To put it into a wider context, it may also be comparable to the invertebrate-based RIVPACS method, although times for all three methods can be significantly less (particularly for RIVPACS) depending on the number of species present and the nature of the sample site. Variation does exist between the three methods, however, in the relative proportion of this time spent in the field and in the laboratory, and the flexibility with which survey programmes can be planned (Table 9). For example, MTR surveys require a greater proportion of time in the field and any laboratory analysis must be undertaken shortly after return to the laboratory. Whereas DQI (TDI) and RIVPACS surveys require a greater proportion of time spent in the laboratory, but as samples can be stored prior to analysis planning of survey programmes can be more flexible (samples can be stored and analysed in batches as appropriate).

A fully costed comparison to take into account capital and running costs was not made, and may not be relevant to the selection of method, as different survey teams have differing levels of access to, for example, equipment and reference material. Such a comparison would require a small additional project. As a crude comparison, however, it is useful to consider the equipment and chemicals required for a survey team starting with nothing. In this scenario, the capital costs of undertaking DQI (TDI) and/or RIVPACS surveys are greater than MTR surveys as the former two methods both require a fume cupboard, and DQI (TDI) also requires a centrifuge, hotplate and appropriate glassware. Microscope facilities are required by all three methods. Running costs in terms of chemicals for preparation and preservation are also higher for DQI (TDI) and RIVPACS.

Bringing capital and revenue (time and chemicals) costs together, the total cost per MTR survey is certainly comparable to that of other biological methods used to assess trophic status or water quality, and may even be cheaper depending on equipment already available. This may be offset to some extent, however, by the restrictions on survey programme flexibility imposed by the requirement to analyse all specimens immediately. Such restrictions may reduce efficiency.

Where two or more of the methods are required within a biological monitoring programme, efficiencies can be made by combining surveys. For example, samples for DQI (TDI) can easily be taken in conjunction with an MTR survey. The only limitation to this would be where by doing so would result in too long a time delay between surveys of sites being compared, eg sites upstream and downstream of a QD. This is only likely to be the case where all three methods (MTR, DQI and RIVPACS) are undertaken in conjunction with each other.

Table 9. Comparison of aspects of the cost effectiveness of different biological and chemical methodologies for assessing water quality, based upon the average trained and experienced practitioner.

Method	Relative effort of major elements of methodology	Benefits	Disadvantages
MTR for aquatic macrophytes	**Survey**, database entry, identification, confirmation and storage of plant material	Identification relatively easy; information immediately available; nationally applicable [compatible with method for assessing conservation status]; shows long-term conditions	Affected by other chemical and physical factors; comparison between rivers not fully understood. [slow to respond]
DQI for diatoms	Survey, **slide preparation and identification** (slide storage)	Samples quick to take; probably nationally applicable; gives measure of organic pollution effect; shows short-term conditions	Limited by substrate suitability; identification may be difficult or time consuming.
RIVPACS for macro-invertebrates	Collection, preservation, **sorting** and identification	Samples quick to take; identification relatively easy with good identification keys; nationally applicable	Sorting and identification often time consuming; derived for organic pollution but limited in ability to reflect trophic status *per se*.
Chemical	**Immediate analysis of 'physical' determinands,** longer period for others (1-2 weeks)	Samples quick and easy to take	Spot check, limited in ability to monitor changes in time unless frequently taken.

5.4 Ease of understanding or interpretation by non-biologists

5.4.1 Introduction

Good communication of survey results facilitates appropriate management decisions and assessment of priorities. Poor communication of results limits the usefulness of the information gathered and may result in poor river management decisions or the inability to prioritise management resources to the areas of greatest need.

The MTR system is a means of presenting complicated information on the macrophyte community, in a simple numeric form. This is potentially both easily communicable and manageable. As with many biological methods of assessment, however, the numbers produced must be interpreted by biologists trained in the methodology, before assessments of trophic status can be made. To enable the interpreted data to be communicated easily to others, standard guidance is required, giving a consistent approach to both the interpretation and presentation of data.

5.4.2 Detection of point-source nutrient inputs

For data gathered for UWWTD purposes, or for the assessment of other point-source inputs, standard guidance has been achieved in the present study by devising 'decision trees' which, with the addition of supplementary helpful information, guide the biologist to one of several 'standard descriptors' both for the trophic status of an individual survey and for whether there is a significant downstream change in trophic status. Guidance is given on the presentation of data in a simple format (see User's Manual, Holmes et al 1999).

5.4.3 Detection of diffuse inputs

Interpretation of results relating to applications other than the assessment of point-source inputs, is more complex, and hence the communication of survey findings more difficult. The main area of concern is the potential mis-use of the system for purposes for which it is not yet deemed appropriate, by those who understand neither the complicating factors which determine the MTR on a local scale nor the unreliability of comparing actual MTR scores across catchments or geographical areas. The inherent problem with a system which produces numbers is that the numerical results are assumed to mean the same thing on all occasions. The evidence produced during the present study shows that this assumption may well not be valid for MTR and that factors other than trophic status may influence the MTR score. For this reason, the cross-comparison of actual MTR values between catchments is NOT recommended at the current time, except where catchments are of the same physical type (and even then the method has not yet been proven).

This advice, however, may restrict the usefulness of the MTR, as without cross-catchment comparisons, individual catchments cannot be placed into context and river management resources cannot be targeted at the catchments in most need of eutrophication control. Two alternative approaches to data presentation, to overcome this limitation, were thus considered: the comparison of MTR scores presented in the form of bands or classes; and the comparison of catchments based on the downstream impacts demonstrated by the MTR.

Banding scheme

A banding/classification scheme, to enable mapping of eutrophication at a catchment or wider scale, is not considered appropriate due to the complexity of factors influencing the MTR. Very broad comparisons can be made in terms of the three 'class ranges' used for interpretation of results for UWWTD purposes (MTRs of < 25, 25 - 65, and > 65; see Holmes et al 1999), but these classes are likely to be too wide to be of much operational use as a banding scheme. Although MTR scores may be placed into arbitrary groups for mapping purposes, such maps must only be used to illustrate MTR scores: no inference should be made to trophic status, which should be based on a site-by-site interpretation. To avoid mis-interpretation of such maps, their use should be discouraged.

It is recommended that any tentative banding scheme be limited to descriptive indicative guidance as to the ranges of MTR values which have been recorded in rivers of differing physical and chemical types (see, for example, Appendix 3). This should not be used to group MTR scores into bands for presentational purposes, but more to assist decisions regarding the trophic status of individual sites.

A robust, scientifically sound, banding or classification scheme to allow national mapping of eutrophication, cannot proceed until after the development of a predictive element to the MTR system. A prediction and classification system for aquatic and riparian plants, currently named 'plantpacs', is in the initial stages of planning with the Agency and should allow a classification scheme to be derived based on the level of eutrophication impact (measured as the degree of disturbance from some undisturbed reference condition – see 4.8). Such a scheme could contribute to monitoring and reporting for a range of applications, including the UWWTD, the proposed EC Water Framework Directive and national eutrophication strategies.

Downstream impacts

Catchments may be compared on the basis of the relative degree of downstream change in trophic status as demonstrated by the MTR. This may help to prioritise those catchments which would benefit most from nutrient control measures. The approach taken for the presentation of evidence for the designation of SA[E]s (Holmes at al 1999), whereby downstream reductions in MTR of 4 units or 15% or more are deemed 'significant', would provide a first stage for prioritisation; cross-catchment comparisons of actual percentage decreases in MTR of more than 15% would provide a second stage.

5.5 Practical considerations: overall conclusions

1. No substantive simplification to the methodology is required. Where minor amendment or clarification would make the method easier to use without losing valuable ecological information, this has been incorporated into the procedural guidance given in the User's Manual. The MTR survey methodology is considered to be a relatively straightforward method to use, provided adequate training is undertaken and the User's Manual is read, understood and used. The main difficulties lie in interpretation of the results, particularly when interpreting single MTR scores and for applications other than UWWTD (or other point-source) monitoring.

2. The MTR delivers a cost-effective assessment of trophic status. No changes are recommended in this respect.

3. Guidance for biologists on the interpretation of results, based upon the findings presented in this report, is given in the current MTR Users' Manual (Holmes et al 1999). Given this guidance and an understanding of the underlying principles behind the method, MTR survey results can be communicated to non-biologists without significant difficulty for those applications requiring the impact of point-source discharges to be assessed (for example, UWWTD monitoring).

4. For applications requiring cross-catchment comparison of trophic status, interpretation of results is more difficult. Mapping of MTR at a catchment or wider scale is not appropriate due to the complexity of factors influencing the MTR. Never-the-less, the MTR methodology may still be useful in gaining an overview of the trophic status of catchments in order to prioritise those areas which would benefit most from further investigation or nutrient reduction effort. The broad principles applied to the assessment of point-source inputs can be adopted, assessing trophic status on a site-by-site basis and comparing the magnitude of downstream changes.

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6 COMPARISON BETWEEN MTR AND OTHER METHODS

6.1 Methods to be compared

At the start of this project, a literature survey was undertaken to find potential methodologies for the assessment of the trophic status of rivers, to compare with MTR. Those found were limited in various ways, for example, by not having a broad national approach or perspective, not utilising species representative of the UK flora, or by not being fully proven scientifically. Thus to address the need for a nationally validated system(s) to assess the trophic status of rivers, the comparison between the macrophyte-based MTR and other potential methods was limited to the other principal method under development for the Agency, ie the diatom-based DQI (based on TDI). This comparison is described in 6.2 and 6.3. The recently developed River Trophic Status Indicator model (Ali et al 1999) is considered briefly in 6.4.

6.2 Criteria and methods for comparing MTR and DQI

The comparison of DQI and MTR was based upon the criteria listed for MTR in 2.2, together with consideration of the similarity or advantageous differences of the results in a range of circumstances such as: stable or variable river flow; perturbations such as flood or spates, pulses of nutrients or pollutants; nutrient-rich or -poor rivers; and physical habitat range. Consideration was also given to situations in which neither method is suitable (eg toxic contamination or heavy organic pollution), to seasonal applicability, and to the circumstances in which each method can be used effectively.

The comparative performance was evaluated by:

a) comparing the operation of the two methods;
b) undertaking DQI survey, in conjunction with MTR, along two rivers to provide case studies for analysis;
c) collating the DQI data available from the Environment Agency's TDI project (R&D Project 618) and, together with data gathered in (b), comparing these with MTR recorded at the same sites.

6.3 Comparative performance of MTR and DQI

6.3.1 Operation of the methods

Comparison of the MTR and DQI showed that both have a relatively easy method of sampling which is suitable for combination with invertebrate programmes; they are both reliant on a finite number (about 100) of mostly easily identifiable, widely-distributed species or taxa; and, analysis of results requires only a straightforward computation. They differ in their permanent record of conditions: DQI samples are stored on microscope slides for future reference or comparison, whereas larger herbarium reference material or samples are required for some but not all macrophytes. Both methods are relatively amenable to analytical quality control or quality assurance.

6.3.2 Field surveys

Field surveys comparing the MTR and DQI systems were undertaken on the Rivers Kennet and Loddon in southern central England. Diatom material from river stones was prepared and the species assemblage determined using the standard TDI/DQI methodology, which requires the identification of some 250 specimens per slide (Kelly 1996b, for update see Kelly in Prygiel et al 1998 et seq.). Diatom Quality Indices, a transformation of TDI (see 1.5.2), were calculated from the data following the guidelines set out by Kelly (1996a, b, & c) and using the equation:-

$$WMS = \Sigma asv / \Sigma av$$

where *WMS* = *Weighted Mean Sensitivity*
a = *taxon abundance of species in sample*
s = *pollution sensitivity of species* (values range 1 to 5)
v = *indicator value* (range 1 to 3)
Σ = *sum of*

and *Trophic Diatom Index = (WMS x 25) - 25*

or *Diatom Quality Index = 100 - TDI.*

The amount of organic pollution is given by the percentage of certain specified diatom taxa ie percentage of Pollution Tolerant Valves (% PTV).

The Rivers Kennet and Loddon were sampled as case studies at 13 sites (Figure 54):

a) The River Kennet from above Marlborough to Aldermaston.

Overall, the MTR was moderately high along the entire study length, although there was a slight downstream decrease (Figure 54a). The DQI followed a similar pattern to the MTR upstream of Hungerford, increasing from 30 above Marlborough, to 41 as the river passed through the town, before falling to 36 below the WWTW discharge. In contrast to the MTR, however, the DQI declined significantly downstream of this point, decreasing to 17 and then to a minimum of 8 above the Newbury WWTW. Organic pollution was highest above Marlborough and Newbury WWTW discharges (15% PTV), with a minimum PTV of 5%.

The low DQI values in the lower reaches of the river may be due to the nature of the river bed at these sample sites, it being silty with a dense cover of filamentous algae. The Kennet and Avon Canal flows into the river just upstream of Hungerford, where the River Dun joins the River Kennet, and then merges with the River Kennet upstream of Newbury. From this point downstream the whole nature of the river changes, this including a decrease in phosphorus which may be a result of it being complexed out of solution by calcite formation. There is also a commercial trout farm and a small wastewater treatment works.

b) The River Loddon between Basingstoke and Reading.

Overall, the MTR was moderately high along the entire study length, although there was a downstream decrease (Figure 54b). The DQI followed a fairly similar pattern to the MTR, except for upstream of Basingstoke WWTW where it was notably lower. The DQI increased from 16 above Basingstoke to 36 downstream of the town, before decreasing to 17 at Reading. The PTV increased from 2.4% upstream of Basingstoke to 7.5% downstream of the discharge and continued to rise downstream to reach 17% at Reading.

Where the pattern between DQI and MTR is dissimilar, as upstream of Basingstoke WWTW, the low DQI and increased proportions of *Amphora pediculus* appear to indicate a greater degree of enrichment than do the macrophytes. The results may have been distorted, however, by the site conditions. The river was densely shaded, with abundant filamentous algae. Diatom abundance was considerably reduced, the diatom material collected was found to be in a poor state and some specimens (eg the planktonic forms) had quite clearly come from elsewhere. Such conditions, which would also affect the macrophyte assemblage, were typical of this section of river and therefore the only available type of sample site. Natural temporal variation in both diatom and macrophyte floras may also have affected results at this site. DQI values from surveys by M. Kelly were 26 (15% PTV) upstream of Basingstoke WWTW and 27 (38% PTV) downstream in November 1996, compared with 13 (15% PTV) upstream of Basingstoke WWTW in May 1996 (Kelly, pers. comm). Similarly, decreases in MTR of 15–20% are reported to have been found previously between upstream and downstream of Basingstoke WWTW, but in this study the MTR was similar at both sites.

c) Conclusion

In both the Kennet and Loddon surveys, DQI and MTR were found to broadly follow the same downstream pattern. Where this was not the case, the difference was likely to be due to poor site conditions: a silty river bed, dense shade and/or abundant filamentous algae. Temporal variation in results may also occur. Levels of organic pollution in both river systems were less than 20% PTV, ie within Kelly's lowest category (Kelly 1996a), meaning that this was unlikely to mask the enrichment of the two rivers.

These results emphasise the value of undertaking both DQI and MTR together at the same site(s), but that for both methods, it is important to take site conditions into account when selecting sites and interpreting results and to sample on more than one occasion (which may mean two or more seasons of the year for DQI, and over two or more years for MTR).

The repeatability of DQI was not tested and only single MTR surveys were used in the comparisons with DQI, but for both these rivers the results recorded were similar to other results available.

It should be noted that DQI and MTR should not be expected to be numerically the same value.

6.3.3 Comparison of DQI and MTR data from various sources

TDI data gathered during the Agency TDI development were provided by Agency staff or by courtesy of Martyn Kelly. The data were then transformed to give DQI values and compared

with MTR values recorded at the same sites. Environmental Quality Index-Average Score Per Taxa (EQI-ASPT) data, where available, were also included in the comparison (based on macro-invertebrates). There was a broad relationship between MTR and DQI, and between MTR and EQI-ASPT (Figure 55), although differences were also apparent. Separation of the MTR and DQI data by phosphate concentration (less than and greater that 1 mg l^{-1}), did show a distinction between the two subsets which indicated that the relationship with phosphate may not be the same for DQI as for MTR.

The joint dataset was too sparse to be examined in detail for the comparative effects of, for example, perturbations (eg flood and flashy spates, pulses of nutrients, pulses of pollutants), or substrata over the full range of river from upland oligotrophic to enriched lowland ones. Some seasonal data were examined for DQI with a relatively stable result being found.

6.3.4 Conclusions

Comparison of the two systems (MTR and DQI) is based upon the use of a less extensive diatom dataset and in the absence of an analysis similar to that of MTR against nutrient status. Nevertheless both systems appear to give a broad assessment of trophic status over the range of conditions studied. The two methods are likely to be complementary when DQI is taken in addition to MTR, particularly as DQI may demonstrate the additional effects of organic enrichment. Practical advantages and disadvantages can be seen for both; *either* in more rapid field work but more extensive and skilled laboratory analysis (DQI) *or* in more field time and less laboratory time (MTR). Both methods, however, rely on a field visit at which habitat data should be recorded and which involves a commitment of time. Field visits could or should, be combined with other field survey or sampling such as for water, sediment or macro-invertebrates. The use of the DQI and MTR methods to complement each other could overcome requirements to sample out of the main growth seasons for macrophytes. It could also demonstrate both short-term (DQI) and longer term (MTR) changes in trophic status.

In conclusion, DQI is a useful complementary technique to MTR, but requires more development to include a wide range of habitats comparable to the Conservation Rivers database.

Footnote An updated guidance standard for European use was the subject of a workshop on Diatom Sampling in Douai, October 1997 (Prygiel et al 1998). Previous work has been presented in Whitton and Rott (1996) and Whitton et al (1991).

6.4 Using functional variables as an alternative to MTR

The River Trophic Status Indicator (RTSI) models are a series of models developed by Ali et al (1999), using functionally-derived variables (mainly based on morphological attributes of freshwater macrophytes) to predict the trophic status of rivers. The approach contrasts with the MTR system which uses the species assemblage. Ali et al (1999) found that both the RTSI model based on plant functional groups and a second model based on field-measured traits perform as well as MTR at predicting river water phosphorus concentration but that better prediction can be obtained by combining both types of RTSI measure. Examples of traits used include the mean number of lateral branches per specimen, the mean submerged-leaf biomass and the mean floating-leaf biomass. However, their comparison between RTSI and MTR did not extend to practical elements of the methodology, such as cost-effectiveness

and ease of use, which would need to be evaluated before recommendation as a routine monitoring method could be considered. In the future, the development of a system to predict the macrophyte flora of a site from environmental parameters (such as the 'plantpacs' system currently being scoped) should not only enhance the performance of the MTR system but would also have the advantage, compared to the RTSI model, of providing species information for conservation purposes.

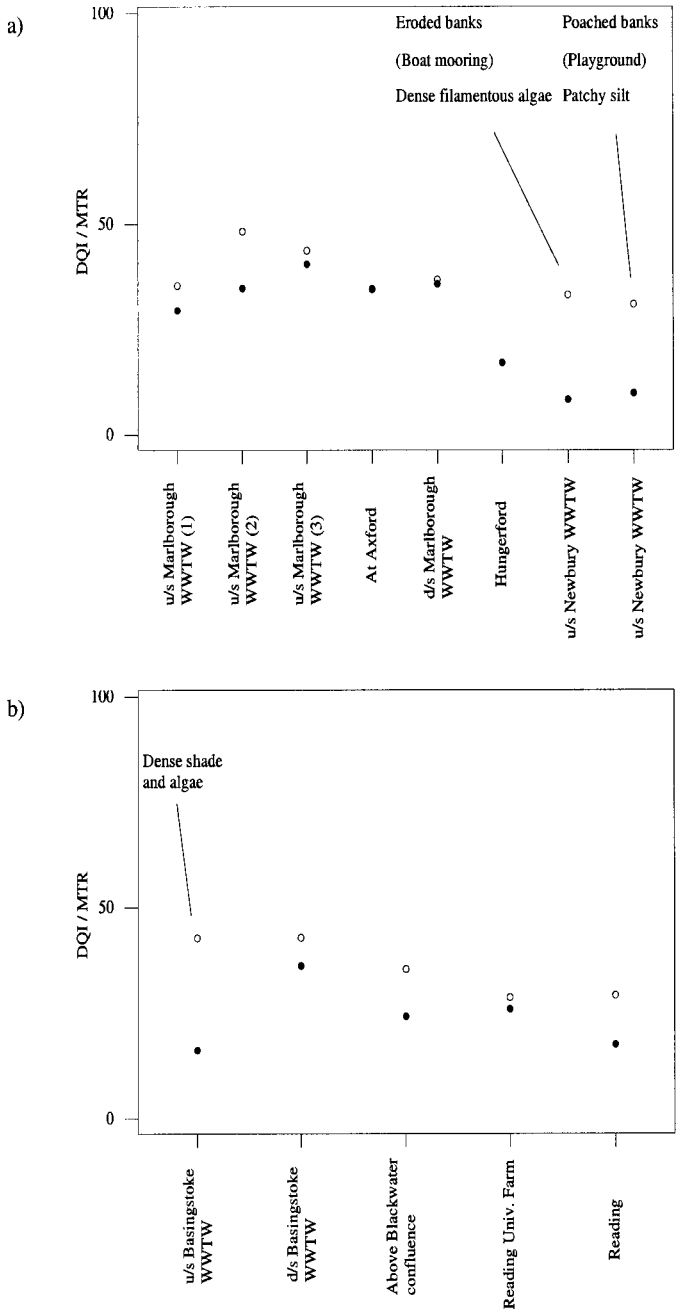
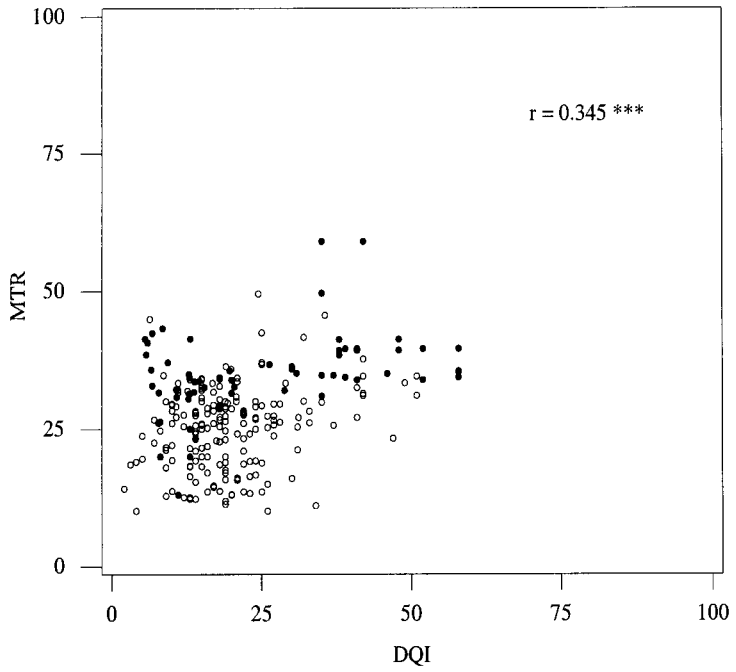


Figure 54. A comparison of MTR surveys (O) with DQI (●) for a) the River Kennet and b) the River Loddon, IFE, 1996.

a)



b)

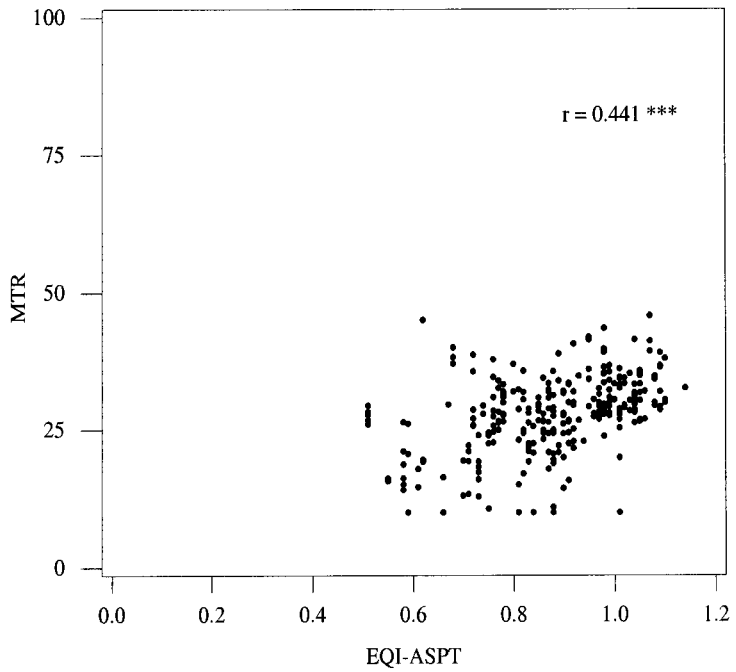


Figure 55. Relationship between a) DQI and MTR from surveys completed within one month of each other (divided into less than 1 mg l^{-1} phosphorus (●) and greater than 1 mg l^{-1} phosphorus (O), n =254) and b) EQI-ASPT and MTR scores from surveys in the same year. EA data.

7 SUMMARY, CONCLUSIONS & RECOMMENDATIONS

The overall objectives of the project were:

- to evaluate the Mean Trophic Rank (MTR) system, in terms of its method, results, deficiencies, limitations and refinements, to produce a robust transportable system for assessing the trophic status of rivers using macrophytes;

- to compare this and other biological methods of assessing trophic status of rivers (principally DQI) and to evaluate them;

- to produce a Recommended Method to assist in the designation of phosphate-based Sensitive Areas (Eutrophic) under the Urban Wastewater Treatment Directive (UWWTD).

Other applications of the MTR were also considered. The research findings are summarised below in terms of these objectives. The Recommended Method is given as an accompanying volume to this report (R&D Technical Report E38, Holmes et al 1999).

7.1 Performance of MTR

The performance of MTR as a useful tool to assess the trophic status of rivers in terms of the macrophyte community, was tested by gathering relevant macrophyte, physical and chemical data from field surveys and subjecting these data to a series of analyses. A set of criteria for the ideal system was established and data gathered to allow the evaluation of the performance of the MTR against these criteria. Following assessment of the suitability of these data, the MTR methodology was evaluated as a means of assessing nutrient status. The source and range of variability and error were investigated and the practical elements of the methodology considered. Throughout this evaluation, deficiencies and limitations of the MTR method were considered and possible refinements suggested, with a view to making recommendations as to operational use.

7.1.1 Suitability of the data

Macrophyte, physical habitat and chemical data from field surveys were gathered from various sources and assembled into a database. The suitability of these data was assessed by considering the overall size of the database and the variety of different types of sites included.

1. The overall size of the database was sufficient for statistical confidence to be obtained from analyses.
2. Datasets from different sources complemented each other and extended the range of data available for analysis.
3. The range of physical, chemical and macrophyte characteristics of sites was sufficient to allow analysis of the influence of the physico-chemical environment on plant species and on the MTR.
4. The geographical distribution of sites gave sufficient national coverage, although some parts of the dataset were incomplete in terms of physico-chemical parameters and bias the applicability of the MTR evaluation towards England.

5. Future gathering of data from sites with a wider range of physical substrate and under a full range of water chemistry, would benefit further method development.
6. It was assumed that the quality of the macrophyte data was consistent between different datasets and was adequate for evaluation of the MTR.
7. As phosphate data were only available for 116 of the 129 MTR-scoring species, it was not possible to analyse the nutrient tolerances of the rarer species. In the future, gathering of distributional data on rarer plants, confirmation of their identification, and concurrent collection of physical and chemical data, would benefit further method development.

It was concluded that the data gathered would allow a sufficiently comprehensive evaluation of the MTR system. The plant data were combined into a relational database ('MTR Database'). Two versions of this database were produced. One holds all the data gathered and is subject to access restrictions under formal written agreement with the collaborating partners (see Project Record, Dawson et al. 1999, for details of access arrangements). The second version holds only those data not subject to access restrictions and includes all data contributed by the Agency, IFE and DoE/IRTU. A reference copy of this second database is held with the Project Record on CD-ROM (Microsoft Access 97), together with instructions on its operation. Although not a specific objective of the project, the second database can be used for operational purposes on a single-user basis.

7.1.2 MTR and nutrient status

MTR is a means of expressing the trophic status of a river in terms of the response of the macrophyte community to nutrient status. Evaluation of the relationship between MTR (and its components) and nutrient status was thus the first step in determining the technical performance of the MTR. Although the evaluation focused primarily on phosphate concentration as a proxy for nutrient status, some analyses were also undertaken using nitrate concentration.

1. The aquatic macrophyte flora showed a spectrum of tolerances to nutrient enrichment. Although most species occurred over a broad nutrient range, differences in species' preferences were apparent. Species occurrence and/or abundance (percentage cover) increased with nutrient concentration for some species, and decreased for others.
2. Species Trophic Ranks (STRs) were found to reflect broad species' tolerances to nutrient enrichment, especially in relation to phosphate enrichment. No changes to STRs are recommended at this stage of method development, with the exception of *Stigeoclonium tenue* which should be removed from the scoring list. Further research is needed to confirm the STRs for several other species. Re-ranking of species based on the mean phosphate or nitrate concentration at which they occurred, helped to identify which species may potentially require a change in STR.
3. MTR was found to decline with increasing phosphate and nitrate concentration. A more significant correlation was found when the logarithm of phosphate concentration was used, but not when the logarithm of nitrate concentration was used. The correlation was particularly strong at concentrations below 1 mg^{-1} P or 10 mg^{-1} N, but even more at concentrations less than 0.5 mg^{-1} P or 5 mg^{-1} N.
4. Although MTR cannot be used to distinguish between phosphate and nitrate enrichment, comparison of phosphate-based ranks with nitrate-based ranks suggests that some individual species may potentially be useful in this respect. However, phosphate is likely to be the limiting factor in most cases.

5. A general pattern of decreasing MTR downstream was often seen at QDs, although MTR may also vary along the length of a river, probably in relation to diffuse nutrient inputs. To gain a better understanding of downstream variation in MTR, future sampling should be extended to intermediate sites between existing MTR sites and to the systematic survey of rivers. These MTR surveys should be concurrent with sampling of water chemistry and should be repeated at intervals of a few years.
6. It was not possible to evaluate the performance of the MTR at assessing temporal changes in MTR, due to the current lack of adequate time-series data. To allow such evaluation, it is recommended that MTR monitoring continue, particularly where phosphate reduction measures are commissioned (or cease).
7. MTR is applicable to use across the UK. The distribution of STR 2 species may merit further investigation to improve national applicability.
8. Species diversity and/or overall percentage cover may, in some cases, provide useful supplementary information to MTR results, although these parameters should not be used alone as indicators of trophic status.
9. No scoring species was found which could be used as a 'key' or 'indicator' species to help interpretation of trophic status.

It was concluded that MTR is a useful broad tool for assessing trophic status, and may be most useful at detecting eutrophication impacts when the concentrations upstream (or prior to) the nutrient input are less than 1 mg^{-1} phosphate-P or 10 mg^{-1} nitrate-N. The methodology detects the symptoms of eutrophication as defined by the UWWTD and the Environment Agency's Eutrophication Strategy: increased abundance of species tolerant of nutrient enrichment results in a reduction in the MTR score, reflecting a disturbance to the 'balance' of macrophyte species present.

7.1.3 Variability and error

The potential sources of variability and/or error in the MTR methodology were considered, including the inherent variability of the method (repeatability), variation between surveyors (reproducibility) and the natural background variation. This allowed an assessment of the robustness of the MTR methodology and the potential for confidence limits to be assigned to MTR scores.

1. The MTR methodology is repeatable but not necessarily reproducible. Both repeatability and reproducibility can be improved by provision of adequate training, correct application of the method, and adoption of quality assurance procedures.
2. MTR may vary within the survey season. Survey results should thus only be compared with confidence if the surveys are undertaken at the same time of the survey season.
3. Any change in MTR must be at least 4 units or 15% to be deemed as a significant change in trophic status, this being twice the mean seasonal change in MTR and greater than the median difference due to inter-surveyor variation.
4. Atypical flow conditions should be avoided, especially as part of a collective dataset over a number of years.
5. MTR is influenced by the physical character of the river and its water chemistry. The size of the river, its slope, substrate size, underlying solid geology, the altitude of its source and water chemistry, should be assumed to influence the MTR and accounted for when interpreting results. Care should be taken when selecting survey lengths to minimise the differences in these factors between sites being compared. Shaded areas

should be avoided although the amount of shade is unlikely to have a strong influence on the MTR score itself.

6. Downstream comparison of MTR scores at physically similar sites along the length of a river can be recommended at this stage. Comparisons between physically similar sites in different river catchments should be treated with caution. Comparisons between physically dissimilar sites should not be made.
7. Further research is required to confirm the precise influence on MTR of depth, cross-sectional area, flow category, substrate size, river slope, solid geology, and the altitude of the source. This may be addressed by the development of 'plantpacs'; a predictive system currently being scoped in a collaborative Agency/IFE project (R&D Project No. W1-017).
8. Surveyors are recommended to continue to assess confidence in (a) the MTR survey, (b) the physical comparability of survey sites, and (c) the resulting MTR scores, by use of the three suffixes of confidence. The way in which comparability is assessed should be changed in line with the methodology for DQI surveys. Clear guidance on the use of these suffixes of confidence is needed for surveyors.
9. Quality assurance measures should be adopted to improve the quality of MTR survey data. In addition to integral measures undertaken as part of the survey method itself, surveyors should be adequately trained and re-surveys (audit surveys) should be undertaken for quality assurance purposes, as described in the MTR User's Manual. The use of herbarium-style or reference plant material should be encouraged, to allow confirmation of plant identification and to provide a training resource. Ongoing training should emphasise the care necessary for assessment of percentage plant cover; location of sparse species; identification of 'difficult' species such as *Ranunculus* species; and recognition of 'natural' events such as washout of species, changes in water quality (other than trophic status) and variations in year-to-year flow conditions.

It was concluded that surveyor error, seasonal patterns of plant growth and differences in the physico-chemical character of the river, can all contribute to variability in the MTR recorded for a given trophic status. Training and adoption of quality assurance measures can help to reduce errors. Natural temporal or spatial variation must be allowed for when selecting site locations and survey timing, and when interpreting results. Future development of a predictive element to the system is required to take account of this natural variation and hence increase the robustness of the method. The scope of such a predictive system ('plantpacs') is currently being established.

7.1.4 Practical considerations

For MTR to be recommended as a routine application in a biological monitoring programme, MTR must perform well on a technical basis but it must also be relatively easy to use, cost-effective, and give results which can be expressed in a way which is easily understood by non-biologists. Assessment of these practical considerations yielded the following conclusions.

1. The MTR survey methodology is considered to be a relatively straightforward method to use, provided adequate training is undertaken and the User's Manual is read, understood and used. No substantive simplification to the methodology is required. Minor amendments or clarifications are incorporated into the procedural guidance given in the MTR User's Manual.

2. The main difficulties in the MTR method lie in the interpretation of the results, particularly when interpreting single MTR scores and for applications other than UWWTD (or other point-source) monitoring. This is due to the natural background variation in MTR, which must be allowed for when determining trophic status (see 7.1.3). Guidance for biologists on the interpretation of results, based upon the findings presented in this report, is given in the MTR User's Manual (Holmes et al 1999).
3. The MTR methodology delivers a cost-effective assessment of trophic status. Costs are comparable with those of other routine biological methods of water quality assessment. No changes are recommended in this respect.
4. Given the guidance provided in the MTR User's Manual and an understanding of the underlying principles behind the method, MTR survey results can be easily communicated to non-biologists for those applications requiring the impact of point-source discharges to be assessed (for example, UWWTD monitoring).
5. For other applications in which cross-catchment comparison of trophic status is required, interpretation of results is more difficult. Mapping of MTR at a catchment or wider scale is not appropriate due to the complexity of factors influencing the MTR. Comparison of the magnitude of downstream changes may be useful, however, in prioritising those areas that would benefit from further investigation or nutrient reduction effort.

It was concluded that the MTR methodology is relatively easy to use and is cost-effective. The main difficulty lies in the interpretation and communication of results. This is relatively straightforward when assessing the downstream impact of point-source nutrient inputs, provided the guidance in the MTR User's Manual is followed. It is more problematic when comparing trophic status between rivers, and hence the MTR is not recommended for such applications at this stage. The future development of a predictive element to the MTR system (such as 'plantpacs', see 7.1.3) is recommended, to overcome these problems.

7.2 Comparison between MTR and other methods

A literature survey of other biological methodologies for assessing the trophic status of rivers, found that the only suitable alternative to MTR was the other principal method under development by the Agency: the Diatom Quality Index (DQI: a derivation of the Trophic Diatom Index or TDI). Thus the comparison of MTR with other methods was restricted to the DQI (see Kelly 1996b, note that an updated guidance standard for diatom sampling methods for European use, is likely to be available mid-1999). Consideration was given to the similarity or advantageous differences of DQI and MTR results over a range of circumstances.

1. DQI and MTR were found to produce similar results.
2. Both systems appear to give a broad assessment of trophic status over the range of circumstances studied. The two methods are likely to be complementary and should be used in parallel. DQI may provide added value when the additional effects of organic enrichment are demonstrable.
3. Practical advantages can be seen for both methods: *either* more rapid field work but more extensive laboratory analysis (DQI); *or* more field time and less laboratory time (MTR).
4. Both methods rely on a field visit at which habitat data should be recorded and which involves a commitment of time, but this visit may be combined with other field survey or sampling such as for water, sediment or macro-invertebrates.
5. The use of the DQI and MTR methods to complement each other could overcome requirements to sample outside the main growth seasons for macrophytes.

It was concluded that wherever possible, the biological assessment of the trophic status of rivers should comprise both DQI and MTR surveys. The two methods are complementary and both provide valuable information. The recently published River Trophic Status Indicator models (Ali et al 1999), which use macrophyte functional variables rather than the species assemblages, may merit further attention as a complementary methodology, but the practical aspects of their application would need to be evaluated before they could be recommended for routine monitoring purposes.

7.3 Recommendations

Recommendations are listed below relating to (i) the operational use of the MTR system, and (ii) research to improve and refine the methodology and to extend the role of macrophytes for river quality assessment.

7.3.1 Operational recommendations

Many recommendations resulting from the MTR evaluation related to where, when and how to undertake MTR surveys. These recommendations have been incorporated into the standard MTR methodology, in the companion volume to this report: the *MTR User's Manual* (Holmes et al. 1999). The manual includes guidance on where and when it is appropriate to use MTR, including its recommended applications; how to undertake the survey in the field; how to process and interpret the survey results; and how to improve the quality of the results. The Manual is intended as 'best practice' standard methodology, applicable throughout the United Kingdom. All MTR surveys should follow the guidance provided in the Manual.

A summary of key operational recommendations incorporated into the manual is as follows.

1. The Mean Trophic Rank system is recommended as suitable to assist in the designation of phosphate-based Sensitive Areas (Eutrophic) under the Urban Wastewater Treatment Directive (UWWTD).
2. Although not yet proven, the MTR system should be equally applicable to the assessment of non-qualifying point discharges.
3. MTR surveys along the length of a river, or throughout a river catchment, may be undertaken, although the influence of factors other than trophic status must be taken into account. Only physically similar sites should be compared.
4. The use of MTR to make comparisons between the trophic status of different rivers is **not** recommended, unless the rivers are of the same physico-chemical type (and even then, results should be treated with caution).
5. *Stigeoclonium tenue* should be removed from the list of scoring species.
6. Ongoing training in plant identification should be undertaken by MTR surveyors. This can include use of reference plant material as well as field visits and attendance on training courses. Particular attention should be paid to the identification of *Ranunculus* species, *Juncus bulbosus*, mosses and rare plants.
7. Ongoing training of surveyors should also include training in the correct application of the method. Emphasis should be placed on the care necessary for estimation of percentage plant cover, location of sparse species, and recognition of 'natural' events (such as 'washout' of species, changes in water quality other than trophic status, or variation in year-to-year flow conditions).

8. When undertaking surveys at the same site over a period of years, surveys should be carried out at the same time of the season. Atypical flow conditions should be avoided.
9. Care should be taken when selecting survey lengths to minimise the physical differences between sites being compared.
10. Surveyors should continue to assess confidence in the MTR survey and the physical comparability of survey sites, using the appropriate suffixes of confidence. The way in which comparability is assessed should be changed in line with the methodology for DQI surveys. Surveyors should follow the guidance in the MTR User's Manual, on how to the use of these suffixes of confidence.
11. Surveyors should continue to assess confidence in the resulting MTR scores, by use of the third suffix of confidence (based on number of highlighted species). Although no conclusive evidence has been found that the highlighted species are more reliable indicators of trophic status, it is recommended that this measure of confidence be retained as an interim measure with the following *proviso*. Achievement of a suffix of 'a' or 'b' (five or more highlighted species) will lend confidence in the results, but achievement of a suffix of 'c' may not necessarily mean that the result is inaccurate. In all cases, emphasis should be placed on obtaining information from as many sources as possible and on drawing conclusions using the balance of evidence available.
12. Wherever possible, both DQI and MTR surveys should be carried out.
13. Quality assurance measures should be adopted to improve the quality of MTR survey data. In addition to integral measures undertaken as part of the survey method itself, surveyors should be adequately trained and re-surveys should be undertaken for quality assurance purposes. Protocols for the resurvey ('audit') surveys are described in the MTR User's Manual.
14. Any change in MTR must be at least 4 units or 15% to be deemed as a significant change in trophic status.
15. The influence of the physical character of the river and its water chemistry on the MTR should be accounted for when interpreting results.
16. Mapping of MTR at a catchment or wider scale is not appropriate due to the complexity of factors influencing the MTR. Comparison of the magnitude of downstream changes may be useful, however, in prioritising those areas that would benefit from further investigation or nutrient reduction effort.

In addition, the following two operational recommendations were made.

17. A link with a more relevant national IDQ aquatic plant examination should be investigated.
18. A mechanism should be derived to allow problems with the MTR methodology to be logged at national level, for future consideration when refining the method and operational guidance; this could operate under the Biology Technical Group for regulatory Agencies.

7.3.2 Recommendations for future method development

The MTR evaluation identified some deficiencies in, and limitations to, the current methodology. To address these, further work is needed. Some of the following recommendations relate to research and development work required, whereas others relate to operational monitoring and others to both. Data from R&D and operational monitoring would be useful in future method development. The recommendations are listed in order of priority, with the highest first:

1. Further research is required to confirm the precise influence on MTR of depth, cross-sectional area, flow category, substrate size, river slope, solid geology, and the altitude of the source. The development of a predictive element to the system is recommended, to address this. One such system, 'plantpacs' (Plant Prediction and Classification System), is currently being scoped in a collaborative Agency/IFE project, (R&D Project No. W1-017). 'Plantpacs' should allow monitoring and classification of trophic status and eutrophication impact for applications other than UWWTD, including the proposed EC Water Framework Directive, national management strategies on eutrophication and individual catchment/river basin management plans. It may contribute to the ecological monitoring of low flows in rivers where associated eutrophication impacts are perceived and should also be useful for detecting other impacts, such as acidification or habitat degradation. Links with other organisations in the UK and the EU should be pursued to assist and enhance future development of 'plantpacs' within a european context.
2. Opportunities should be sought to extend the reference framework of 'un-enriched' sites (especially in lowland areas).
3. To allow evaluation of the performance of MTR at assessing temporal changes in trophic status, it is recommended that MTR monitoring continue, where phosphate reduction measures are commissioned (or have ceased); monitoring is required under the UWWTD.
4. To improve the national applicability of the MTR method, the distribution of STR 1-3 species in Scotland and the possible need for selection of additional STR 2 species for high-energy rivers, should be investigated.
5. Links should be made with other and future macrophyte distribution projects, and use of common survey methodologies should be investigate and directed at:
   a) sampling of plant communities from a wider range of physical substrate;
   b) resolving the STR of rare plants and other candidates for STR change identified in this project;
   c) investigating the distribution of those species which show potential as 'key' or 'indicator' species (either to indicate nutrient enrichment, or to distinguish between phosphate and nitrate enrichment);
   d) re-examining numbers of species present or biodiversity as an alternative measure of trophic status (within 'plantpacs');
   e) examining the effect of weighting factors in MTR for cosmopolitan (& other) species;
   f) examining the effect of nutrient reduction on plant communities.

   In confirming STRs, species cover should be taken into account.
6. MTR sampling at regular intervals along a selection of rivers throughout the UK should be undertaken, to investigate downstream variation in MTR and the influence of diffuse nutrient inputs. The surveys should include major tributaries and sections at which phosphate-reductions measures are being undertaken or proposed. These MTR surveys should be concurrent with regular sampling of water chemistry and should be repeated at intervals of a few years.
7. The practical aspects of the River Trophic Status Indicator model (Ali et al 1999) should be evaluated and compared with MTR and 'plantpacs'.
8. Physical and chemical data collected or assembled should be standardised.
9. In addition to links with projects on aquatic macrophyte distribution, MTR could also be linked to projects on marginal communities (such as Ecofact or Countryside Survey, Bunce at al 1999), or to work currently in hand on plant surveys in relation to the River Habitat Survey. MTR could also form one element of a system to predict the effects of nutrient stripping on the macrophyte community.

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List of Appendices

Appendix 1 Method of estimation of mean for width, depth, substrate size and shade.

A1 Estimation of mean width, depth, substrate size and percentage of site unshaded.

A1.1 Conversion of plant cover data.

The conversion of the macrophyte cover scales to an estimated percentage assumes:

a) every category given a particular cover score has an equal portion of the whole (ie two categories with a 3 each cover 50 percent of the whole);

b) the total cover recorded equals 100 percent.

c) the standard cover scale C is C1 <0.1%; C2 0.1-1%; C3 1-2.5%; C4 2.5-5%; C5 5-10%; C6 10-25%; C7 25-50%; C8 50-75%; and, C9 >75% (Holmes et al 1999).

A1.2 Conversion of 3-point cover scale of physical data to an estimated percentage. (scale points are: '1' = < 5%; '2' = 5-25%; and '3' = >25%)

For each variable (width, depth, etc.):

1. Count the number of categories (<1m,1-5m etc.) with a '3' or a '2'.

2. Estimate the percentage values for each '2' (ie 5-25%) category from Table A1.2.1.

Table A1.2.1. Percentage value for cover 2 determined by the number of twos and threes in the variable.

	Number of 'two's							
No. of 'three's	1	2	3	4	5	6	7	8
	N/A	N/A	N/A	25	20	16	14	12
1	15	15	15	13	11	9	8	7
2	15	14	11	9	7	6	6	5
3	14	9	6	5	N/A	N/A	N/A	N/A

3. The estimated percentage for conversion of the 'three's (ie >25% cover) is then determined by:

Percentage = (100 - x) / (No. of 'three's)

where x = (percentage value for 'two's) x (No. of 'two's)

Note: In some cases the surveyor has entered four or more values of '3' in the substrate category, sometimes as well as covers of '2'. As a cover of '3' is for percentages over 25% this results in an estimated percentage greater than 100%.

4. Covers of '1' are converted to 1%.

A1.3 Conversion of 4-point cover scale for physical data from Conservation Rivers database.

(cover scale points are: '1' = < 5%; '2'= 5-25%; '3' = 25-50%; and, '4' = 50-100%)

The estimated percentage for covers of 'two' (5-25%) and 'three' (25-50%) are calculated as above, with an additional condition to account for any covers of four recorded.

1. When a cover of 'four' is recorded for any of the categories the estimated conversion for any 'three's is set to twenty five percent, the value for conversion of 'two's is determined from Table A1.2.1 (assuming 'two' more 'three's than actually recorded to allow for the four), and the conversion for the 'four' is determined by;
   Percentage = (100 - (25 x No of threes) - x)
   where x = (Percentage value for 'two's) x (No. of 'two's)

2. Covers of '1' are converted to 1%.

A1.4 Conversion of 10-point physical cover scale (from Conservation Rivers database).

1. The estimated percentage cover for conversion of the 10-point scale is determined by reference to Table A1.4.1.

Table A1.4.1. Estimated percentage for conversion of 10 point cover scale.

Cover value	Cover range	Percentage conversion
1	1-10%	3.33%
2	11-20%	13.33%
3	21-30%	23.33%
:	:	:
10	91-100%	93.33%

A1.5 Calculation of mean values for physical data.

The converted cover scores are used to estimate a mean value for width, depth, substrate size (phi) and percentage unshaded.

A1.5.1 Width and depth.

1. The percentage cover in each category is multiplied by the mid point of the range for that category, as given in Table A1.5.1.

Table A1.5.1. Mid point values used to estimate mean width and depth

Width Category (m)	Mid point value (m)
<1	.5
1-5	2.5
5-10	7.5
10-20	15
>20	30

Depth category (m)	Mid point value (m)
<0.25	0.1
0.25-0.5	0.375
0.5-1.0	7.5
>1.0	1.5

2. The resulting values are summed and the total is divided by 100 to give the estimated mean value of depth or width.

A1.6 Mean substrate size.

1. The percentage values in each substrate category are used in the following equation to determine the mean substrate size (phi, as RIVPACS combining equation);

phi = (-7.75 x %boulders/cobbles
- 3.25 x %pebbles/gravel
+ 2 x %sand
+ 8 x % silt/clay)

where the percentage in the separate categories are combined to give percentages of boulders and cobbles, pebbles and gravel etc.

A1.7 Percentage unshaded.

The converted percentage in left bank unshaded and right bank unshaded are averaged to give the total percentage of river that is unshaded.

Appendix 2 Data collated as part of MTR project with percentage of surveys for which each parameter was available. Data are mainly from the EA in England & Wales or DoE, NI.

	EA	IFE	Conservation Rivers	IRTU	RHS Benchmark
No. Sites	523	105	1563	272	110
No Surveys	1655	117	3128	272	110
Survey Length: 100m	94%	100%		100%	
500m	6%		100%		100%
Dates of Surveys	'93-'96	'96	'76-'94	'95	'95-'96
Physical data	92%	97%	97%		
Chemical data (<1km, any year)	58% [a]	58% [a]	25%	100%	
Chemical data (<5km, any year)			69%		
Chemical data (<1km, within 2 years)	58% [a]	58% [a]	3%	100%	
Chemical data (<5km, within 2 years)			10%		
Plant species recorded	All [b]	All	All	All	All
Width (3 point / % cover)	92/41%	97/95%	96% / 0%		
Depth (3 point / % cover)	92/41%	97/85%	97% / 0%		
Substrate (3 point / % cover)	91/41%	94/85%	97% / 0%		
Habitat (3 point / % cover)	89/40%	94/85%	96% / 0%		
Shade (3 point / % cover)	89/40%	97/89%			
Clarity (3 point / % cover)	89/41%	97/97%			
Bed Stability (3 point / % cover)	75/31%	94/86%			
Flow (from gauging stations)	22%	11%			
pH	48%	35%		100%	
Alkalinity	53%	41%		100%	
Conductivity	35%	17%			
Phosphate	55%	50%	25%	100%	
Nitrate	24%	21%	25%	100%	
Potassium	4%				
Sulphate	4%				
Chloride	42%	31%			
Magnesium	10%	3%			
Calcium	10%	3%			
Nitrite	12%	9%		100%	
BOD	49%	32%			
Ammonia	47%	32%			
Suspended Solids	47%	23%			
Dissolved Oxygen	31%	27%			

a Chemical site confirmed on OS maps to match survey point with no tributaries or other inputs between them but may be > 1km apart.

b Survey emphasis on 'scoring' plants but others recorded.

Appendix 3 Summary relating MTRs derived from Conservation Agencies data to river community types

River Community Type [1.]	General description [1.]	Mean MTR[2.]	Mean of top 10%	No. of samples
I	Lowland rivers with minimal gradients. Predominantly in south and east England, but may occur wherever substrates are soft and chemistry enriched	34.0	40.8	49
II	Rivers flowing in catchments dominated by clay	32.9	42.0	429
III	Rivers flowing in catchments dominated by soft limestone such as Chalk and Oolite	40.2	47.3	136
IV	Rivers with impoverished floras, confined to lowlands or eutrophic systems	39.5	58.2	361
V	Rivers of sandstone, mudstone and hard limestone catchments in England and Wales, with similar features to those of Type VI	47.6	65.3	559
VI	Rivers predominantly in Scotland and northern England in catchments dominated by sandstone, mudstone and hard limestone; substrates usually mixed coarse gravels, sands and silts mixed with cobbles and boulders	46.2	59.6	258
VII	Mesotrophic rivers where fine sediments occur with boulders and cobbles, so a mix of bryophytes and higher plants is typical; often downstream from Type VIII communities	52.9	75.0	97
VIII	Oligo-mesotrophic, fast-flowing, rivers, where boulders are common and bryophytes typify the plant assemblages; intermediate, and often between, Types IX and VII	68.1	82.0	537
IX	Oligotrophic rivers of mountains and moorlands where nutrient and base levels low; bedrock, boulders and coarse substrates dominate	68.8	86.2	70
X	Ultra-oligotrophic rivers in mountains, or streams flowing off acid sands; substrates similar to Type IX but often more bedrock	83.0	95.5	327

1. Revised by Holmes et al (1998) from the version previously published in the SSSI selection guidelines (Nature Conservancy Council, 1989).
2. MTRs calculated from 1km sites, each split into consecutive 500m lengths, using a 5-point abundance scale, in England, Scotland and Wales. Data are unpublished but were prepared for the purposes of Holmes (1995). Macrophyte data are courtesy of English Nature, Countryside Council for Wales and Scottish Natural Heritage.
Source of table: Holmes at al (1999)

Appendix 4

The MTR Survey form as used by IFE (slightly reduced & blank).

For new forms see methodology in Environment Agency R&D Technical Report E38 (Holmes et al 1999).

(Record % cover on the 9 point scale given below). Surveyor:

	Cover		Cover		Cover
ALGAE		*Lotus pedunculatus*		*Lemna gibba*	
Batrachospermum sp		*Menyanthes trifoliata*		*Lemna minor*	
Cladophora agg.		*Montia fontana*		*Lemna minuta*	
Enteromorpha sp		*Myriophyllum alterniflorum*		*Lemna trisulca*	
Hildenbrandia rivularis		*Myriophyllum spicatum*		*Phragmites australis*	
Hydrodictyon reticulatum		*Myriophyllum sp.*		*Potamogeton alpinus*	
Lemanea fluviatilis		*Nuphar lutea*		*Potamogeton berchtoldii*	
Stigeoclonium tenue		*Nymphaea alba*		*Potamogeton crispus*	
Vaucheria sp		*Nymphoides peltata*		*Potamogeton freisii*	
LIVERWORTS		*Oenanthe crocata*		*Potamogeton gramineus*	
Chiloscyphus polyanthos		*Oenanthe fluviatilis*		*Potamogeton lucens*	
Jungermannia atrovirens		*Polygonum amphibium*		*Potamogeton natans*	
Marsupella emarginata		*Potentilla erecta*		*Potamogeton obtusifolius*	
Nardia compressa		*Ranunculus aquatilis*		*Potamogeton pectinatus*	
Pellia endiviifolia		*Ranunculus circinatus*		*Potamogeton perfoliatus*	
Pellia epiphylla		*Ranunculus flammula*		*Potamogeton polygonifolius*	
Scapania undulata		*Ranunculus fluitans*		*Potamogeton praelongus*	
MOSSES		*Ranunculus hederaceus*		*Potamogeton pusillus*	
Amblystegium fluviatile		*Ranunculus omiophyllus*		*Potamogeton trichoides*	
Amblystegium riparium		*Ran. penic.* subsp *pseudofluitans*		*Sagittaria sagittifolia*	
Blindia acuta		*Ran. penic.* subsp *penicillatus*		*Schoenoplectus lacustris*	
Brachythecium plumosum		*Ran. penic.* subsp *vertumnus*		*Sparganium emersum*	
Brachythecium rivulare		*Ranunculus peltatus*		*Sparganium erectum*	
Brachythecium rutabulum		*Ranunculus trichophyllus*		*Spirodela polyrhiza*	
Bryum pseudotriquetrum		*Ranunculus sceleratus*		*Typha latifolia*	
Calliergon cuspidatum		*Ranunculus sp.*		*Typha angustifolia*	
Cinclidotus fontinaloides		*Rorippa amphibia*		*Zannichellia palustris*	
Dichodontium flavescens		*Rorippa nasturtium-aquaticum*			
Dichodontium palustre		*Rumex hydrolopathum*		OTHER SPECIES / SAMPLE	
Dicranella palustris		*Veronica anagallis-aquatica*			
Fontinalis antipyretica		*Veronica catenata*			
Fontinalis squamosa		*Veronica scutellata*			
Hygrohypnum luridum		*Viola palustris*			
Hygrohypnum ochraceum		MONOCOTYLEDONS			
Hyocomium armoricum		*Acorus calamus*			
Philonotis fontana		*Alisma plantago aquatica*			
Polytrichum commune		*Alisma lanceolatum*			
Racomitrium aciculare		*Bolboshoenus maritimus*			
Rhynchostegium riparioides		*Butomus umbellatus*			
Sphagnum species		*Carex acuta*			
Thamnobryum alopecurum		*Carex acutiformis*			
VASCULAR CRYPTOGRAMS		*Carex riparia*			
Azolla filiculoides		*Carex rostrata*			
Equisetum fluviatile		*Carex vesicaria*			
Equisetum palustre		*Catabrosa aquatica*		% / Score / Area	
DICOTYLEDONS		*Eleocharis palustris*		<0.1% 1	
Apium inundatum		*Elodea canadensis*		0.1-1% 2	
Apium nodiflorum		*Elodea nuttallii*		1-2.5% 3	
Berula erecta		*Eleogiton fluitans*		2.5-5% 4	
Callitriche hamulata		*Glyceria maxima*		5-10% 5	
Callitriche obtusangula		*Groenlandia densa*		10-25% 6	
Ceratophyllum demersum		*Hydrocharis morsus-ranae*		25-50% 7	
Hippurus vulgaris		*Iris pseudacorus*		50-75% 8	
Nardia compressa		*Juncus bulbosus*		>75% 9	

Physical Records River: NGR:

(Use 3 point scale, 1 = <5%, 2 = 5-25% and 3 = >25%)

Width (m) <1 ___% ☐ 1-5 ___% ☐ >5-10 ___% ☐ >10-20 ___% ☐ >20 ___% ☐

Depth (m) <0.25 ___% ☐ 0.25-0.5 ___% ☐ >0.5-1 ___% ☐ >1.0 ___% ☐

Substrate Bedrock ___% ☐ Boulders ___% ☐ Cobbles ___% ☐ Pebbles ___% ☐ Gravel ___% ☐
Sand ___% ☐ Silt/Mud ___% ☐ Clay ___% ☐ Peat ___% ☐ Not visible ☐

Habitat Pool ___% ☐ Run ___% ☐ Riffle ___% ☐ Slack ___% ☐

Shading: Left Bank None ___% ☐ Broken ___% ☐ Dense ___% ☐
Right Bank None ___% ☐ Broken ___% ☐ Dense ___% ☐

Water Clarity Clear ___% ☐ Cloudy ___% ☐ Turbid ___% ☐

Bed Stability Firm ___% ☐ Stable ___% ☐ Unstble ___% ☐ Soft ___% ☐

Photograph Facing downstream ☐ Facing upstream ☐

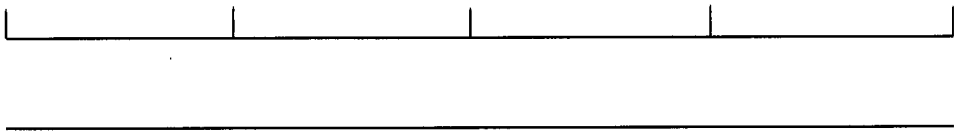
(Enter no. of photos taken in each direction)

Photograph comments _____

Sketch Map 100m ☐ 1-2km site features ☐ (below) (Tick if done)

(Indicate: Location of STW discharge, other inputs, tributaries, weirs and other artificial features, and areas of dense shading.)

u/s ← Length (km) _____ → d/s



Measure of confidence for comparability of u/s and d/s sites (I > 75% similar, II 50-75%, III <50%)

Irradiation (1-5, Excellent - Very Bad) ☐ **Substrate** ☐ **Flow type** ☐ **Depth & width** ☐

Confidence in survey conditions (% of site affected by adverse survey conditions, A < 25%, B 25-50%, C >50%) ☐

Comments _____

Physical impact of STW discharge (1-5, minor to major, + comment) ☐ _____

Plant samples

	No. of samples	Sample codes used (e.g. a-d, 1-4)
Bryophytes	☐	_____
Algae	☐	_____
Others	☐	_____

Chemical Samples

Filtered water ☐ Unfiltered water ☐ Sieved (<2mm) sediment ☐
Subsampled before drying

Plants: _____ ☐ Oven dried at 60°C overnight ☐
(list spp.) _____ ☐ Oven dried at 60°C overnight ☐
_____ ☐ Oven dried at 60°C overnight ☐

Appendix 5

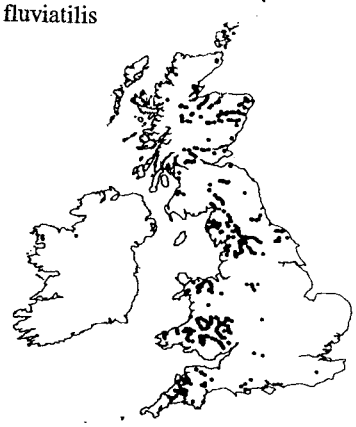
Distribution of MTR scoring species in the Britain and Northern Ireland. (see 3.9)

Distribution of scoring species of algae.

Batrachospermum sp

Hildenbrandia rivularis

Lemanea fluviatilis



Vaucheria spp

Enteromorpha spp

Stigeoclonium tenue

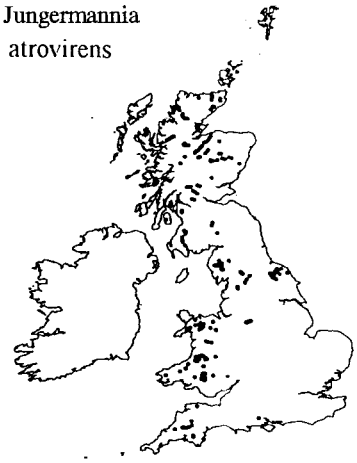
Hydrodictyon reticulatum

Cladophora agg

Distribution of scoring species of liverworts.

Chiloscyphus polyanthos

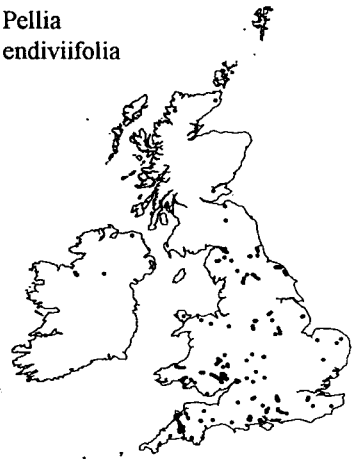
Jungermannia atrovirens



Marsupella emarginata

Nardia compressa

Pellia endiviifolia



Pellia epiphylla

Scapania undulata

Distribution of scoring species of mosses.

Amblystegium riparium

Amblystegium fluviatile

Blindia acuta

Brachythecium plumosum

Brachythecium rivulare

Brachythecium rutabulum

Bryum pseudotriquetrum

Calliergon cuspidatum

Cinclidotus fontinaloides

Distribution of scoring species of mosses.

Dichodontium flavescens

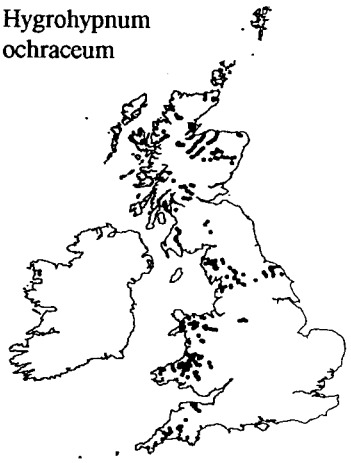
Dichodontium pellucidum

Dicranella palustris

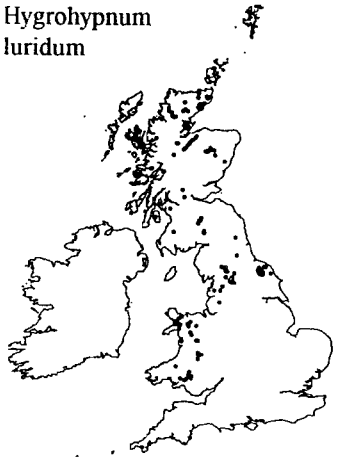
Fontinalis antipyretica

Fontinalis squamosa

Hygrohypnum ochraceum

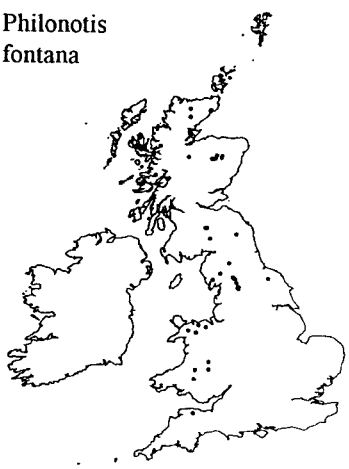


Hygrohypnum luridum



Hyocomium armoricum

Philonotis fontana



Distribution of scoring species of mosses.

Polytrichum commune

Racomitrium aciculare

Rhynchostegium riparioides

Sphagnum species

Thamnobryum alopecurum

Distribution of scoring species of vascular cryptograms and dicotyledons.

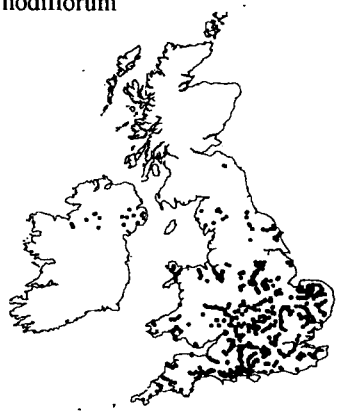
Azolla filiculoides

Equisetum fluviatile

Equisetum palustre

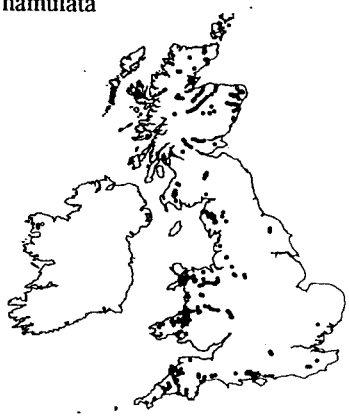
Apium inundatum

Apium nodiflorum



Berula erecta

Callitriche hamulata



Callitriche obtusangula

Ceratophyllum demersum

Distribution of scoring species of vascular cryptograms and dicotyledons.

Hippurus vulgaris

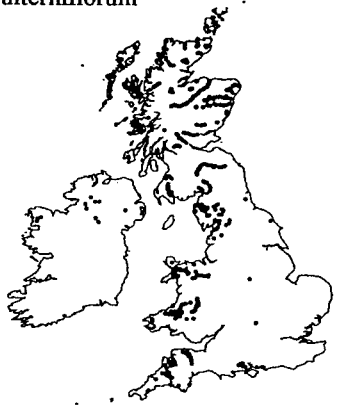
Littorella uniflora

Lotus uliginosum

Menyanthes trifoliata

Montia fontana

Myriophyllum alterniflorum



Myriophyllum spicatum

Myriophyllum spp

Nuphar lutea

Distribution of scoring species of vascular cryptograms and dicotyledons.

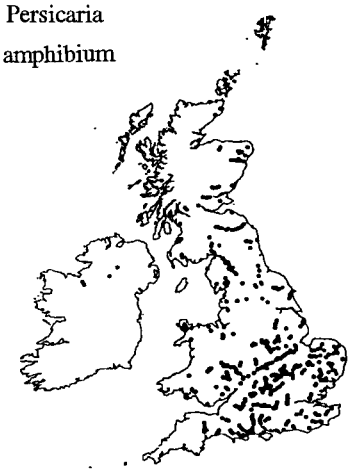
Nymphaea alba

Nymphoides peltata

Oenanthe crocata

Oenanthe fluviatilis

Persicaria amphibium

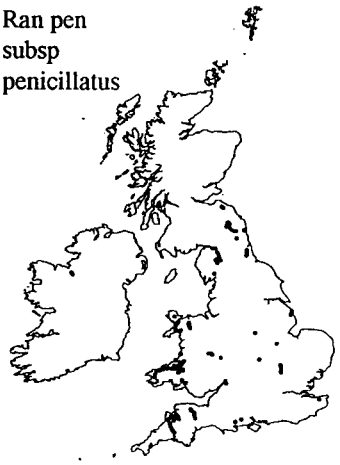


Potentilla erecta

Ranunculus aquatilis

Ran pen subsp pseudofluitans

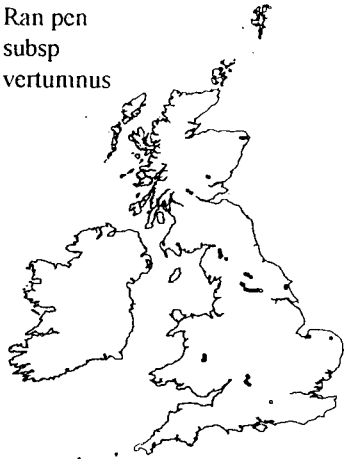
Ran pen subsp penicillatus



The distribution of Ranuculus penicillatus species are by sub-species only.

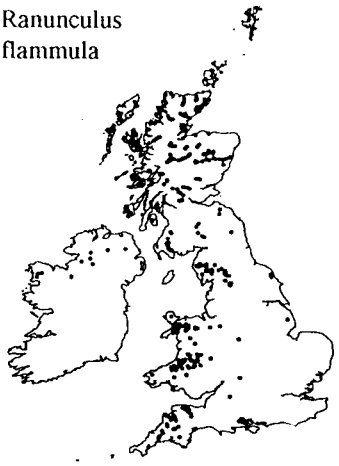
Distribution of scoring species of vascular cryptograms and dicotyledons.

Ran pen subsp vertumnus

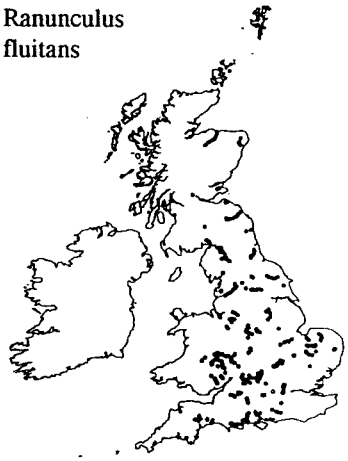


Ranunculus circinatus

Ranunculus flammula



Ranunculus fluitans



Ranunculus hederaceus

Ranunculus omiophyllus

Ranunculus peltatus

Ranunculus trichophyllus

Ranunculus sceleratus

Distribution of scoring species of vascular cryptograms and dicotyledons.

Ranunculus spp

Rorippa amphibia

Rorippa nasturtium-aquaticum

Rumex hydrolopathum

Veronica anagallis-aquatica

Veronica catenata

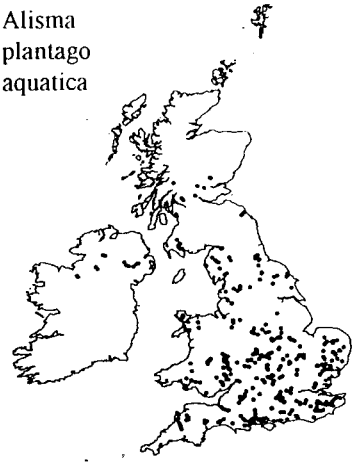
Veronica scutellata

Viola palustris

Distribution of scoring species of monocotyledons.

Acorus calamus

Alisma plantago aquatica



Alisma lanceolatum

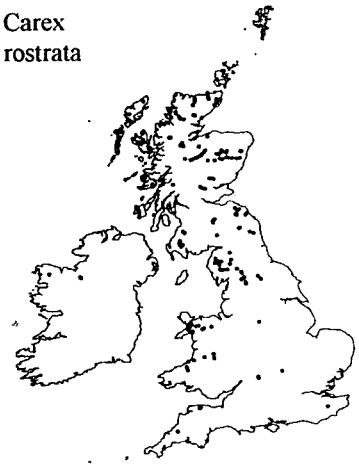
Butomus umbellatus

Carex acuta

Carex acutiformis

Carex riparia

Carex rostrata

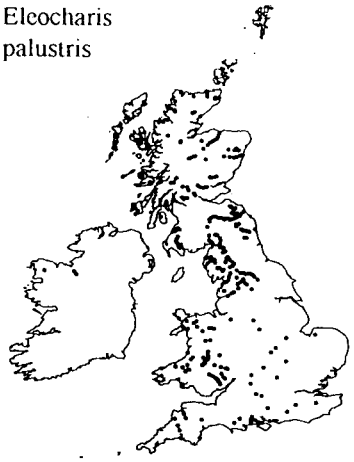


Carex vesicaria

Distribution of scoring species of monocotyledons.

Catabrosa aquatica

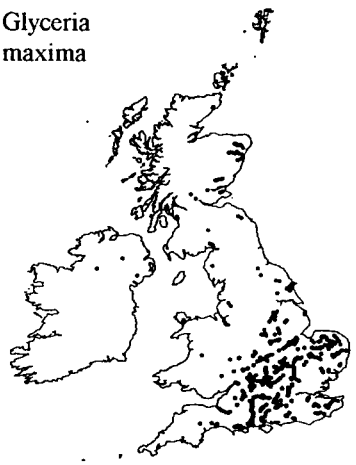
Eleocharis palustris



Elodea canadensis

Elodea nuttallii

Glyceria maxima



Groenlandia densa

Hydrocharis morsus-ranae

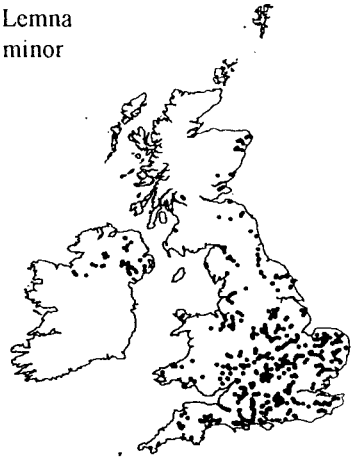
Iris pseudacorus

Juncus bulbosus

Distribution of scoring species of monocotyledons.

Lemna gibba

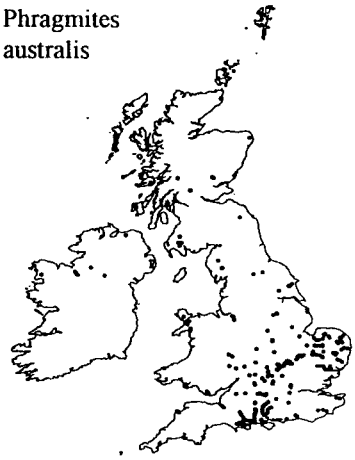
Lemna minor



Lemna minuta

Lemna trisulca

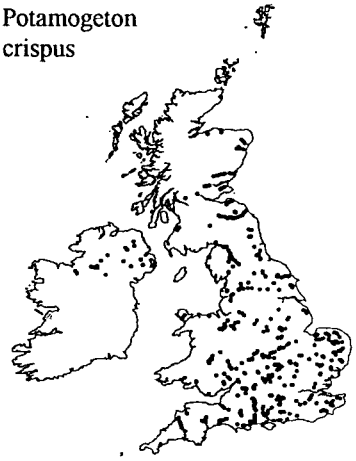
Phragmites australis



Potamogeton alpinus

Potamogeton berchtoldii

Potamogeton crispus



Potamogeton freisii

Distribution of scoring species of monocotyledons.

Potamogeton gramineus

Potamogeton lucens

Potamogeton natans

Potamogeton obtusifolius

Potamogeton pectinatus

Potamogeton perfoliatus

Potamogeton polygonifolius

Potamogeton praelongus

Potamogeton pusillus

Distribution of scoring species of monocotyledons.

Potamogeton trichoides

Sagittaria sagittifolia

Eleogiton fluitans

Schoenoplectus lacustris

Bolboschoenus maritimus

Sparganium emersum

Sparganium erectum

Spirodela polyrhiza

Typha latifolia

Distribution of scoring species of monocotyledons.

Typha angustifolia

Zannichellia palustris

Appendix 6

Table A6 List of Mean Trophic Rank Scoring Taxa

Notes:

1. Authorities are not included for the purposes of brevity, except where to omit the authority would cause confusion.
2. Recent synonyms are included (in italics). These are based upon Stace (1991), with in addition: Kent (1992) for check list of vascular plants, Smith (1990, 1991) for bryophytes and Jermy et al (1982) for sedges. Previous genus name abbreviated as 'g.'
3. 'h' denotes that hybrids of the species indicated should be included in the taxon for scoring purposes.
4. The codes are taken from Holmes, Whitton and Hargreaves (1978) and Whitton, Holmes and Sinclair (1978).
5. Species selected for national aquatic plant identification test (id) found in this list.
6. Highlighted 'species' are given in bold type.
7. *Stigeoclonium tenue* is included in this list, but one of the reccomendations from this project is that it be removed (see 3.4.1).

ALGAE	**STR**	**comments and code number**
Batrachospermum species	6	0202, id
Hildenbrandia rivularis	6	020601, id
Lemanea fluviatilis	7	020701, id
Vaucheria species	1	0914
Enteromorpha species	1	1715, id
[Stigeoclonium tenue][7]	[1]	173205
Hydrodictyon reticulatum	3	162201, id
Cladophora aggregate	1	all species except *C. aegagropila* & *C. rhizoclonium* 1901, id

LIVERWORTS		
Chiloscyphus polyanthos v	8	341102, id
Jungermannia atrovirens *Solenostoma triste*	8	345508
Marsupella emarginata v	10	343402, id
Nardia compressa ***Alicularia compressa***	10	343701, (id)
Pellia endiviifolia *P. fabbroniana*	6	344101, id
Pellia epiphylla	7	344102, id
Scapania undulata	9	345410, (id)

MOSSES		
Amblystegium fluviatile *g. Hygroamblestegium*	5	320201, (id)
Amblystegium riparium *g. Hypnum or Leptodictyum*	1	
Blindia acuta	10	320901
Brachythecium plumosum	9	321001
Brachythecium rivulare	8	321002, id
Brachythecium rutabulum	3	321003
Bryum pseudotriquetrum	9	321212
Calliergon cuspidatum *g. Acrocladium Calliergonella or Hypnum*	8	321302
Cinclidotus fontinaloides	5	321802, id
Dichodontium flavescens	9	322501
Dichodontium pellucidum	9	322502
Dicranella palustris *D. squarrosa*	10	322604
Fontinalis antipyretica	5	323401, id
Fontinalis squamosa	8	323402
Hygrohypnum ochraceum ***g. Hypnum***	9	323905, id
Hygrohypnum luridum ***Hypnum palustre***	9	323903, id
Hyocomium armoricum ***H. flagellare***	10	324001
Philonotis fontana	9	325404
Polytrichum commune	10	326201
Racomitrium aciculare	10	326601, id
Rhynchostegium riparioides *g. Eurynchium*	5	326902, id
Sphagnum species	10	3274, id
Thamnobryum alopecurum	7	327801, id

FERNS & HORSETAILS		
Azolla filiculoides	3	350101, id
Equisetum fluviatile h	5	350202, id
Equisetum palustre h	5	350204, id

DICOTYLEDONS		
Apium inundatum	9	360402
Apium nodiflorum h *g. Sium*	4	360403
Berula erecta *g. Sium or Siella*	5	360801
Callitriche species indeterminate	no score	3611
Callitriche hamulata *C. intermedia ssp hamulata*	9	361103
Callitriche obtusangula	5	361105
Ceratophyllum demersum	2	361401
Hippuris vulgaris	4	363201
Littorella uniflora	8	363901
Lotus pedunculatus *L. uliginosus*	8	suggested code number 370003
Menyanthes trifoliata	9	364701
Montia fontana	8	365001
Myriophyllum alterniflorum	8	365401
Myriophyllum spicatum	3	365403
Myriophyllum species indeterminate	6	3654, BUT NOT *M. aquaticum (M. brasiliense)*
Nuphar lutea h	3	365501
Nymphaea alba	6	365601
Nymphoides peltata *Limnanthemum*	2	365701
Oenanthe crocata *O. phellandrium*	7	365802
Oenanthe fluviatilis	5	365804
Persicaria amphibia *Polygonum amphibium*	4	366501
Potentilla erecta h *g. Tomentilla or Comarum*	9	366702
Ranunculus species indeterminate h	6	3669
Ranunculus aquatilis	5	366901
Ranunculus circinatus	4	366903
Ranunculus flammula	7	366904
Ranunculus fluitans h	7	366906
Ranunculus hederaceus	6	366907
Ranunculus omiophyllus h *R. lenormandii*	8	366909
Ranunculus peltatus h ***R. aquatilis ssp. peltatus***	4	366911
Ranunculus penicillatus h	5	3669## Use this category for historical data when subspecies/ variety has not been recorded.
Ranunculus penicillatus h subspecies penicillatus	6	366913

Ranunculus penicillatus h **subspecies pseudofluitans** *R. pseudofluitans* *R. aquatilis v.pseudofluitans* *R. peltatus v. pseudofluitans* **- variety calcareus** *R. penicillatus v. calcareus* **- variety vertumnus** *R. penicillatus v. vertumnus*	5 5 5	366912 3669## 366914
Ranunculus trichophyllus h	6	366918
Ranunculus sceleratus	2	366917
Rorippa amphibia h *g. Nasturtium or Sisymbrium*	3	367101
Rorippa nasturtium-aquaticum ***Nasturtium officinale*, or** ***g. Sisymbrium***	5	367905
Rumex hydrolapathum h	3	367303
Veronica anagallis-aquatica or V. catenata indeterminate or hybrid	4	3698 Hybrids of *V. anagallis -aquatica* or *V. catenata*, or indeterminate forms of these species
Veronica anagallis-aquatica	4	369801
Veronica catenata ***V. aquatica***	5	369803
Veronica scutellata	7	369804
Viola palustris	9	369901

MONOCOTYLEDONS		
Acorus calamus	2	380101
Alisma plantago-aquatica h	3	380303
Alisma lanceolatum	3	380302
Bolboschoenus maritimus *g. Scirpus maritimus or Schoenoplectus*	3	384505
Butomus umbellatus	5	380301
Carex acuta h *C. gracilis* Curtis	5	381101
Carex acutiformis h	3	381102
Carex riparia h	4	381128
Carex rostrata h *C. ampullacea*	7	381129
Carex vesicaria h *C. inflata*	6	381131
Catabrosa aquatica *g. Aira*	5	381201
Eleocharis palustris h *g. Scirpus*	6	382004
Eleogiton fluitans ***g. Scirpus or Isolepis***	10	384502
Elodea canadensis ***g. Anacharis***	5	382101
Elodea nuttallii ***g. Anacharis or Hydrilla!***	3	382103
Glyceria maxima	3	382503
Glyceria notata/fluitans/ declinata *G. notata = G. plicata*	no score	(382504/02/01 respectively)

Groenlandia densa *g. Potamogeton densus*	3	382601
Hydrocharis morsus-ranae	6	382701
Iris pseudacorus	5	382901
Juncus bulbosus	10	383066
Lemna gibba	2	383301
Lemna minor	4	383302
Lemna minuta *L. minuscula*	3	383305 NEW NUMBER
Lemna trisulca	4	383304
Phragmites australis *P. communis*	4	383801
Potamogeton indeterminate	no score	3840
Potamogeton alpinus	7	384002
Potamogeton berchtoldii h	4	384003
Potamogeton crispus	3	384006
Potamogeton freisii h	3	384009
Potamogeton gramineus h	7	384010
Potamogeton lucens h	3	384011
Potamogeton natans h	5	384012
Potamogeton obtusifolius	5	384014
Potamogeton pectinatus	1	384015
Potamogeton perfoliatus	4	384016
Potamogeton polygonifolius h	10	384017
Potamogeton praelongus h	6	384018
Potamogeton pusillus h *P. panormitanus*	4	384019
Potamogeton trichoides h	2	384021
Sagittaria sagittifolia	3	384202
Schoenoplectus lacustris h *Scirpus lacustris*	3	384504
Sparganium emersum h *S. simplex*	3	384602
Sparganium erectum *various subspecies*	3	384603
Spirodela polyrhiza *g. Lemna*	2	383303
Typha latifolia h	2	384902
Typha angustifolia	2	384901
Zannichellia palustris	2	385201
(mean STR score)	**(5.5)**	

Appendix 7. Calculation of MTR - example

7.1 Calculation of Mean Trophic Rank

Selected, usually common, aquatic macrophytes have been assigned a number from 1 - 10 according to their tolerance/preference for enriched or un-enriched waters; this is the Species Trophic Rank (STR). The STR for each selected taxa can be found in Appendix 6.

Mean Trophic Ranks (MTR) are only to be calculated for 100m survey lengths. All scoring species should be included in the calculation of the MTR, but non-scoring species should be excluded.

MTR scores are calculated as follows:

1. For all scoring taxa recorded, multiply the Species Trophic Rank (STR) by the Species Cover Value (SCV) to give a Cover Value Score (CVS) for each scoring species.

   Example

Species	STR		SCV		CVS
Enteromorpha sp(p)	1	x	1	=	1
Cladophora agg.	1	x	1	=	1
Nuphar lutea	3	x	1	=	3
Lemna minor	4	x	7	=	28
Potamogeton pectinatus	1	x	1	=	1
Mentha aquatica	-	x	(2)	=	-
Zannichellia palustris	2	x	1	=	2
Amblystegium riparium	1	x	1	=	1
Ranunculus fluitans	7	x	7	=	49
TOTAL			20		86

2. Add up all the numbers in the CVS column.

3. Add up the numbers in the SCV column associated with scoring species ONLY. Do not include non-scoring species in this calculation.

4. Divide the total score for the CVS by the total of the SCV and multiply by 10 to give the Mean Trophic Rank (MTR).

eg

MTR = (sum of CVS ÷ sum of SCV) x 10

In above case: (86 ÷ 20) x 10 = **43.0**

Present MTR score to ONE decimal place only

NB Where only non-scoring species are present in the survey length, there is no MTR for the survey. An MTR value of 'zero' may be recorded for data archiving purposes but this value must ***not*** *be used to indicate trophic status.*