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The use of pressure-response relationships between nutrients and biological quality elements: A method for establishing nutrient supporting element boundary values for the Water Framework Directive

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Foreword

The Water Framework Directive requires Member States (MS) to follow an intercalibration process to ensure comparability of status class boundaries (specifically the Good/Moderate boundary) for biological quality elements (BQEs). This process is well established, and has been successfully followed by many MS for a range of BQEs. However, concerns have been raised that an apparently wide range of nutrient boundary values have been established by MS to support good ecological status. ECOSTAT has initiated a project to investigate this issue. The work is being led by UK (Freshwaters), Germany (Saline waters) and JRC. The aim of the work is to investigate and establish the reasons for any differences between MS in the development and application of nutrient boundaries, leading to the production of best practice guidance.

One of the recommendations from the work on freshwaters was to compare boundary values with pressure response relationships using information gathered during the intercalibration exercise and this report addresses this issue.

This work is being co-ordinated by the steering group members listed below:

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Abstract

The Water Framework Directive requires nutrient boundary concentrations to be established as part of the assessment of ecological status. In this report we use data and relationships developed during the intercalibration exercise for lakes and national monitoring data for rivers to determine ranges of potential nutrient (N & P) boundary concentrations at the intercalibrated boundaries for high/good and good/moderate biological status.

Where data were available we compared the use of different regression models, including multivariate (N+P), and both type I and type II univariate (N or P) models. We suggest that the most appropriate statistical approach is to use either multivariate ordinary least squares (OLS) regression with both N and P as predictor variables or, for univariate relationships, to use type II regression, as the slope of a conventional ordinary least squares regression is likely to be underestimated unless model uncertainty is low, resulting in incorrect predicted boundary values.

We also used two categorical methods to determine boundary values. Firstly, by calculating the distribution of mean nutrient concentrations for water bodies categorised by biological status. Secondly, we developed a method to determine the nutrient concentration at which the mis-match between biological and nutrient status was minimised. Both methods produced boundary values that were similar to those from regression models.

We compiled the results from all of these approaches, together with uncertainty estimates, to provide ranges for the "most likely" and "possible" ranges of boundary values for intercalibration and broad water body types.

For many relationships, particularly in rivers, uncertainty was relatively high, with nutrient concentration typically accounting for only 35-45% of variability. As a result of this uncertainty the range of boundary values that might be predicted if a different, but similar (for example water body type) data set were used was relatively high.

We were only able to use data or published relationships from a limited range of lake and river intercalibration types, but comparing the resulting boundary values to those currently being used by Member States, we demonstrate that in most cases the majority of national boundary values fall within the range of predicted values if uncertainty is taken into consideration.

Given the high degree of variability in the relationships between nutrients and biological status we suggest that further discussion and guidance is needed on how they can be used to support the objectives of the WFD as it is clear that even for well-defined water body types a range of values occur in water bodies that are considered to be in good status according to the most sensitive biological quality element.

1 Introduction

1.1 Background

To achieve good ecological status under the Water Framework Directive (WFD) the directive specifies that "nutrient concentrations do not exceed the levels established so as to ensure the functioning of the ecosystem and the achievement of the values specified for the biological quality elements" (WFD Annex V, Section 1.2). Member states thus need to establish the concentrations of nutrients that meet this requirement. A review of these values has recently been carried out which has revealed a relatively wide range of concentrations currently being used (Phillips and Pitt 2015). To provide greater clarity on the range of nutrient concentrations that might be considered to support good ecological status this work has used data collected during the intercalibration of ecological status, or where these data were not available national data sets, to examine relationships between nutrients and biological status.

The CIS guidance on eutrophication assessment (European Commission 2009b) outlines potential methods of establishing nutrient standards, which should be linked to the setting of biological boundaries for ecological assessment. In this report we explore the use of methods to achieve this, particularly the use of regression to quantify pressure-response relationships between nutrients and biological ecological quality ratios (EQRs). From such a relationship it should be possible to quantify the nutrient concentration at a particular EQR value which can be used to establish appropriate nutrient levels that support good ecological status.

The report summarises relationships to explore the issues associated with the use of regression models. It also compares the results with values determined using categorical analysis. A detailed account is provided for one lake type, shallow high alkalinity lakes, to illustrate the approach. The results of the analysis of other types are summarised with further details in an appendix.

2 Approach and methods used

2.1 Choice of regression approach, type I or type II models

Regression models allow the relationship between nutrients and biological status to be established. However, one of the issues with the use of regression is that ordinary least squares regression (OLS) minimises the variation in the dependent variable and thus assumes no uncertainty in the predictor variable. This is often the case for experimental studies, but unlikely to be so when using data from monitoring programmes such as are used for the WFD. Thus, when using OLS regression to quantify the relationship between nutrient concentration and biological status we have to make a choice concerning whether biological status (EQR) or nutrient concentration are considered the dependent variable. The choice of the dependent variable is important as where both variables contain error an OLS regression will underestimate to slope of the relationship (Legendre 2008) and thus influence the nutrient concentration we determine for the biological boundary.

As the purpose of the model is to predict the nutrient concentration that occurs at a given ecological status, for example the good/moderate boundary, it might be logical to make the dependent (y) variable nutrient concentration, with biological status as the independent (x) variable. However, when considering the relationship between nutrients and biological status we generally assume that the nutrient concentration "causes" the ecological status, which is why we seek to establish the nutrient concentrations that will support good status. Thus it is also logical to make the dependent variable biological status, predicted from nutrient status, with boundary values subsequently determined by re-arranging the regression equation. However, the fact that nutrient concentrations are also influenced by the biology through uptake should not be completely ignored.

The choice of regression approach depends on the degree of asymmetry in the relative uncertainty of the dependent and predictor variable (McArdle 2003; Smith 2009). It is clear that estimates of both the biological EQR and nutrient concentration will contain error due to sampling, however this is not the only source of uncertainty we need to consider. In addition to the uncertainty associated with sampling regimes, the uncertainty in the relationship between nutrients and biology, sometimes called equation error, also needs to be taken into account (McArdle 2003). As other environmental factors also influence the biology the relationship between nutrients and biology is likely to be asymmetric in relation to uncertainty, as equation error will increase the error of the EQR. Thus, it is not unreasonable to conclude that the total uncertainty in the biological EQR is greater than that of nutrients. However, the issue is whether it is "much greater", as required for the use of OLS regression. Where R^2 values are high (>0.6) there is little practical difference in the nutrient boundaries resulting from a regression of EQR on nutrient or nutrient on EQR, but for less certain relationships the differences are more substantial.

The alternative is to use a type II regression (Sokal and Rohlf 1995), where the fitting procedure minimises the variation of both dependent and independent variables. The disadvantage of a type II regression is that it is less appropriate where the purpose of the model is to make predictions (Legendre and Legendre 2012), it is more difficult to interpret uncertainty (Smith 2009), is less easily available in statistical software and it can only be used with a single predictor variable. It is also important to only apply type II regression to relationships with a significant correlation. (Smith 2009) suggests a critical value of at

least 0.6 (R^2 = 0.36), as the method will generate a line with a slope significantly different from zero from random data.

In the analysis reported here we apply both conventional type I OLS regression, using both nutrient and biological status as the dependent variables in turn and a type II regression, presenting a range of predicted nutrient concentrations at the good/moderate and high/good boundaries.

2.2 Data and analysis method

Data collated for the intercalibration of phytoplankton, macrophytes, phytobenthos (most countries assume diatoms as proxies for phytobenthos, but some use non diatom methods, see Poikane et al. (2016)) and invertebrates for lakes and large rivers from Central Baltic, Northern and Cross GIGs, supplemented by some additional national river macrophyte and phytobenthos data sets were used for the analysis (see Figure 3-4 for illustration of method). The EQR's used were the benchmark standardised common metrics or for the additional river data national EQRs normalised by linear transformation from national to standard EQR boundary values (0.8, 0.6, 0.4, and 0.2). Nutrient concentrations were growing season or annual mean total phosphorus (TP) and total nitrogen (TN) values for each lake or lake/year. For rivers, soluble P ("orthophosphate-P", "soluble reactive P") was used.

The approach was to fit regression lines using OLS with both nutrient and EQR as dependent variables in turn and additionally to fit a line using reduced major axis (RMA) regression, the most commonly recommended alternative to OLS (Legendre and Legendre 2012). For graphical display, where nutrients were the dependent variable the regression equations were algebraically re-arranged so that the slope was directly comparable with the approach where EQR was the dependent variable. After fitting relationships, the value of the nutrient concentration was determined using the intercalibrated common metric boundary values or for national data sets the normalised EQR boundaries (0.80 & 0.60). Univariate regression models were fitted for TP and TN independently and for lakes also in combination using multivariate OLS. As not all water bodies had data for both N and P a separate multivariate analysis was carried out to maximise the number of points for the univariate analysis. The significance of including both TP and TN in the model was assessed using AIC comparing univariate and multivariate models from this reduced data set.

The multivariate analysis results in an infinite range of potential TN and TP concentrations at the specified boundary EQR values. These are presented as contour lines overlaid on a scatter plot of mean TP v mean TN. The values identified as boundaries were those where the contour line intersected with an RMA regression line fitted to the relationship between TN and TP.

In all cases uncertainty in the predicted nutrient boundary values is derived from the upper and lower quartiles of the residuals of the regression lines. Thus the range of boundary values will contain 50% of the observed data and the most likely value associated with a particular status will be given by the regression line. All statistical analysis was carried out with R, RMA was fitted using the lmodel2 package (Legendre 2011).

For lakes the protocol used for the analysis was to initially identify outliers using scatter plots and to exclude these from subsequent analysis by marking the data set. To maximise the number of data points for the univariate analysis records for TP and TN were marked

for exclusion independently. Linearity was then assessed using a combination of GAM models (mgcv package) and segmented regression (segmented package). The significance of potential break points were determined using the Davies test. For the univariate analysis only linear parts of the relationship were used for fitting regressions.

2.3 Categorical analysis

Categorical analysis provides an alternative and potentially simpler method of analysis which is not dependent on establishing a statistically significant modelled relationship. Two different approaches were used, the first used the distribution of nutrient concentration in classified water bodies, and the second identified the nutrient boundary that minimised the difference between classifications based on biological and supporting element classifications.

2.3.1 Distribution of concentration by biological class

Boxplots of the distribution of nutrient concentrations by biological class were produced. Potential nutrient boundary values were determined by averaging quantiles of adjacent classes. Two methods were used to determine boundary values.

1. The first approach was to average the higher class 75th percentile (or 0.75 quantile) with the lower class 25th percentile (or 0.25 quantile). The logic being that for the good/moderate boundary this was the average of the highest common (<75%) nutrient concentration associated with Good status and the lowest common (>25%) nutrient concentration of Moderate status.
2. The second approach was to average the 0.25 and the 0.75 quantiles respectively of adjacent classes, the logic being that this was the mid-point of conditions in the good and moderate classes. This approach has the advantage of providing a potential range of boundary values, by using the averaged upper and lower quartiles of the distribution.

2.3.2 Mismatch of biological and nutrient classifications

A second approach was to minimise the mismatch in biological and nutrient classifications using discrete steps of nutrient boundary values. This was a variation of a method proposed in the CIS guidance on eutrophication assessment (European Commission 2009a) which proposed looking at the proportion of water bodies where both biology and supporting element were in good status. The analysis was carried out using Excel. Data were arranged to provide a series of nutrient classifications using a logarithmic series of potential nutrient boundary values. Both biology and nutrients were recorded using a binary classification, for example "good or better" and "moderate or worse". The resulting percentage of misclassified water bodies where biology was good or better, but nutrients were moderate or worse were compared with the opposite form of misclassification where biology was moderate or worse but nutrient good or better. The results were displayed graphically by plotting the percentage of misclassification against the nutrient boundary concentration used. The point where the two forms of misclassification intersected was identified as the minimum mismatch and the nutrient concentration determined. Analysis was carried out for both the high/good and good/moderate boundaries.

2.3.3 Summarising results

The regression models and boxplot approaches provide estimates of uncertainty. Thus the nutrient boundary value predicted by the regression line represents the "most likely" concentration that occurs at the biological good/moderate boundary. At this value 50% of sites at good ecological status would have lower and 50% higher nutrient concentrations. Alternatively, higher or lower values can be derived, using the confidence limits of the predicted line, where more or fewer sites at good ecological status would have lower nutrient concentrations. The use of a lower concentration as a boundary value would ensure that more sites were likely to be at good status if this value were achieved. This precautionary approach however, would also result in more than 50% of sites being at good ecological status despite nutrient concentrations being higher than the boundary value. Ultimately the choice of approach is dependent on the way that boundary values are used to support water management, but as ecological status is assessed as the worse of both biological and supporting elements, the CIS guidance on classification (European Commission 2005) points out that these levels need to be established so that they are no more or less stringent than required by the WFD and hence do not cause water bodies to be wrongly downgraded to moderate status. This implies that the most appropriate approach is to use the regression line rather than an upper or lower confidence limit.

In our analysis the methods used provide a range of potential nutrient boundary values for each BQE/type combination. The results are tabulated in the appendix but have also been summarised in the main text in the following way.

a) A range for the "most likely" boundary value derived from the minimum and maximum value predicted from the different regression and categorical approaches.
b) The boundary value from the "best" regression model, together with a range defined by the upper and lower quartiles of the residuals of the regression. The "best" regression was defined as the one with the highest R^2 value or for the univariate analysis was the RMA regression.
c) The maximum range of values suggested by the analysis, derived from the minimum and maximum values of the upper and lower quartiles of the regressions or categorical analysis.

These results are compared with the range of values reported for the lake/river type by member states. It is important to note that some member states may have used larger data sets when determining national boundary values and that as a consequence they are likely to have more robust relationships and potentially a wider range of residuals that would influence their selected boundaries.

3 Results for lakes

3.1 High Alkalinity Shallow Lakes (IC type L-CB1)

To facilitate understanding of the methods used and the implications of using different approaches to regression, this section describes in detail the results obtained for high alkalinity shallow lakes. Subsequent sections provide summaries of results for other lake types.

3.1.1 Univariate regression models

Relationships between TP and the common metric for phytoplankton are shown in Figure 3-1. The OLS regression relationship is linear where TP < 100µgl^{-1} but the gradient is steeper when the uncertainty of TP is minimised in comparison to when the biological EQR is minimised (compare Figure 3-1 a & b). The RMA regression slope is intermediate and given that the R^2 (0.53) is substantially greater than the threshold value of 0.36 provides the best unimodal modelled relationship from which boundary values can be predicted, a value for the good/moderate boundary of 39 µgl^{-1} with 50% of the results having values between 28-51 µgl^{-1} (Table 3-1). The relationship between TP and common metric for macrophytes was only linear from 40 µgl^{-1}. The R^2 was highly significant but lower than that for phytoplankton (R^2=0.43 p<0.001) resulting in a larger difference in gradients for the OLS regressions (Figure 3-2 a & b). This was above the critical threshold and the RMA regression predicted a good/moderate boundary value of 64 µgl^{-1} with a range of 46-93 µgl^{-1}.

The univariate relationships for TN had lower R^2 values than those for TP and the value for macrophytes was higher than that for phytoplankton (Table 3-2). However, the R^2 values were below the critical threshold and thus less reliable for predicting boundary values.

Table 3-1 Predicted total phosphorus boundary values for high alkalinity shallow lakes (L-CB1) using regression models and categorical methods

IC Type	Phytoplankton Models	R^2	nutrient range TP µgl^{-1}	GM TP µgl^{-1}			HG TP µgl^{-1}		
				Pred	25th	75th	Pred	25th	75th
LCB1	EQR v TP + TN (OLS)	0.55	4 - 100	40	28	57	22	15	32
	EQR v TP (OLS)	0.53	4 - 91	41	28	60	22	15	32
	TP v EQR (OLS)		4 - 91	35	26	48	25	18	34
	EQR v TP (RMA)		4 - 91	39	28	51	23	17	31
	Average adjacent quartiles			44			24		
	Average adjacent classes			40	30	61	23	18	37
	Minimise class difference			40			32		

IC Type	Macrophyte Models	R^2	nutrient range TP µgl^{-1}	GM TP			HG TP		
				Pred	25th	75th	Pred	25th	75th
LCB1	EQR v TP + TN (OLS)	0.40	10 - 597	45	24	82	15	8	30
	EQR v TP (OLS)	0.43	41 - 597	59	41	97	26	18	43
	TP v EQR (OLS)			73	50	102	51	35	72
	EQR v TP (RMA)			64	46	93	34	24	50
	Average adjacent quartiles			39			31		
	Average adjacent classes			47	25	68	31	20	44
	Minimise class difference			45			21		

Table 3-2 Predicted total nitrogen boundary values for high alkalinity shallow lakes (L-CB1) using regression models and categorical methods, predictions from models where $R^2 < 0.36$ shown in grey type as potentially less reliable.

IC Type	Phytoplankton Models	R^2	nutrient range TN mgl^{-1}			GM TN mgl^{-1}			HG TN mgl^{-1}		
						Pred	25th	75th	Pred	25th	75th
LCB1	EQR v TP + TN (OLS)	0.55	0.11	-	3.00	1.05	0.75	1.50	0.60	0.43	0.85
	EQR v TN (OLS)		0.11	-	1.58	1.04	0.54	1.81	0.55	0.28	0.95
	TN v EQR (OLS)	0.28	0.11	-	1.58	0.85	0.69	1.07	0.71	0.58	0.90
	EQR v TN (RMA)		0.11	-	1.58	0.92	0.65	1.29	0.65	0.46	0.91
	Average adjacent quartiles					1.06			0.77		
	Average adjacent classes					0.97	0.73	1.36	0.81	0.58	1.09
	Minimise class difference					0.76			0.61		

IC Type	Macrophyte Models	R^2	nutrient range TN mgl^{-1}			GM TN mgl^{-1}			HG TN mgl^{-1}		
						Pred	25th	75th	Pred	25th	75th
LCB1	EQR v TP + TN (OLS)	0.40	0.22	-	6	1.05	0.58	1.75	0.40	0.22	0.70
	EQR v TN (OLS)					1.17	0.77	1.93	0.55	0.37	0.92
	TN v EQR (OLS)	0.31	0.8	-	6.39	1.44	1.12	1.78	1.14	0.89	1.42
	EQR v TN (RMA)					1.27	0.94	1.78	0.75	0.56	1.05
	Average adjacent quartiles					1.10			0.79		
	Average adjacent classes					1.03	0.69	1.53	0.82	0.57	1.20
	Minimise class difference					0.90			0.49		

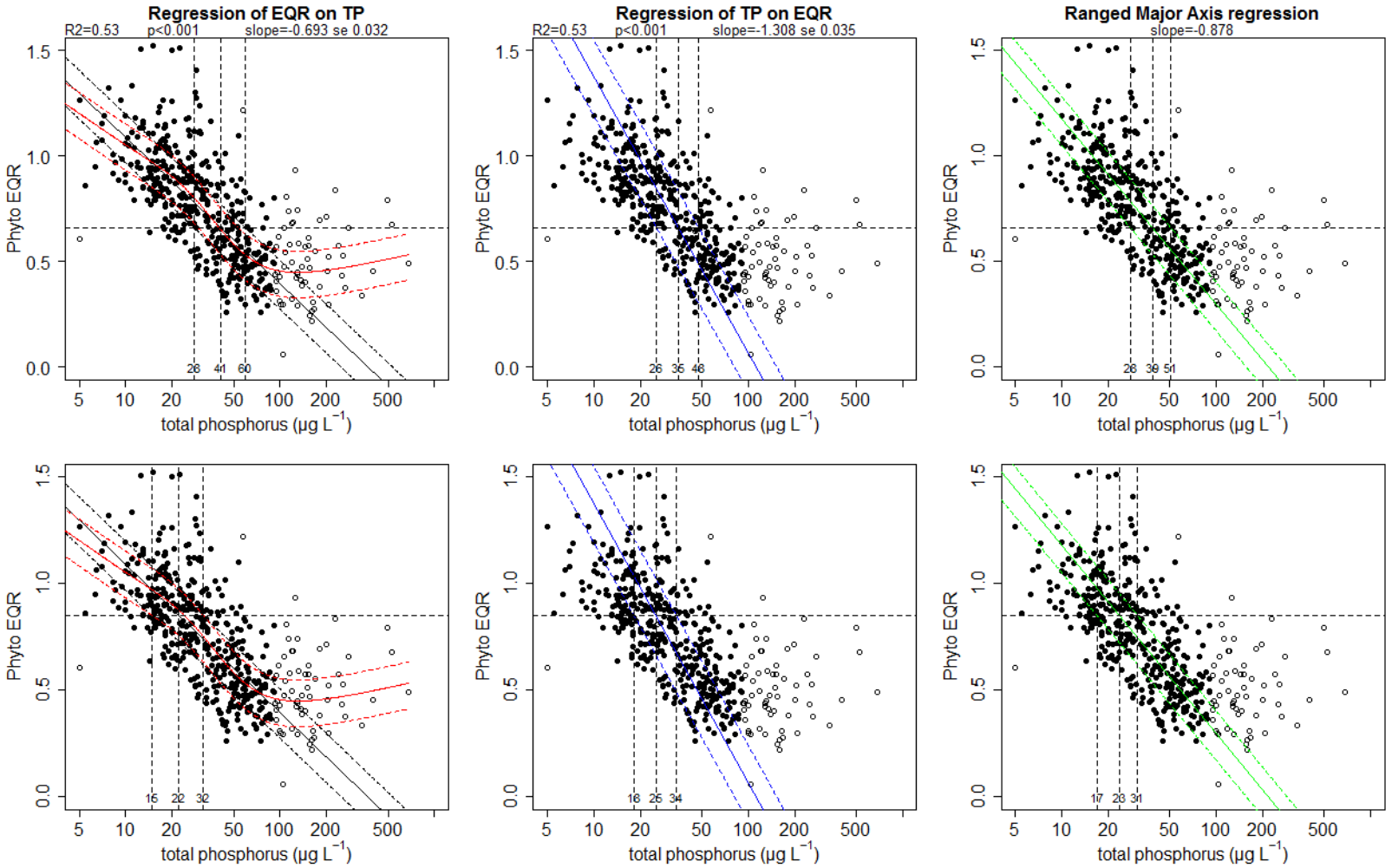

Figure 3-1 Regression relationships between common metric EQR for phytoplankton and total phosphorus ($\mu g l^{-1}$) for shallow high alkalinity lakes (Intercalibration type L-CB1) showing a) G/M and b) H/G boundaries. Solid points used for fitting relationship, dotted lines represent area containing 50% of the data.

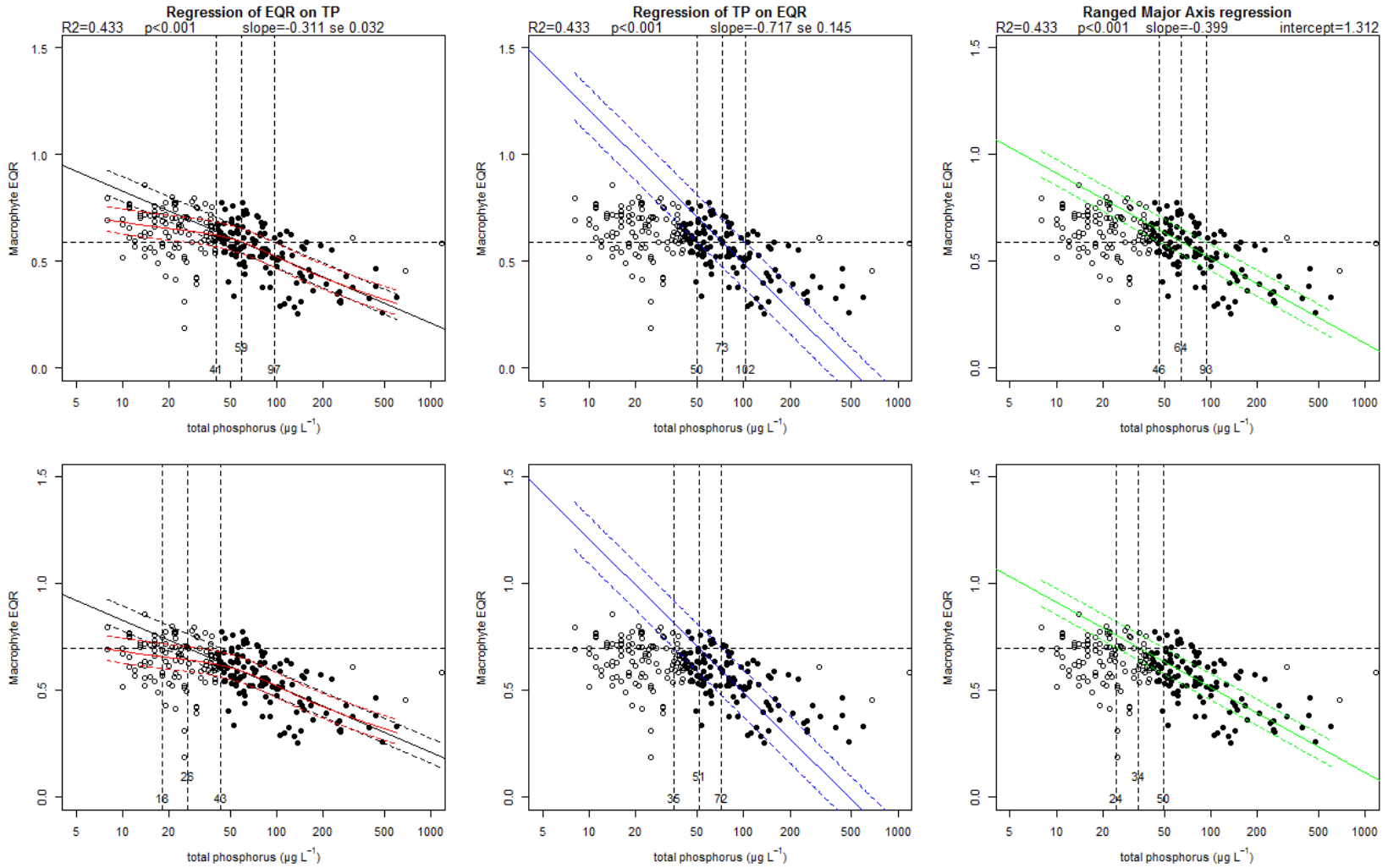

Figure 3-2 Regression relationships between common metric EQR for macrophytes and total phosphorus ($\mu g l^{-1}$) for shallow high alkalinity lakes (Intercalibration type L-CB1) showing a) G/M and b) H/G boundaries. Solid points used for fitting relationship, dotted lines represent area containing 50% of the data

3.1.2 Bivariate regression models

Including both TP and TN in models for phytoplankton and macrophytes increased the R^2 value significantly relative to the use of TN only but not for TP only in the case of macrophytes (Table 3-1 & Table 3-2). The resulting good moderate boundary values are similar to those from the univariate models (TP 40 range 28 - 57; TN 1.05 range 0.75 - 1.50) but are more reliable. It is interesting to note that the contour lines showing boundary values for macrophytes intersect the relationship between TP and TN at an angle much closer to 90° than they do for phytoplankton (Figure 3-3) showing that macrophyte status is more influenced by TN than it is for phytoplankton.

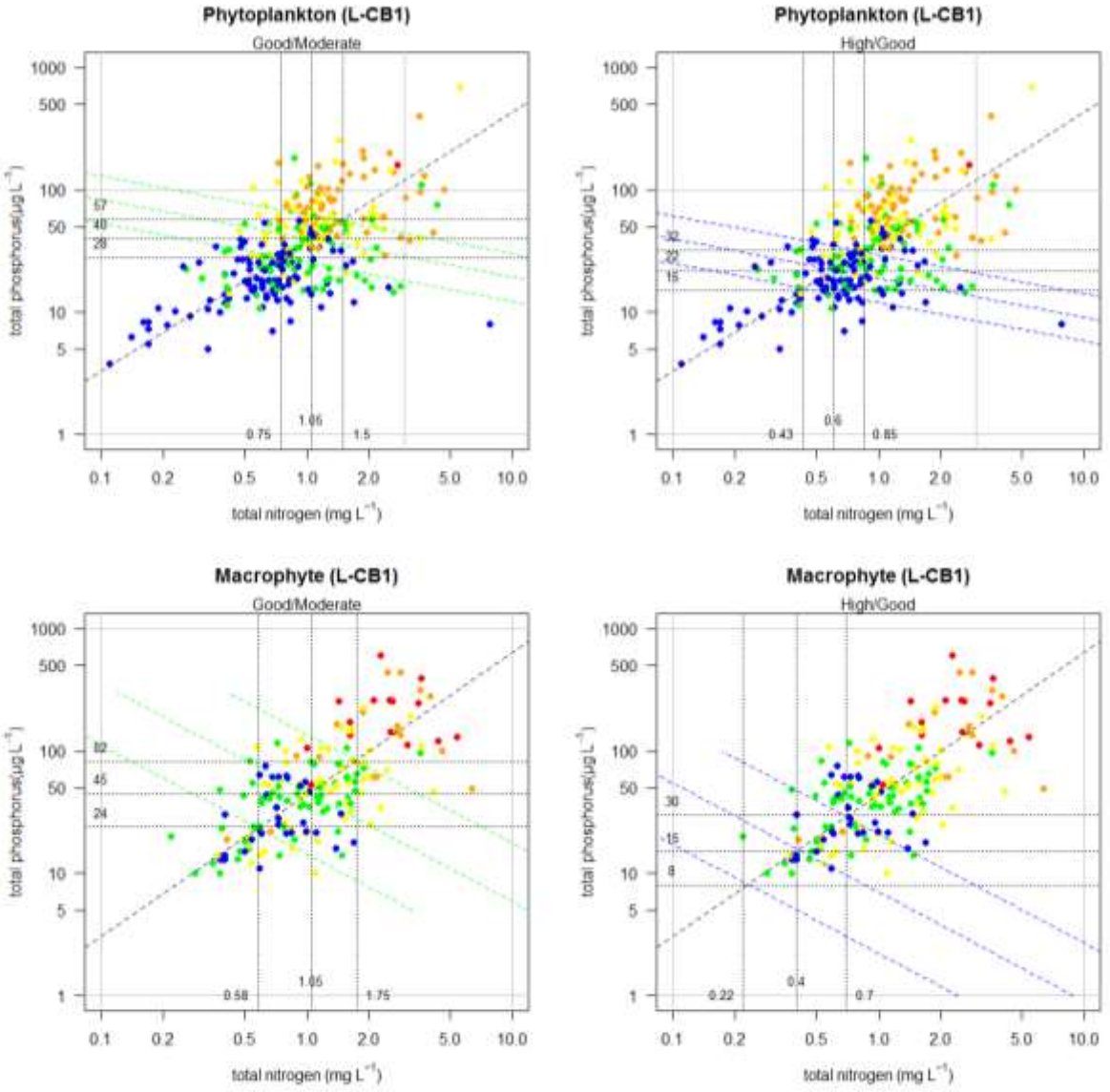

Figure 3-3 Relationship between mean TP and TN, points coloured by WFD class for phytoplankton and macrophytes in high alkalinity shallow lakes (Intercalibration type L-CB1). Coloured dotted lines contours of predicted TN and TP concentration when phytoplankton EQR is at a) good/moderate boundary (green) ± 25th and 75th residuals of prediction, b) high/good boundary (blue) ± 25th and 75th residuals of prediction. Horizontal and vertical lines show intersection with RMS regression of observed TP and TN showing boundary concentrations.

3.1.3 Categorical relationships

Box plots showing the range of TP and TN concentrations in lakes classified using phytoplankton and macrophytes are shown in Figure 3-4 and Figure 3-5. One approach to defining a good/moderate boundary value is to take the average of the upper 75th quantile of lakes classified as Good and the lower 25th quantile of lakes classified as Moderate. At this value less than 25% of lakes would be at moderate status and more than 75% would be at good status. A similar and potentially simpler approach would be to take the average of the median value of nutrient concentration at good and moderate status. The results for both phytoplankton and macrophytes provide very similar boundary values to those from regression modelling and the outcomes for macrophytes and phytoplankton are more similar to each other than they are using regression approaches (Table 3-1 & Table 3-2), suggesting that this categorical approach can be used, at least for relatively large data sets.

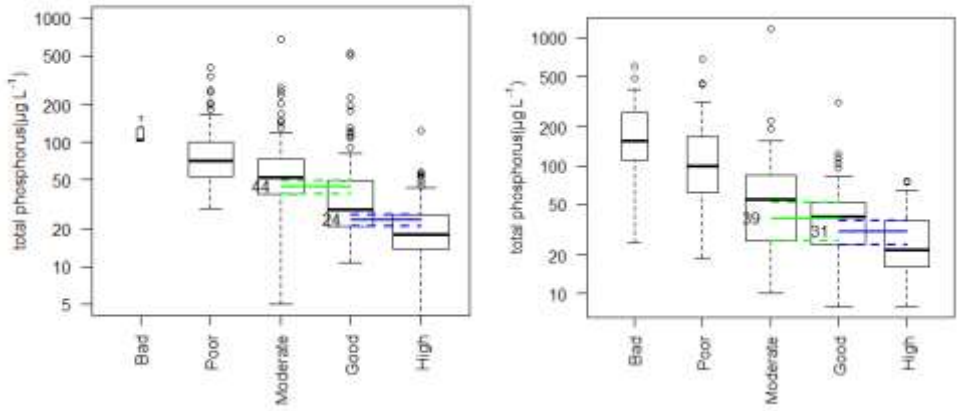

Figure 3-4 Range of TP (µgl^{-1}) for shallow high alkalinity lakes (Intercalibration type L-CB1) classified using common metric for a) phytoplankton (left graph) and b) macrophytes (right graph). Values show average of the 75th of the upper class and 25th of the lower class as potential values for G/M (green) and H/G (blue)

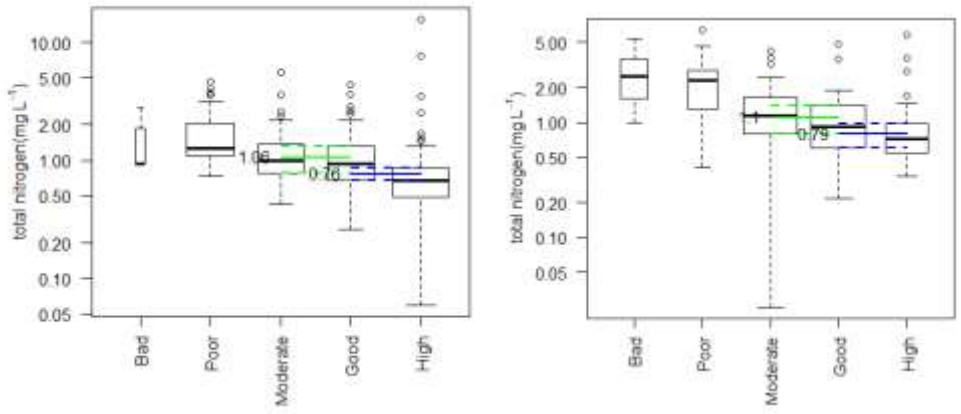

Figure 3-5 Range of total nitrogen (mgl^{-1}) for shallow high alkalinity lakes (Intercalibration type L-CB1) classified using common metric for a)phytoplankton and b) macrophytes. Values show average of the 75th of the upper class and 25th of the lower class as potential boundary values for G/M (green) and H/G (blue)

3.1.4 Minimise the mismatch between biological and supporting element classification

By plotting the percentage of water bodies that would be at good or better status for biology but moderate or worse for nutrients for different potential boundary values can identify nutrient good moderate boundary concentrations where the rate of mismatch decreases. By overlaying a similar plot showing the percentage of water bodies where biology is moderate or worse but nutrients are good or better a point of intersection can be estimated where the mismatch of classifications is minimised (Figure 3-6). For good/moderate status using phytoplankton this occurs at a TP concentration of 40 μgl^{-1} and a TN concentration of 0.76 mgl^{-1}. For macrophytes the values are slightly higher, TP of 45 μgl^{-1} and a TN of 0.90 mgl^{-1} (Figure 3-7). These values are similar to those produced by both the categorical and regression analysis (Table 3-1 & Table 3-2). This approach also demonstrates that it is possible to achieve relatively low rates of mismatch, for TP around 10% and for TN slightly higher at 20%.

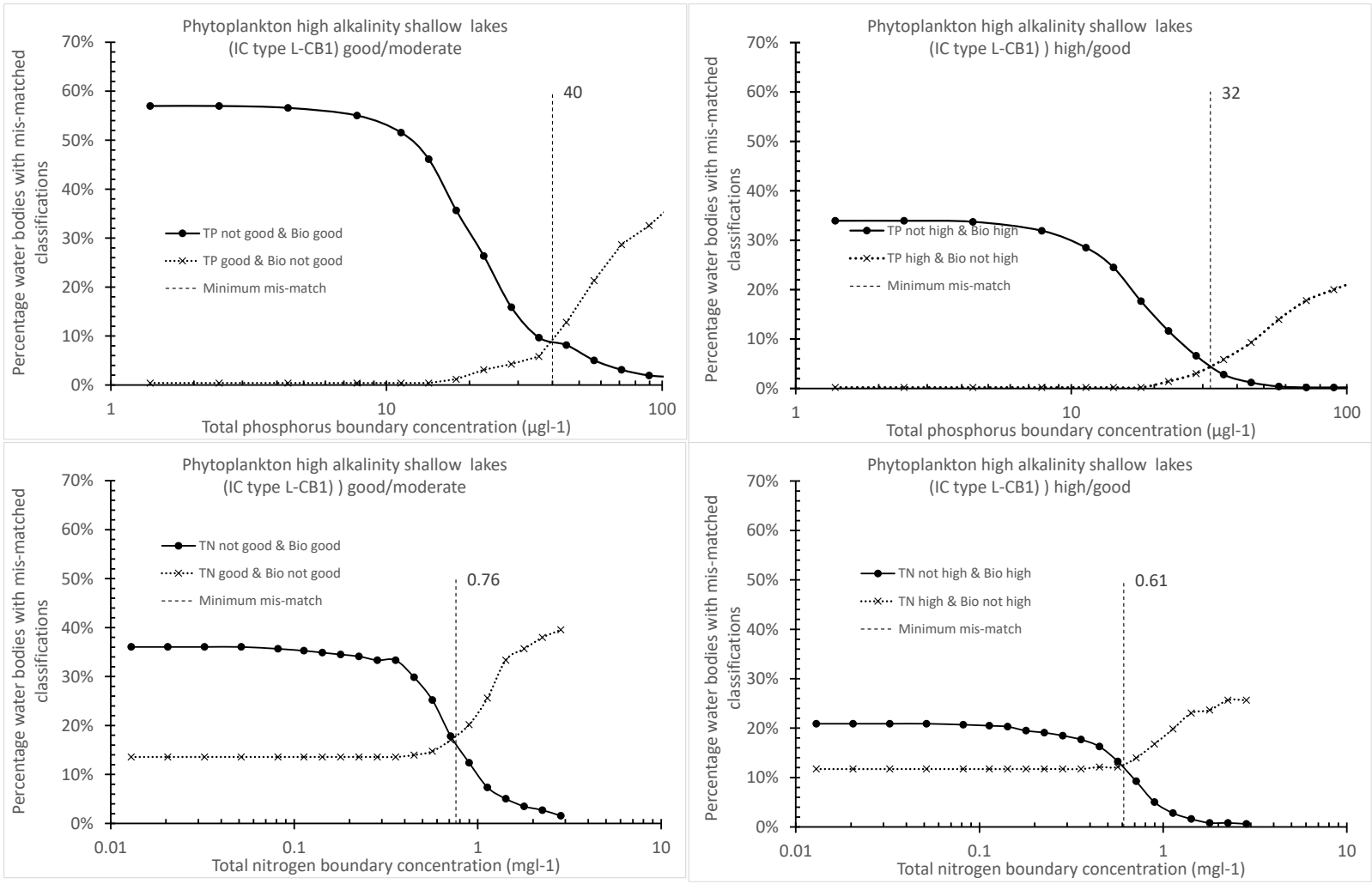

Figure 3-6 Percentage of water bodies where phytoplankton or nutrient classifications for ecological status differ in comparison to the level used to set the boundary values for a) total phosphorus and b) total nitrogen in high alkalinity shallow lakes, intercalibration type L-CB1. Vertical line marks intersection of curves where mismatch is minimised and equal.

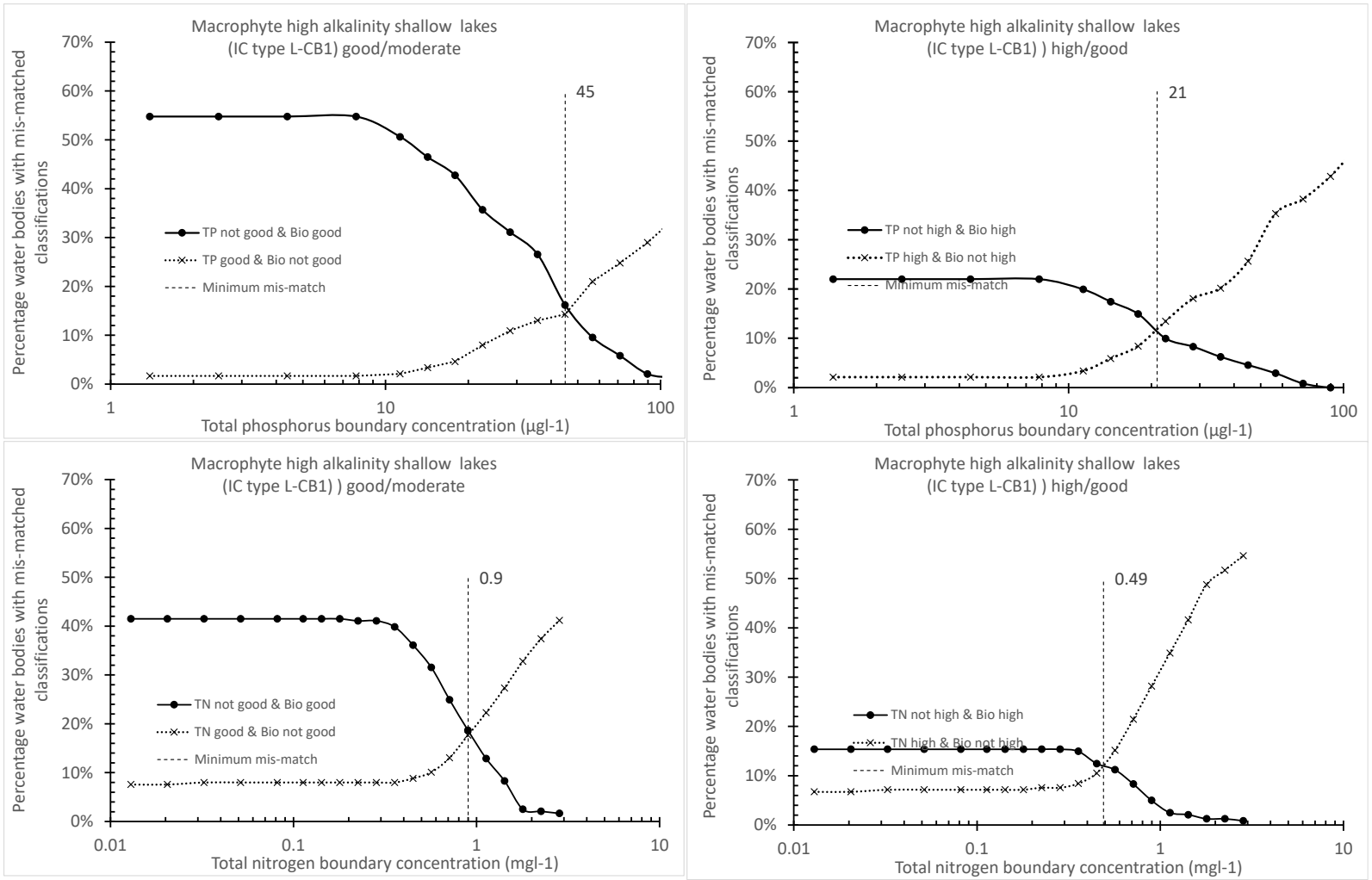

Figure 3-7 Percentage of water bodies where macrophyte or nutrient classifications for ecological status differ in comparison to the level used to set the boundary values for a) total phosphorus and b) total nitrogen in high alkalinity shallow lakes, intercalibration type L-CB1. Vertical line marks intersection of curves where mismatch is minimised and equal.

3.2 Summary high alkalinity lakes

Intercalibration data for phytoplankton and macrophytes from CBGIG very shallow high alkalinity lakes, phytobenthos from XGIG high alkalinity lakes and invertebrates from high alkalinity lakes were used. Detailed results are shown in the Appendix, section 6.1 and are summarised in Table 3-3 &

Table 3-4.

Table 3-3 Summary of predicted total phosphorus boundary values for high alkalinity lakes

IC Type	BQE used		GM TP µgl^{-1}			HG TP µgl^{-1}		
			Pred	range		Pred	range	
LCB1	Phytoplankton	most likely boundary		35	44		22	32
		best model R^2 0.55	40	28	57	22	15	32
		possible range		26	61		15	37
	Macrophytes	most likely boundary		39	73		15	51
		best model R^2 0.40	45	24	82	15	8	30
		possible range		24	102		8	72
LCB2	Phytoplankton	most likely boundary		45	70		32	35
		best model R^2 0.68	52	40	75	34	27	42
		possible range		35	122		22	55
	Macrophytes	most likely boundary		66	90		23	53
		best model R^2 0.47	70	36	125	30	16	56
		possible range		25	156		9	87
XGIG LCB1 LCB2	Phytobenthos	most likely boundary		36	47		16	29
		best model R^2 0.50	45	24	83	19	10	35
		possible range		22	96		7	42
	Invertebrates	most likely boundary		41	49		16	27
		best model R^2 0.38	43	22	90	21	11	44
		possible range		15	119		5	48

Table 3-4 Summary of predicted total nitrogen boundary values for high alkalinity lakes

IC Type	BQE used		GM TN mgl^{-1}			HG TN mgl^{-1}		
			Pred	range		Pred	range	
LCB1	Phytoplankton	most likely boundary		0.76	1.06		0.55	0.81
		best model R2 0.55	1.05	0.75	1.50	0.60	0.43	0.85
		possible range		0.54	1.81		0.28	1.09
	Macrophytes	most likely boundary		0.90	1.44		0.40	1.14
		best model R^2 0.40	1.05	0.58	1.75	0.40	0.22	0.70
		possible range		0.58	1.93		0.22	1.42
LCB2	Phytoplankton	most likely boundary		1.10	1.47		0.94	1.06
		best model R^2 0.68	1.15	1.00	1.40	0.96	0.85	1.15
		possible range		0.92	1.83		0.65	1.39
	Macrophytes	most likely boundary		1.36	1.55		0.71	1.27
		best model R^2 0.47	1.36	0.92	2.10	0.80	0.52	1.20
		possible range		0.81	2.39		0.42	1.66

For the shallow high alkalinity lakes (L-CB1) the lowest predicted good/moderate TP boundary values were from phytoplankton, with a range from 35 – 44 μgl^{-1} which is similar to the ranges predicted from the XGIG phytobenthos (36 – 47 μgl^{-1}) and CBGIG invertebrates (all types 41 – 49 μgl^{-1}). The predictions derived from macrophytes were higher (39 – 73 μgl^{-1}), although the categorical and multivariate analysis suggested lower values (39 – 45 μgl^{-1} Table 3-1) similar to those from the other BQEs.

Taking into consideration the uncertainty derived from the multivariate models suggests that the good/moderate boundary for this lake type should be within the range of 28 – 57 μgl^{-1} TP, if based on phytoplankton, higher for macrophytes (24 – 82 μgl^{-1}) which is similar to the range predicted from phytobenthos and invertebrates. The most similar broad type to this intercalibration type is broad type 3, lowland calcareous/mixed stratified lakes, and c.70% of countries with lakes of this type report boundaries that fall within this range (Figure 3-8 red dotted lines). If the wider possible range is considered (blue line), then only two countries (RO, HU) have national good/moderate boundaries that are higher. It is however important to note that neither of these two countries were involved in the L-CB1 intercalibration exercise and thus their data were not influencing the regression outcome.

As for TP the range of TN good/moderate boundaries is lowest for phytoplankton (0.76 – 1.06 mgl^{-1}), although the multivariate model for macrophytes suggested that nitrogen had more influence on macrophytes than on phytoplankton. Comparing the modelled boundary values with those being used in broad type 3 shows that fewer national type boundaries for TN fall within the possible range of values (Figure 3-9)

The values can be compared with modelled values determined from regressions between member state national phytoplankton metrics calculated during the intercalibration exercise. Only scatter plots and R^2 values were reported in the intercalibration technical report, but the original regression equations were available to the authors and were used to determine boundary values (Table 3-5). These regressions were derived from the same data set as those discussed above, but use the standardised national phytoplankton metrics applied to all countries data. The range of boundary values for TP (29 – 58 μgl^{-1}) and TN (0.73 – 1.47 mgl^{-1}) are very similar to the range derived from the multivariate phytoplankton model.

Boundary predictions from the very shallow lake type (L-CB2) produced higher values, although again models using phytoplankton had lower values than those from macrophytes. Comparing the modelled ranges with the most comparable broad lake type, type 4 lowland calcareous/mixed very shallow lakes, shows that again the majority of countries fall within the uncertainty range of the models, particularly if macrophytes are considered (Figure 3-10 & Figure 3-11). As for L-CB1, it should be noted that not all countries shown in Figure 3-10 and Figure 3-11 were part of the L-CB2 intercalibration and thus did not have national data influencing the relationship.

In summary, analysis of the available data for high alkalinity lakes demonstrates a relatively wide range of potential boundary values. Those generated from phytoplankton are the lowest and comparing these with the values used by member states in similar lake

types suggests that 60-70% currently use boundary values lower than these for TP, but only 30% for TN.

Table 3-5 Range of predicted total phosphorus boundary values for high alkalinity shallow lakes using national metrics (taken from notes produced for intercalibration technical report)

		National Metrics			Good/Moderate		High/Good	
Country	IC Type	intercept	slope	adj R^2	EQR	TP µg/l	EQR	TP µg/l
BE	L-CB1	1.339	-0.465	0.335	0.6	39	0.8	14
DE	L-CB1	1.241	-0.417	0.381	0.6	34	0.8	11
DK	L-CB1	1.274	-0.477	0.450	0.6	26	0.8	10
EE	L-CB1	-0.556	1.863	0.233	2.5	44	1.5	13
IE	L-CB1	1.257	-0.447	0.447	0.6	29	0.8	11
NL	L-CB1	1.380	-0.517	0.497	0.6	32	0.8	13
PL	L-CB1	1.390	-0.448	0.337	0.6	58	0.8	21
UK	L-CB1	1.645	-0.631	0.550	0.6	46	0.8	22

Table 3-6 Range of predicted total nitrogen boundary values for high alkalinity shallow lakes using national metrics (taken from notes produced for intercalibration technical report)

		National Metrics			Good/Moderate		High/Good	
Country	IC Type	intercept	slope	adj R^2	EQR	TN mg/l	EQR	TN mg/l
BE	L-CB1	0.614	-0.378	0.149	0.6	1.09	0.8	0.32
DE	L-CB1	0.618	-0.337	0.274	0.6	1.13	0.8	0.29
DK	L-CB1	0.552	-0.344	0.179	0.6	0.73	0.8	0.19
EE	L-CB1			ns	2.5	ns	1.5	ns
IE	L-CB1	0.545	-0.468	0.319	0.6	0.76	0.8	0.28
NL	L-CB1	0.555	-0.462	0.268	0.6	0.80	0.8	0.30
PL	L-CB1	0.679	-0.474	0.209	0.6	1.47	0.8	0.56
UK	L-CB1	0.662	-0.542	0.299	0.6	1.30	0.8	0.56

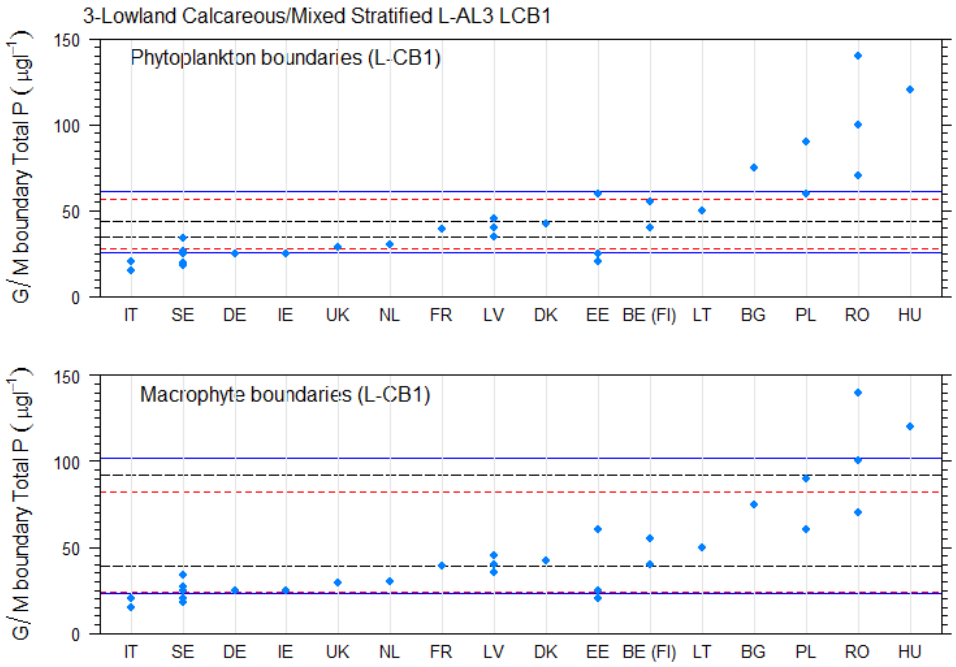

Figure 3-8 Comparison of range of reported good/moderate total phosphorus boundary values for broad type 3 lowland calcareous/mixed stratified lakes in comparison to range of modelled values for shallow high alkalinity lakes (intercalibration type L-CB1) using a) phytoplankton and b) macrophytes. Most likely range (black broken line) best model upper/lower quartiles of model residuals (red dotted line), possible range (blue solid line)

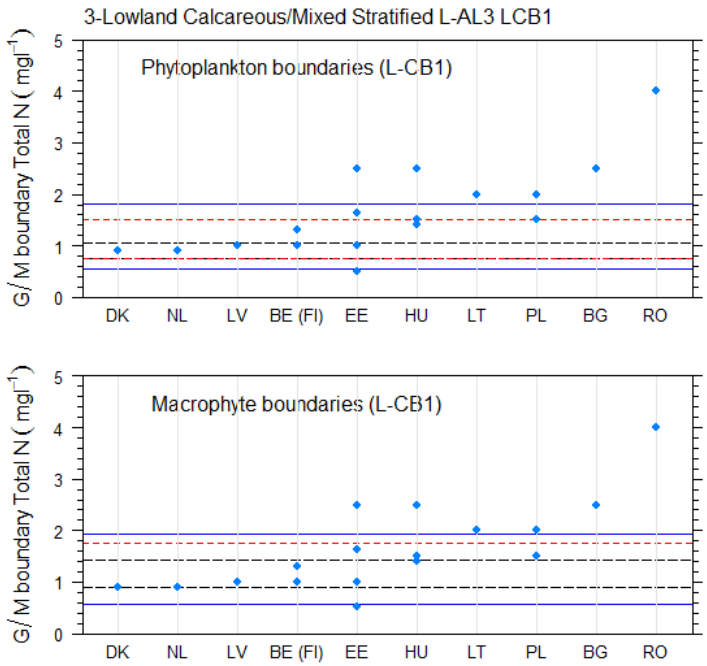

Figure 3-9 Comparison of range of reported good/moderate total nitrogen boundary values for broad type 3 lowland calcareous/mixed stratified lakes in comparison to range of modelled values for shallow high alkalinity lakes (intercalibration type L-CB1) using a) phytoplankton and b) macrophytes. Most

likely range (black broken line) best model upper/lower quartiles of model residuals (red dotted line), possible range (blue solid line)

Table 3-7 Range of predicted total phosphorus boundary values for high alkalinity very shallow lakes using national metrics (taken from notes produced for intercalibration technical report)

		National Metrics			Good/Moderate		High/Good	
Country	IC Type	intercept	slope	adj R^2	EQR	TP µg/l	EQR	TP µg/l
BE	L-CB2	1.259	-0.385	0.225	0.6	52	0.8	16
DE	L-CB2	1.395	-0.447	0.342	0.6	60	0.8	21
DK	L-CB2	1.139	-0.339	0.409	0.6	39	0.8	10
EE	L-CB2	0.150	1.249	0.269	2.5	76	1.5	12
IE	L-CB2	1.347	-0.545	0.522	0.6	23	0.8	10
NL	L-CB2	1.365	-0.431	0.422	0.6	59	0.8	20
PL	L-CB2	1.389	-0.436	0.321	0.6	65	0.8	23
UK	L-CB2	2.041	-0.779	0.565	0.6	71	0.8	39

Table 3-8 Range of predicted total nitrogen boundary values for high alkalinity very shallow lakes using national metrics (taken from notes produced for intercalibration technical report)

		National Metrics			Good/Moderate		High/Good	
Country	IC Type	intercept	slope	adj R^2	EQR	TN mg/l	EQR	TN mg/l
BE	L-CB2	0.636	-0.544	0.194	0.6	1.17	0.8	0.50
DE	L-CB2	0.649	-0.716	0.594	0.6	1.17	0.8	0.62
DK	L-CB2	0.608	-0.473	0.280	0.6	1.04	0.8	0.39
EE	L-CB2			ns	2.5	ns	1.5	ns
IE	L-CB2	0.435	-0.565	0.336	0.6	0.51	0.8	0.23
NL	L-CB2	0.669	-0.665	0.329	0.6	1.27	0.8	0.63
PL	L-CB2	0.709	-0.613	0.268	0.6	1.50	0.8	0.71
UK	L-CB2	0.818	-0.945	0.302	0.6	1.70	0.8	1.05

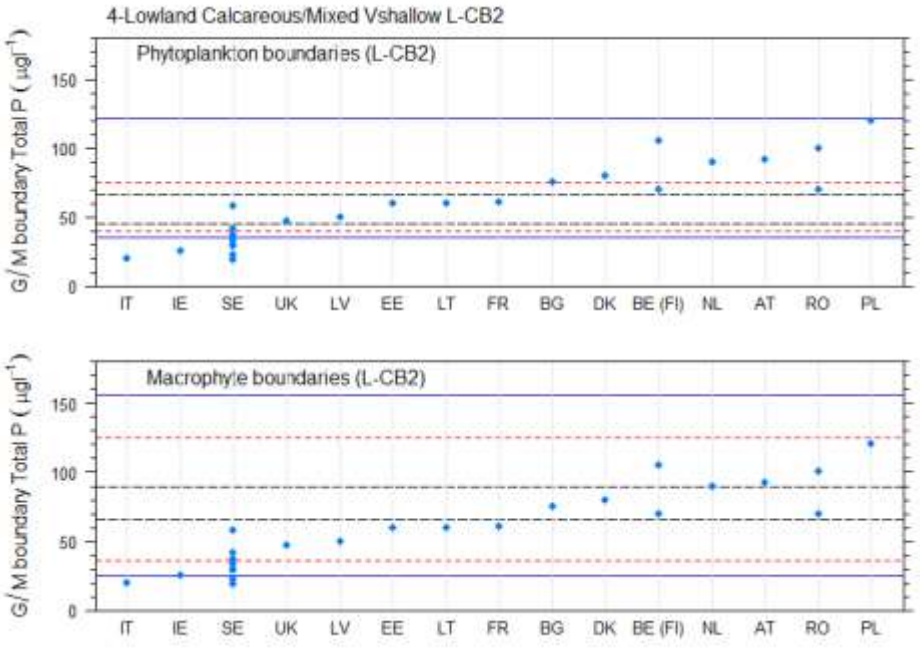

Figure 3-10 Comparison of range of reported good/moderate total phosphorus boundary values for broad type 4 lowland calcareous/mixed very shallow lakes in comparison to range of modelled values for very shallow high alkalinity lakes (intercalibration type L-CB2) using a) phytoplankton and b) macrophytes. Most likely range (black broken line) best model upper/lower quartiles of model residuals (red dotted line), possible range (blue solid line)

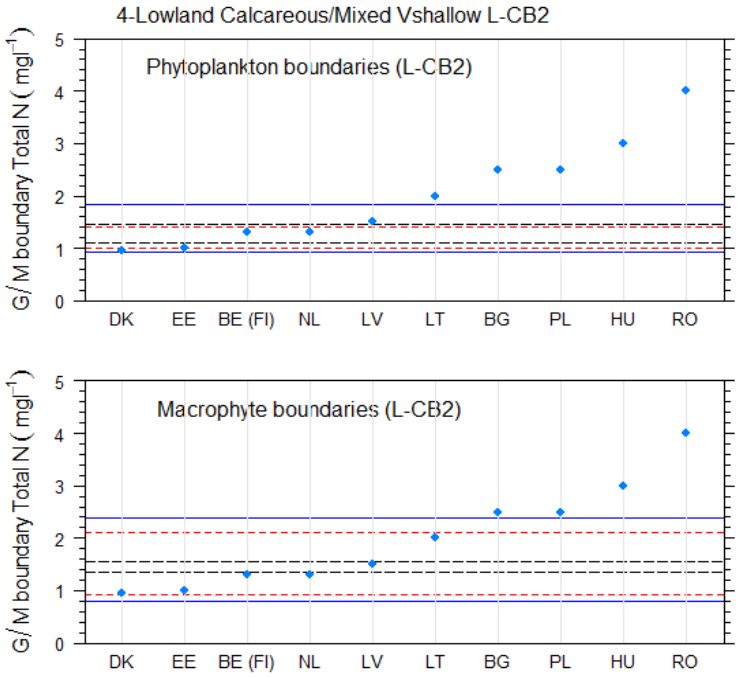

Figure 3-11 Comparison of range of reported good/moderate total nitrogen boundary values broad type 4 lowland calcareous/mixed very shallow lakes in comparison to range of modelled values for very shallow high alkalinity lakes (intercalibration type L-CB2) using a) phytoplankton and b) macrophytes. Most likely range (black broken line) best model upper/lower quartiles of model residuals (red dotted line), possible range (blue solid line)

3.3 Low and moderate alkalinity clear water lakes

Data from the NGIG intercalibration process were available that allowed modelled TP and TN boundary values for phytoplankton to be determined using the common metric for phytoplankton and for TP using the common metric for macrophytes (

Table 3-9). The types used for both BQEs covered low and moderate alkalinity lakes. For macrophytes the pressure gradient was too short to produce an adequate model for low alkalinity lakes (type 101) so the data for both low and moderate alkalinity lakes were combined for analysis. With the exception of boundaries predicted from phytoplankton for low alkalinity deep lakes (L-N2a) the range of boundary values for phytoplankton and macrophytes were similar (10 – 22 $\mu g l^{-1}$). This range of predicted good/moderate boundary values was very similar to the range of values reported by the majority of MS for broad type 2, lowland siliceous lakes (Figure 3-12).

The relationships for total nitrogen were mostly poor, with only the results from moderate alkalinity lakes (L-N1) providing reliable estimates of boundary values (Table 3-10). The range of predicted good/moderate boundary values (0.3 – 1.0 $mg l^{-1}$) was similar to the majority of MS boundary values (Figure 4-13).

Full details of models are shown in the appendix section 6.2

Table 3-9 Summary of predicted total phosphorus boundaries for low and moderate alkalinity lakes

IC Type	BQE used		GM TP $\mu g l^{-1}$			HG TP $\mu g l^{-1}$		
			Pred	range		Pred	range	
L-N2a	Phytoplankton	most likely boundary		11	22		8	10
		best model R^2 0.37	20	15	27	9	7	12
		possible range		9	31		6	13
L-N2b	Phytoplankton	most likely boundary		8	15		6	8
		best model R^2 0.37	14	11	19	8	6	10
		possible range		7	20		5	10
L-N1	Phytoplankton	most likely boundary		18	20		11	12
		best model R^2 0.81	18	15	22	11	9	13
		possible range		15	23		9	15
101 201	Macrophytes	most likely boundary		10	22		6	17
		best model R^2 0.41	22	16	29	14	10	19
		possible range		6	31		5	24

Table 3-10 Summary of predicted total nitrogen boundaries for low and moderate alkalinity lakes

IC Type	BQE used		GM TN mgl^{-1}			HG TN mgl^{-1}		
			Pred	range		Pred	range	
L-N2a	Phytoplankton	most likely boundary		0.41	0.65		0.35	0.41
		best model R^2 0.10	ns			ns		
		possible range		0.32	0.56		0.26	1.05
L-N2b	Phytoplankton	most likely boundary		0.39	0.55		0.29	0.39
		best model R^2 0.26	ns			ns		
		possible range		0.28	0.53		0.18	0.40
L-N1	Phytoplankton	most likely boundary		0.51	0.70		0.33	0.41
		best model R^2 0.81	0.65	0.52	0.79	0.36	0.28	0.44
		possible range		0.43	1.04		0.26	0.52

Table 3-11 Summary of predicted total phosphorus and total nitrogen boundaries for lakes in broad type 2 (lowland siliceous)

Broad Type	BQE used		GM TP μgl^{-1}		HG TP μgl^{-1}	
			range		range	
2	Phytoplankton	most likely boundary	8	22	6	12
		possible range	7	31	5	15
2	Macrophytes	most likely boundary	8	22	6	17
		possible range	6	31	5	24

Broad Type	BQE used		GM TN mgl^{-1}		HG TN mgl^{-1}	
			range		range	
2	Phytoplankton	most likely boundary	0.39	0.70	0.29	0.64
		possible range	0.28	1.04	0.18	1.05

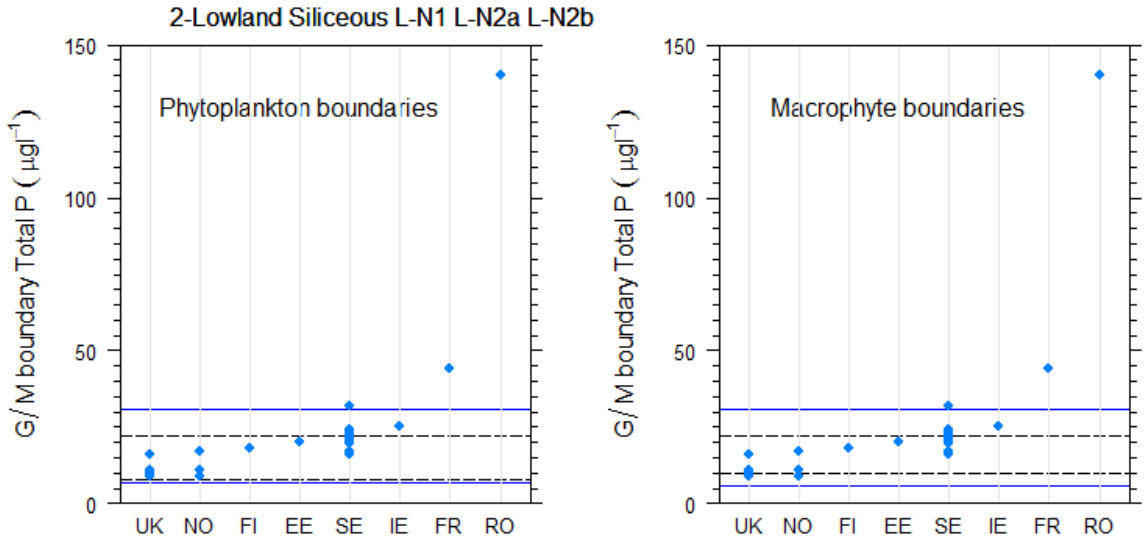

Figure 3-12 Comparison of range of reported good/moderate total phosphorus boundary values for broad type 2, lowland siliceous lakes, in comparison to range of modelled values for low/moderate alkalinity NGIG lakes using phytoplankton (intercalibration types L-N1, L-N2a, L-N2b) and b) macrophytes (intercalibration types 101, 201). Most likely range (black broken line), possible range (blue solid line)

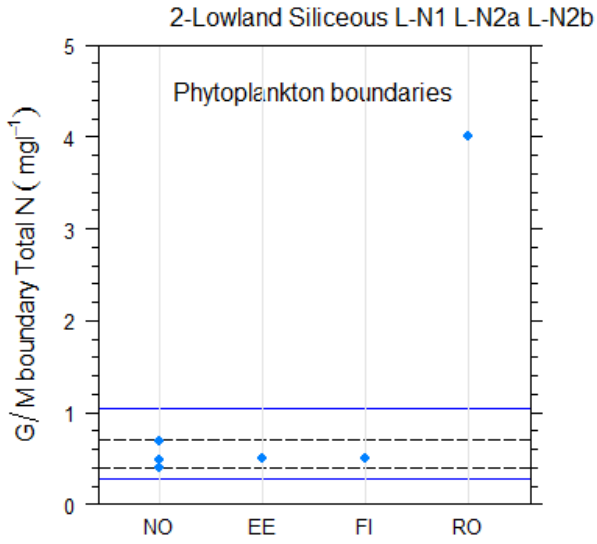

Figure 3-13 Comparison of range of reported good/moderate total nitrogen boundary values for broad type 2, lowland siliceous lakes, in comparison to range of modelled values for low/moderate alkalinity NGIG lakes using phytoplankton (intercalibration types L-N1, L-N2a, L-N2b). Most likely range (black broken line), possible range (blue solid line)

3.4 Low and moderate alkalinity humic lakes

As for the clear water lakes intercalibration data were used to estimate boundary values (

Table 3-12). As expected for humic lakes TP boundary values were higher than for the clear water lakes with moderate alkalinity lakes (phytoplankton type L-N6a & macrophyte type 202) having higher boundaries. For macrophytes the low alkalinity humic lakes (type 102) had a short pressure gradient with considerable scatter and a significant regression model could not be fitted to these data, either independently or in combination with the moderate alkalinity lake type (type 202).

The predicted ranges of the good/moderate boundary values for macrophytes were slightly higher than those for phytoplankton (Table 3-14). The majority of the reported member state boundary values for broad type 5, lowland humic and siliceous lakes, were within the range of these predicted values (Figure 3-14 & Figure 3-15)

Full details of models are shown in appendix 6.3.

Table 3-12 Summary of predicted total phosphorus boundaries for low and moderate alkalinity humic lakes

IC Type	BQE used		GM TP µgl^{-1}			HG TP µgl^{-1}		
			Pred	range		Pred	range	
L-N3a	Phytoplankton	most likely boundary		17	24		11	14
		best model R^2 0.61	22	18	27	12	10	15
		possible range		14	31		9	16
L-N8a	Phytoplankton	most likely boundary		26	28		14	19
		best model R^2 0.80	27	23	32	16	13	19
		possible range		20	38		11	23
L-N6a	Phytoplankton	most likely boundary		14	31		10	15
		best model R^2 0.41	25	19	34	14	10	19
		possible range		10	44		8	21
102	Macrophytes	most likely boundary		23	37		18	19
		best model	ns					
		possible range		16	33		11	25
202	Macrophytes	most likely boundary		30	36		18	28
		best model R^2 0.31	36	19	54	20	10	29
		possible range		16	61		9	39

Table 3-13 Summary of predicted total nitrogen boundaries for low and moderate alkalinity humic lakes

IC Type	BQE used		GM TN mgl^{-1}			HG TN mgl^{-1}		
			Pred	range		Pred	range	
L-N3a	Phytoplankton	most likely boundary		0.53	0.72		0.41	0.43
		best model R^2 0.61	0.72	0.61	0.84	0.46	0.40	0.53
		possible range		0.47	0.63		0.36	0.60
L-N8a	Phytoplankton	most likely boundary		0.80	0.86		0.55	0.68
		best model R^2 0.80	0.85	0.72	1.07	0.47	0.39	0.58
		possible range		0.68	1.03		0.53	0.87
L-N6a	Phytoplankton	most likely boundary		0.37	0.70		0.31	0.44
		best model R^2 0.41	0.6	0.50	0.72	0.41	0.34	0.50
		possible range		0.31	0.89		0.27	0.56

Table 3-14 Summary of predicted total phosphorus and total phosphorus boundaries for lakes in broad type 5, (lowland organic and siliceous)

Broad Type	BQE used		GM TP µgl^{-1}		HG TP µgl^{-1}	
			range		range	
5	Phytoplankton	most likely boundary	17	28	11	19
		possible range	14	38	9	23
5	Macrophytes	most likely boundary	23	37	18	28
		possible range	16	61	9	39

Broad Type	BQE used		GM TN mgl^{-1}		HG TN mgl^{-1}	
			range		range	
5	Phytoplankton	most likely boundary	0.53	0.86	0.41	0.68
		possible range	0.47	1.07	0.37	0.87

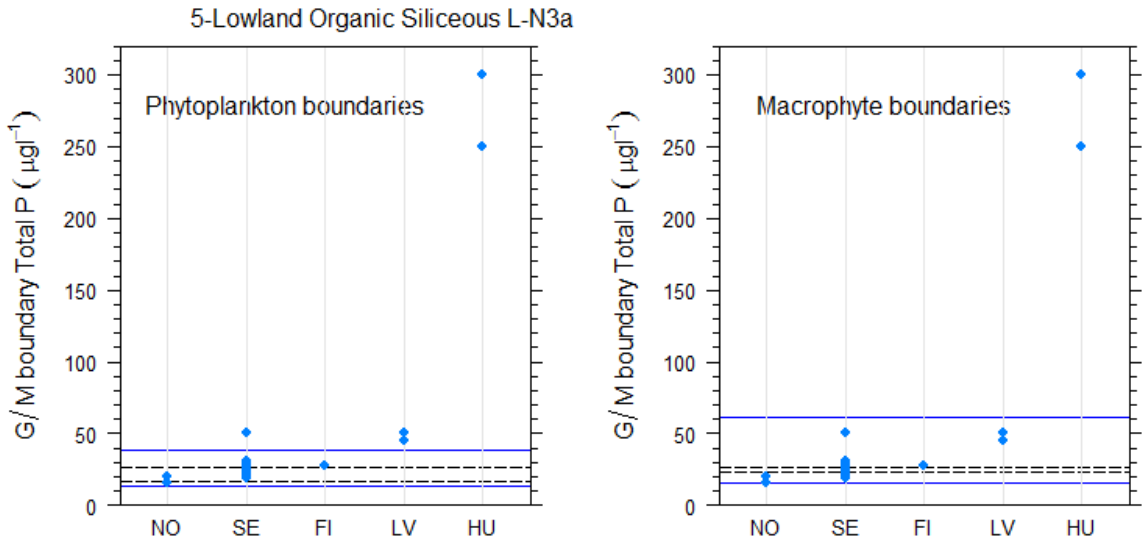

Figure 3-14 Comparison of range of reported good/moderate total phosphorus boundary values for broad type 5, lowland organic siliceous lakes, in comparison to range of modelled values for low/moderate alkalinity humic NGIG lakes using phytoplankton (intercalibration types L-N3a, L-N8a) and b) macrophytes (intercalibration types 102, 202). Most likely range (black broken line), possible range (blue solid line)

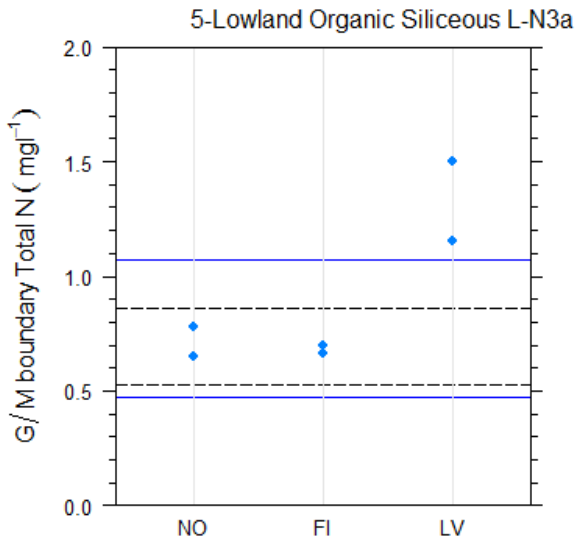

Figure 3-15 Comparison of range of reported good/moderate total nitrogen boundary values for broad type 5, lowland organic siliceous lakes, in comparison to range of modelled values for low/moderate alkalinity humic NGIG lakes using phytoplankton (intercalibration types L-N3a, L-N8a). Most likely range (black broken line), possible range (blue solid line)

3.5 Alpine Lakes

The relationship between national normalised EQR values for phytoplankton metrics from the Alpine GIG (Wolfram et al. 2014) were used to derive boundary values for total phosphorus (Table 3-15). It is assumed that these were OLS type I regression, but as the R^2 values were relatively high the estimated boundary values are unlikely to be significantly different from those that would have been generated using the preferred type II approach.

This gives a range of 14 – 32 μgl^{-1} for the good/moderate boundary which can be compared with the reported boundary values for broad type 8, mid-altitude calcareous mixed lakes (black broken line Figure 3-16). The majority of reported MS boundary metrics fall within this range.

No uncertainty values for the parameters were available so it is not possible to determine a wider range of potential boundaries. However, using the average values of the upper and lower quantiles of the residuals of the regression (-0.21 and +0.28) a typical range of 11 – 40 μgl^{-1} might be expected. All countries with lakes in broad type 8 reported boundaries within this wider range.

Table 3-15 Regression (OLS) parameters and estimated boundary values for Alpine lakes, parameters taken from Figure 2.2 in Wolfram et al. (2014).

Country	IC Type	Regression equation National Metrics			Good/Moderate boundary		High/Good boundary	
		intercept	slope	R^2	EQR	TP μgl^{-1}	EQR	TP μgl^{-1}
AT/SI	L-AL3	-0.1618	-0.178	0.62	0.6	14	0.8	5
DE	L-AL3	-0.1415	-0.176	0.57	0.6	15	0.8	5
IT	L-AL3	-0.1199	-0.176	0.52	0.6	17	0.8	5
AT/SI	L-AL4	-0.2523	-0.230	0.62	0.6	24	0.8	10
DE	L-AL4	-0.3173	-0.256	0.70	0.6	28	0.8	13
IT	L-AL4	-0.1023	-0.203	0.52	0.6	32	0.8	12

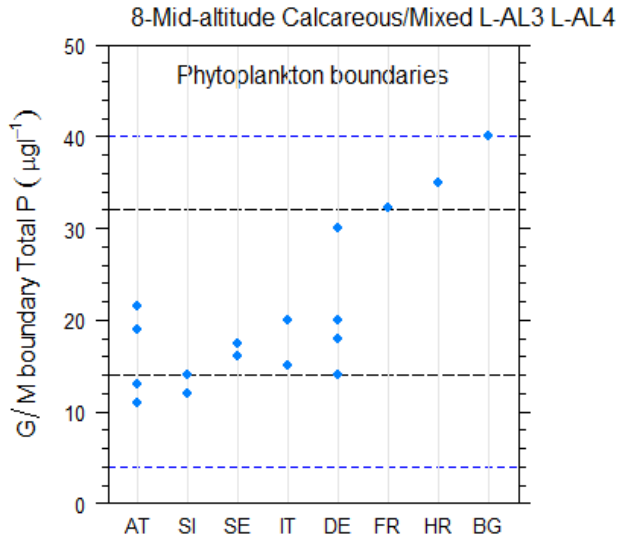

Figure 3-16 Comparison of range of reported good/moderate total phosphorus boundary values for broad type 8, mid-altitude calcareous/mixed lakes, in comparison to range of modelled values for Alpine lakes using regression parameters for national metric EQRs calculated during intercalibration for types L-AL3 and L-AL4 (Wolfram et al. 2014). Most likely range (black broken line), possible range estimated assuming ± 27% of predicted values (blue broken line).

3.6 Comparison of methods used to estimate boundary values

For lakes the relationships between biological status, expressed as an EQR, and total phosphorus concentration were relatively good, with few non-significant relationships. Phytoplankton typically had higher R^2 values than macrophytes and slightly higher than phytobenthos. Relationships with total phosphorus were better than those for total nitrogen.

When R^2 values were low the gradient of a type I OLS regression was lower than that of the type II RMA regression. The effect this has on the predicted boundary value depends

on the mean values for EQR and nutrient concentration, as the two regression lines intersect at the mean value of x and y. For the data analysed the mean values were typically higher than the good/moderate boundaries and thus the OLS regression produced higher good/moderate boundary values (Figure 3-17). The multivariate OLS regression tended to have higher R^2 values, they were less sensitive to outliers and the predicted boundary values were closer to the univariate RMA regression.

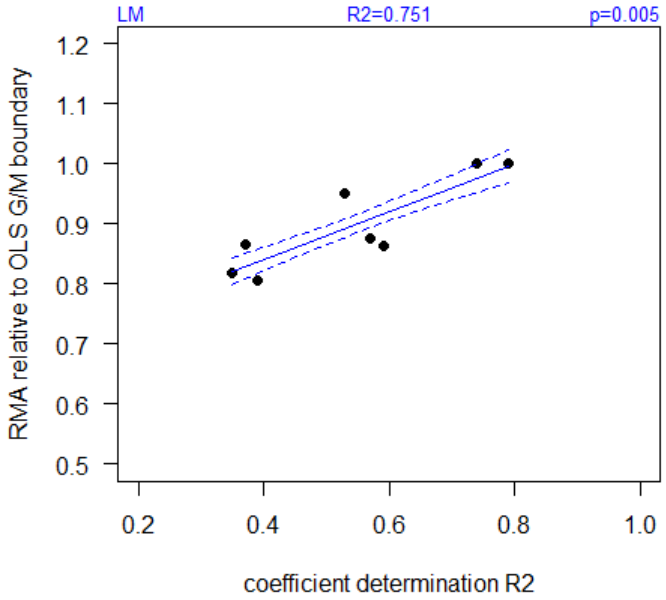

Figure 3-17 Relationship between the proportion of estimated good/moderate boundary value using type II RMA rather than type I OLS regression with coefficient of determination of the regression. (Phytoplankton models for CBGIG and NGIG)

The method of minimising classification mismatches and the categorical approach using box plots produced similar boundary values (Figure 3-19). Their reliability can be assessed from Figure 3-18, the relationship between values estimated using box plots and the best regression model has a slope that is not significantly different from 1. The method that minimises the mismatch of classifications has a slope that is significantly greater than 1 and thus tends to underestimate low boundary values and over estimate at higher values, although the differences are relatively small. As these approaches are not dependent on fitting a reliable linear model they are potentially a useful approach and would be worth further investigation with larger data sets.

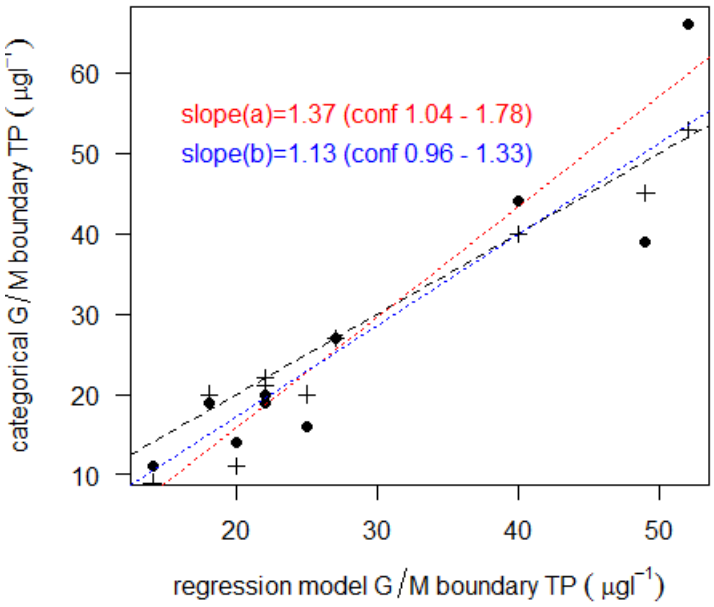

Figure 3-18 Relationship between good/moderate boundary values predicted from best regression model and a) minimising mismatch of classification (closed circles) and b) boxplots (cross). Black dotted line shows 1:1 relationship, red line RMA regression for mismatch method, blue line for RMA regression box plots.

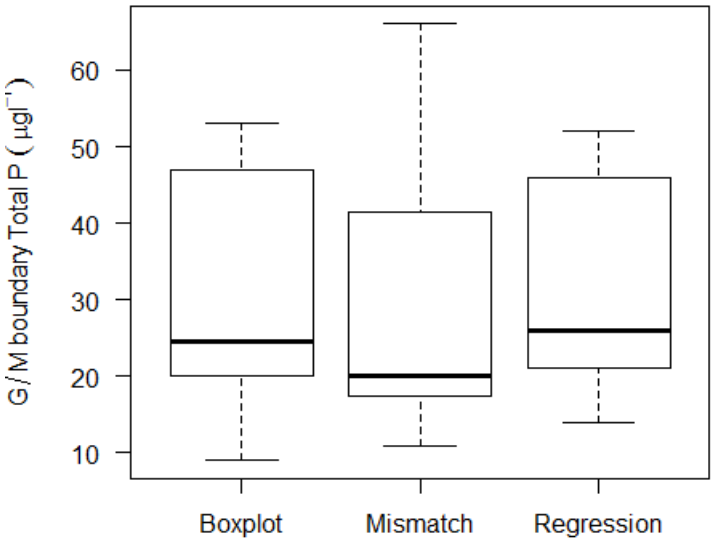

Figure 3-19 Range of good/moderate TP boundary values estimated using best regression model, mismatch of classification and boxplots.

4 Results for Rivers

4.1 Introduction

In general, the same approach followed for rivers although there were no data for phytoplankton.

For very large rivers (see section 4.4) the intercalibration data were used for phytobenthos (12 countries) and macroinvertebrates (20 countries). These data were highly comparable with the common metric EQRs as biological assessment results. Data for macroinvertebrates were analysed together, because further subtypes could not be differentiated, while two subtypes had to be distinguished for phytobenthos.

For small to large rivers only very few data were available from the intercalibration exercise and thus national data sets were used (see sections 4.2-4.3). In total, data from 16 countries were available which, when sub-setted by river type, nutrient (N or P) and sub-element (macrophtye or phytobenthos) yielding 94 relationships. However, relationships between nutrients and biology were much lower than was the case for lakes, with an average R^2 of 0.223. Only 51 of these relationships were statistically-significant.

A second stage of the exercise, therefore, grouped national datasets into broad types and repeated the analyses. Analyses were possible for low alkalinity lowland and upland river types rivers (including intercalibration types R-C1 and R-C3, and corresponding to broad types 2 and 3, and 8 and 9, respectively) as well as high alkalinity lowland rivers (intercalibration type R-C4, corresponding to broad type 4).

The merged datasets contained data from Austria, Denmark, Luxembourg, the Netherlands, Poland and United Kingdom. All these countries, with the exception of Austria reported the nutrient concentrations as annual averages. Sampling frequencies ranged from single (spot) to monthly measurements. Austria provided 90th percentile values; these were halved before being included into the analysis. The biological data were normalised EQR values i.e. status class boundaries adjusted to 0.8, 0.6, 0.4 and 0.2), based on national metrics. In addition to estimating nutrient thresholds for macrophytes and phytobenthos separately, a third set of models were constructed, for "combined macrophytes and phytobenthos", calculated as the minimum of the EQRs of the two sub-elements.

Of these three river types, however, only the low alkalinity rivers revealed significant relationships between nutrients and biology for total nitrogen and ortho- phosphorus (ortho-P). The relationships obtained for total phosphorus were not significant.

Table 4-1 Summary of datasets used for analyses of relationships between nutrients and biology for rivers.

BQE	Determinant	Type	Total number of samples	Country	Samples per country
Phytobenthos	Total nitrogen	Low alkalinity lowland	179	Netherlands	19
				Poland	60
				United Kingdom	100
		Low alkalinity upland	58	Poland	11
				United Kingdom	47
	Ortho-phosphorus	Low alkalinity lowland	120	United Kingdom	120
		Low alkalinity upland	230	Austria	73
				Luxembourg	85
				United Kingdom	72
Macrophytes	Total nitrogen	Low alkalinity lowland	263	Denmark	34
				Netherlands	52
				Poland	78
				United Kingdom	99
		Low alkalinity upland	58	Poland	11
				United Kingdom	47
	Ortho-phosphorus	Low alkalinity lowland	247	Denmark	129
				United Kingdom	118
		Low alkalinity upland	128	Luxembourg	56
				United Kingdom	72
Macrophytes & Phytobenthos (minimum)	Total nitrogen	Low alkalinity lowland	177	Netherlands	19
				Poland	59
				United Kingdom	199
		Low alkalinity upland	42	United Kingdom	47
	Ortho-phosphorus	Low alkalinity lowland	126	United Kingdom	126
		Low alkalinity upland	128	Luxembourg	56
				United Kingdom	72

4.2 Low alkalinity lowland rivers (R-C1)

The range of boundaries produced for lowland, low alkalinity rivers (broadly corresponding to R-C1) using phytobenthos, macrophytes and the combined macrophyte/phytobenthos model were generally lower to those for the upland rivers (see 4.3). Values for different sub-elements were similar, with the combined model giving the most stringent predictions (Figure 4-1, Figure 4-2; Table 4-2, Table 4-3).

The majority of the reported member state boundary values for broad type 3, lowland siliceous very small-small rivers, were within the range of these predicted values for macrophytes and phytobenthos separately (Figure 4-5 - Figure 4-7); however, the widest possible range for TN for macrophytes and phytobenthos was very high (> 4.9 mg l^{-1} and,

if this figure is discounted, then about half of all member states fall outside the limits. This is also the case for the combined model, where seven of the twelve participating MS have boundaries that fall outside the limits predicted by this exercise. High potential boundary values were also predicted for TN using phytobenthos although this value only protects one member state.

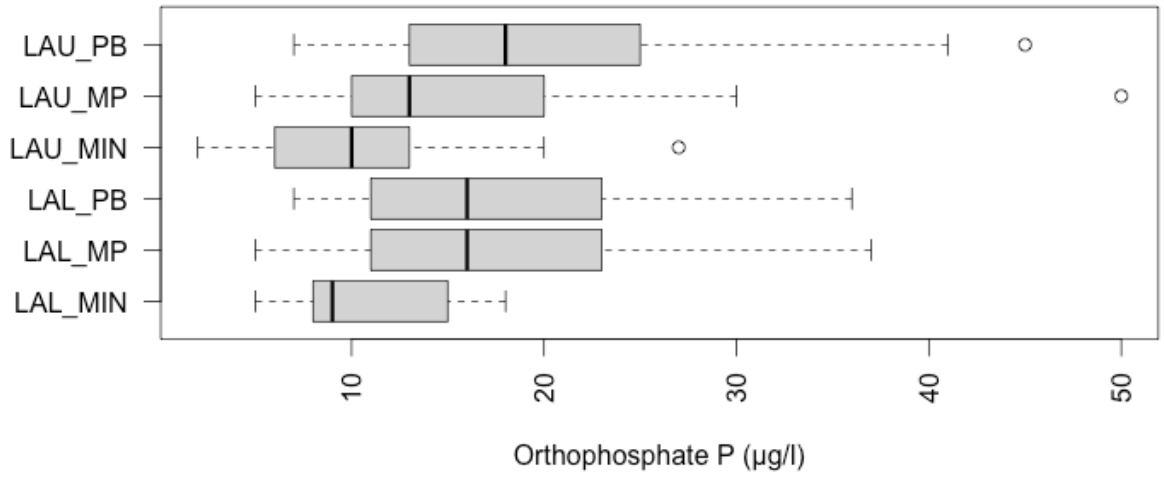

Figure 4-1 Ranges of the 14 threshold values (gained from regression analysis and categorical methods) for ortho-P for the high-good boundary for low alkalinity upland (LAU) and low alkalinity lowland (LAL) rivers. PB = phytobenthos; MP = macrophytes; MIN = minimum of PB and MP.

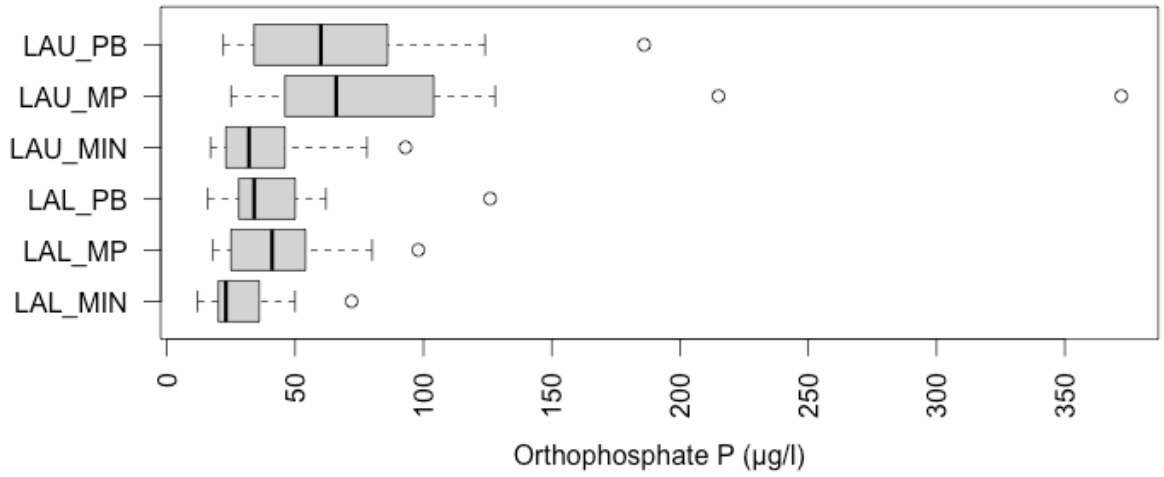

Figure 4-2 Ranges of the 14 threshold values (gained from regression analysis and categorical methods) for ortho-P for the good-moderate boundary for low alkalinity upland (LAU) and low alkalinity lowland (LAL) rivers. PB = phytobenthos; MP = macrophytes; MIN = minimum of PB and MP.

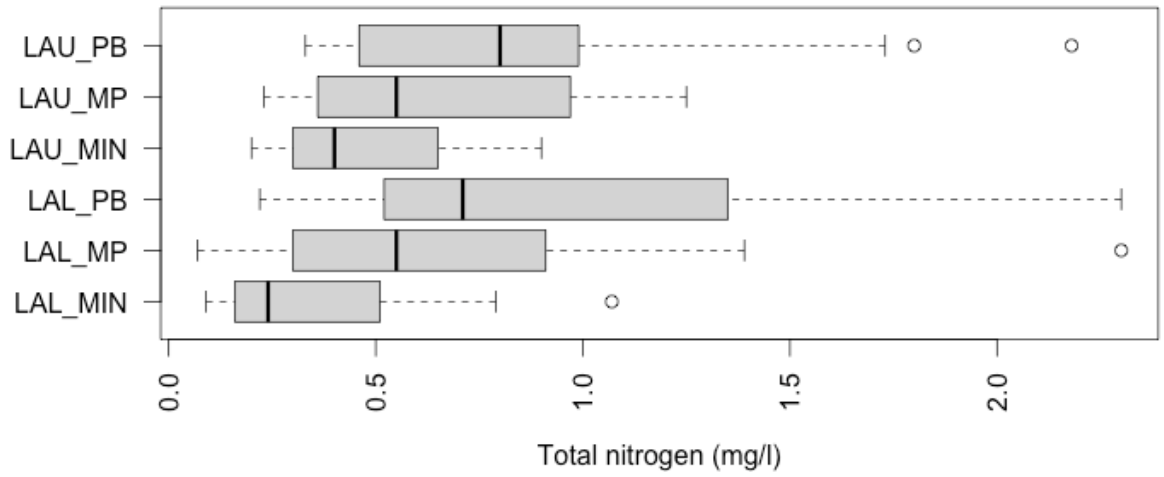

Figure 4-3 Ranges of the 14 threshold values (gained from regression analysis and categorical methods) for total nitrogen for the high-good boundary for low alkalinity upland (LAU) and low alkalinity lowland (LAL) rivers. PB = phytobenthos; MP = macrophytes; MIN = minimum of PB and MP.

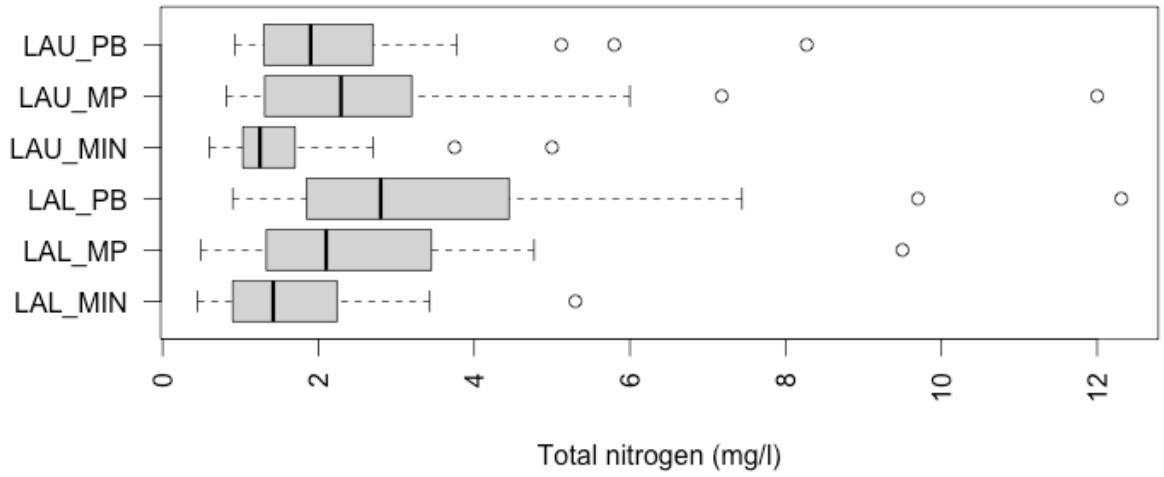

Figure 4-4 Ranges of the 14 threshold values (gained from regression analysis and categorical methods) for total nitrogen for the good-moderate boundary for low alkalinity upland (LAU) and low alkalinity lowland (LAL) rivers. PB = phytobenthos; MP = macrophytes; MIN = minimum of PB and MP

Table 4-2 Summary of predicted soluble phosphorus (ortho-P) boundaries for low alkalinity lowland rivers (LAL)

IC Type	BQE used		GM ortho-P µgl^{-1}			HG ortho-P µgl^{-1}		
			Pred	range		Pred	range	
LAL	Macrophytes	most likely boundary		32	45		11	22
		best model R^2 0.48	45	25	80	13	7	23
		possible range		18	98		5	37
LAL	Phytobenthos	most likely boundary		31	62		10	17
		best model R^2 0.49	39	26	55	16	11	23
		possible range		16	126		7	36
LAL	Combined	most likely boundary		20	36		8	12
		best model R^2 0.50	28	19	43	10	7	15
		possible range		12	72		6	18

Table 4-3 Summary of predicted total nitrogen boundaries for low alkalinity lowland rivers (LAL)

IC Type	BQE used		GM TN mgl^{-1}			HG TN mgl^{-1}		
			Pred	range		Pred	range	
LAL	Macrophytes	most likely boundary		1.38	3.50		0.23	0.93
		best model R^2 0.48	3.50	1.25	9.50	0.35	0.13	1.00
		possible range		0.49	9.50		0.07	2.30
LAL	Phytobenthos	most likely boundary		1.93	4.63		0.60	0.93
		best model R^2 0.49	3.50	1.54	7.44	0.64	0.28	1.37
		possible range		0.90	12.31		0.22	2.30
LAL	Combined	most likely boundary		1.03	2.10		0.18	0.57
		best model R^2 0.54	1.14	0.56	2.46	0.24	0.12	0.51
		possible range		0.45	5.30		0.09	1.07

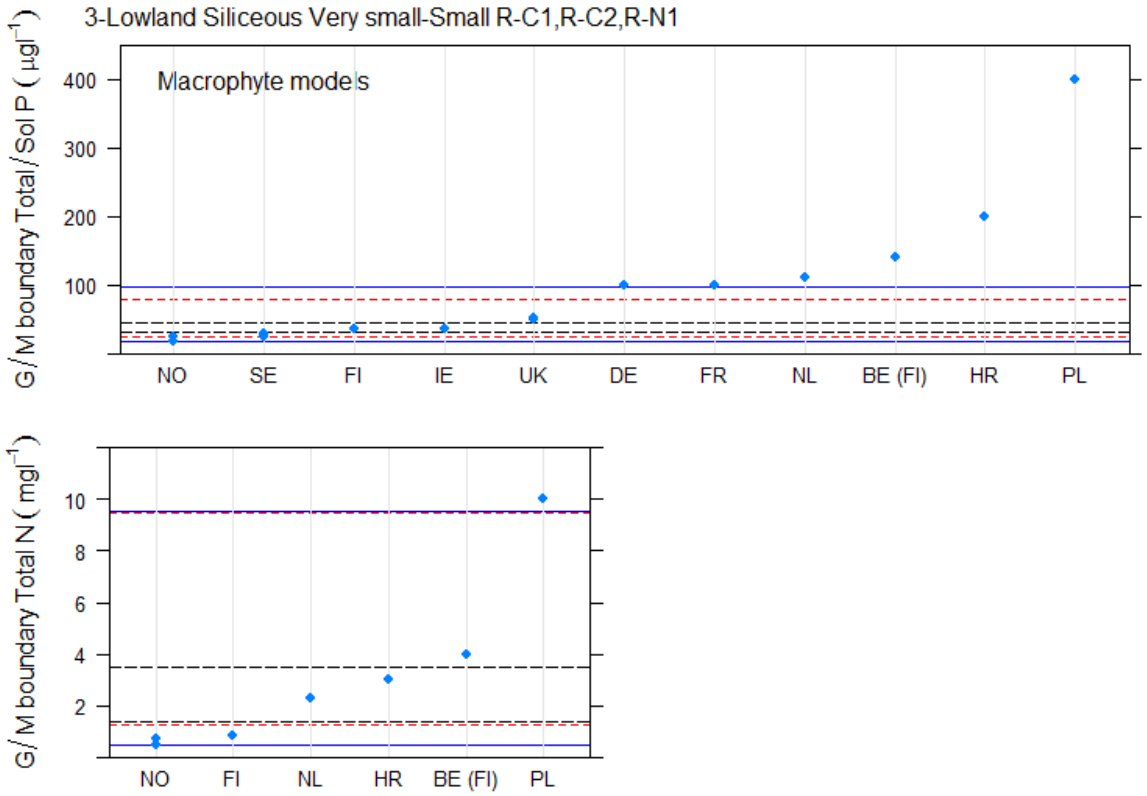

Figure 4-5 Comparison of range of reported good/moderate boundary values for a) ortho-phosphorus boundary values and b) total nitrogen for broad type 3, lowland siliceous very small-small rivers, in comparison to range of modelled values of ortho-P and TN for low alkalinity lowland rivers (including intercalibration type R-C1) using macrophytes. Most likely range (black broken line), best model upper/lower quartiles of model residuals (red dotted line), possible range (blue solid line) Note this figure was reproduced from Phillips & Pitt (2015), the boundary values for phosphorus are total phosphorus or where this was not available are total reactive phosphorus (UK & IE), values for FR are for 90th percentiles

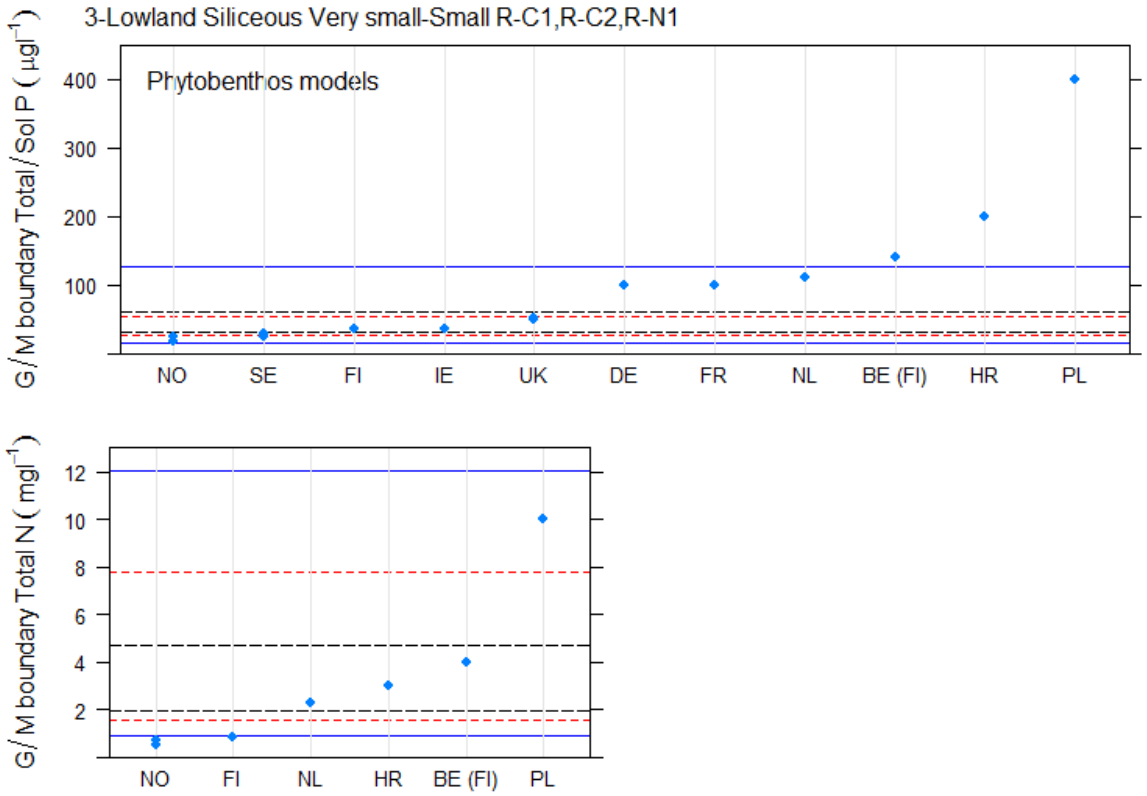

Figure 4-6 Comparison of range of reported good/moderate boundary values for a) ortho-phosphorus and b) total nitrogen for broad type 3, lowland siliceous very small-small rivers, in comparison to range of modelled values of ortho-P and TN for low alkalinity lowland rivers (including intercalibration type R-C1) using phytobenthos. Most likely range (black broken line), best model upper/lower quartiles of model residuals (red dotted line), possible range (blue solid line) Note this figure was reproduced from Phillips & Pitt (2015), the boundary values for phosphorus are total phosphorus or where this was not available are total reactive phosphorus (UK & IE), values for FR are for 90th percentiles

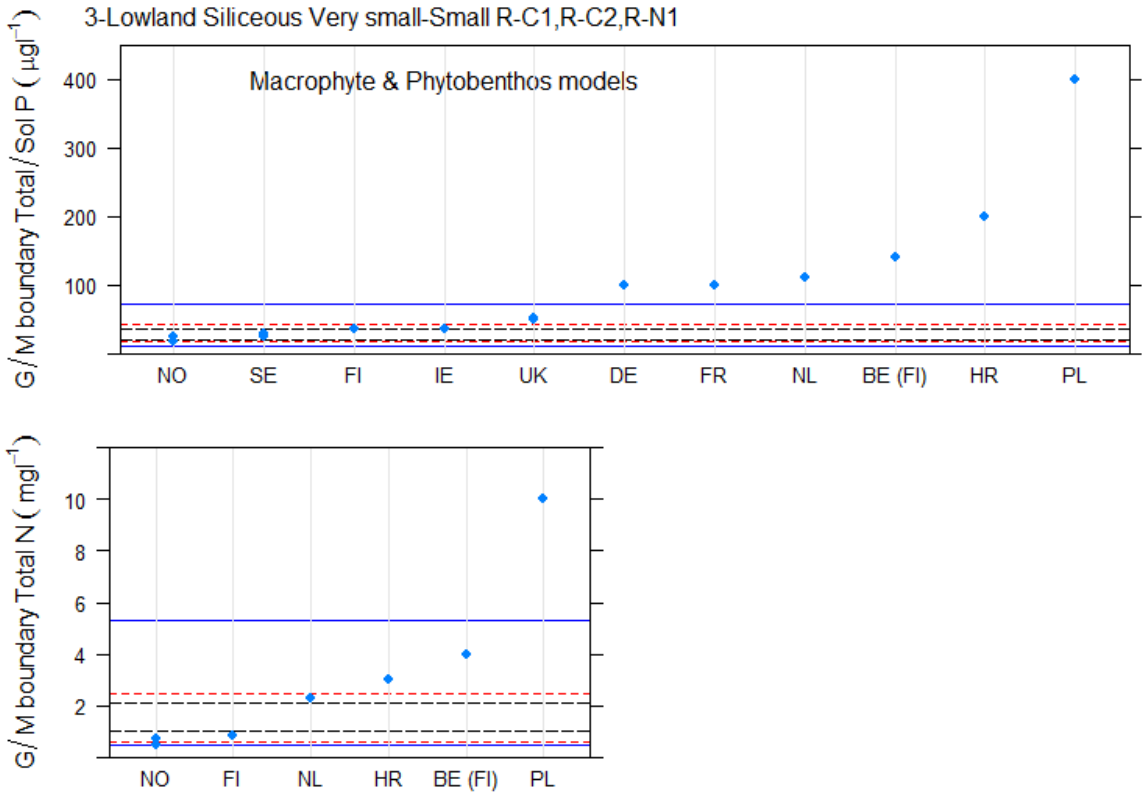

Figure 4-7 Comparison of range of reported good/moderate boundary values for a) ortho-phosphorus (Sol P) and b) total nitrogen for broad type 9 mid-altitude siliceous very small-small rivers in comparison to range of modelled values of ortho-P and TN for low alkalinity lowland rivers (including intercalibration type R-C1) using combined macrophytes and phytobenthos. Most likely range (black broken line) best model upper/lower quartiles of model residuals (red dotted line), possible range (blue solid line) Note this figure was reproduced from Phillips & Pitt (2015), the boundary values for phosphorus are total phosphorus or where this was not available are total reactive phosphorus (UK & IE), values for FR are for 90th percentiles

4.3 Low alkalinity upland rivers

Once again the range of boundaries produced for upland, low alkalinity rivers using phytobenthos, macrophytes and the combined macrophyte/phytobenthos model was similar, with phytobenthos being slightly less precautionary than macrophytes at the high-good boundary whilst macrophytes were slightly less precautionary than phytobenthos for predictions of the good-moderate boundary for ortho-P (Figure 4-1, Figure 4-2; Table 4-4-Table 4-5). The combined model gave the most stringent predictions in both cases.

The majority of the reported member state boundary values for broad types 9 mid-altitude siliceous very small-small rivers were within the range of these predicted values (Figure 4-8 - Figure 4-10).

Table 4-4 Summary of predicted ortho-phosphorus (ortho-P) boundaries for low alkalinity upland rivers (LAU)

IC Type	BQE used		GM ortho-P µgl^{-1}			HG ortho-P µgl^{-1}		
			Pred	range		Pred	range	
LAU	Macrophytes	most likely boundary		48	128		11	18
		best model R^2 0.40	48	25	106	18	10	30
		possible range		25	128		5	50
LAU	Phytobenthos	most likely boundary		34	86		13	25
		best model R^2 0.43	51	28	90	25	14	45
		possible range		22	124		7	45
LAU	Combined	most likely boundary		25	46		6	13
		best model R^2 0.50	25	20	35	10	8	13
		possible range		17	93		2	27

Table 4-5 Summary of predicted total nitrogen (TN) boundaries for low alkalinity upland rivers (LAU)

IC Type	BQE used		GM TN mgl^{-1}			HG TN mgl^{-1}		
			Pred	range		Pred	range	
LAU	Macrophytes	most likely boundary		1.31	6.00		0.50	0.69
		best model R^2 0.49	2.44	1.18	5.10	0.50	0.24	1.05
		possible range		0.82	12.00		0.23	1.25
LAU	Phytobenthos	most likely boundary		1.44	3.78		0.65	0.92
		best model R^2 0.53	2.48	1.27	5.12	0.84	0.43	1.73
		possible range		0.93	8.27		0.33	2.18
LAU	Combined	most likely boundary		0.89	2.16		0.39	0.43
		best model R^2 0.54	1.65	1.25	2.70	0.40	0.30	0.65
		possible range		0.60	5.00		0.20	0.90

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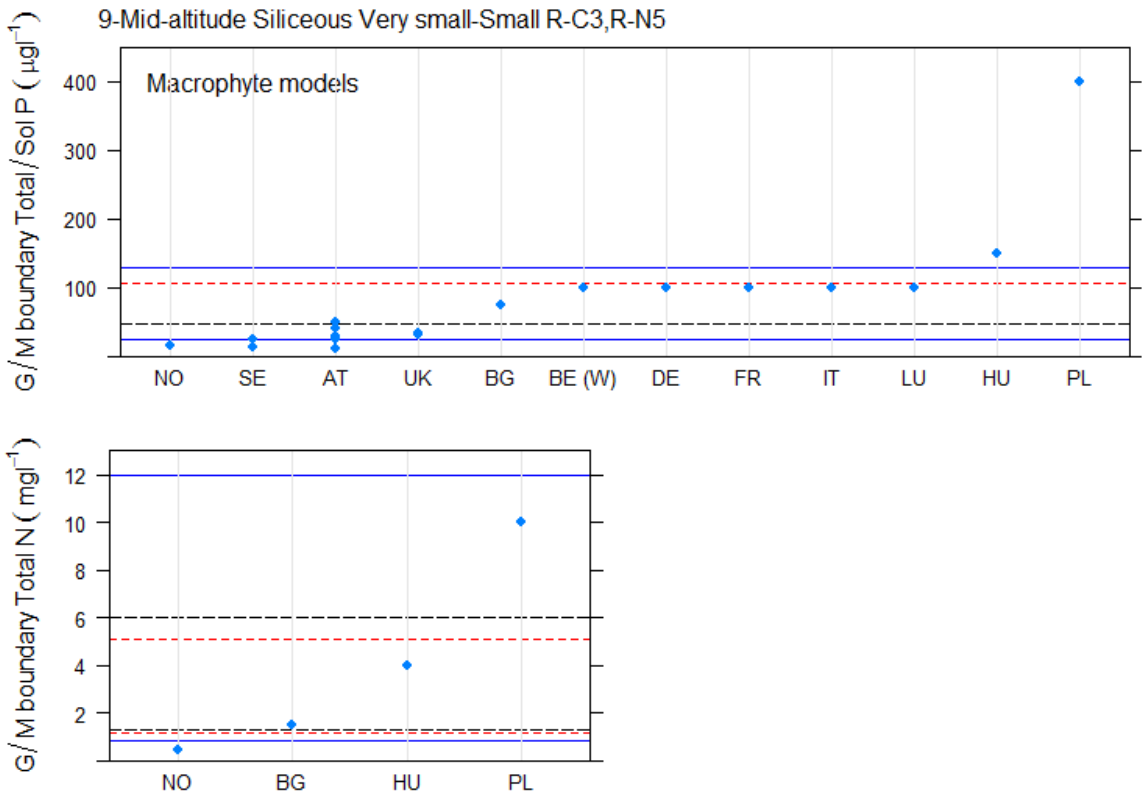

Figure 4-8 Comparison of range of reported good/moderate boundary values for a) ortho-phosphorus boundary values and b) total nitrogen for broad type 9 mid-altitude siliceous very small-small rivers in comparison to range of modelled values of soluble P and TN for low alkalinity upland rivers (including intercalibration type R-C3) using macrophytes. Most likely range (black broken line), best model upper/lower quartiles of model residuals (red dotted line), possible range (blue solid line) This figure was reproduced from Phillips & Pitt (2015) phosphorus boundary values for AT, BE(W) and FR are for 90th percentiles, those for AT are soluble phosphorus.

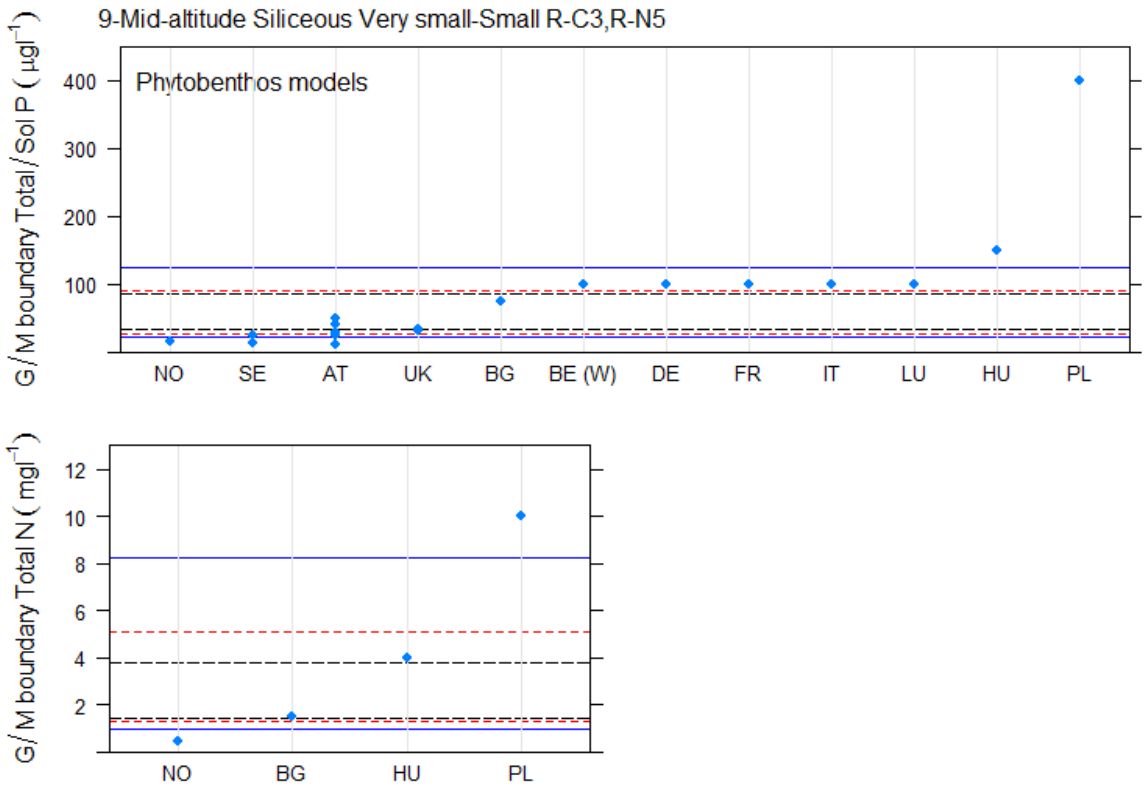

Figure 4-9 Comparison of range of reported good/moderate boundary values for a) ortho-phosphorus boundary values and b) total nitrogen for broad type 9 mid-altitude siliceous very small-small rivers in comparison to range of modelled values of soluble P and TN for low alkalinity upland rivers (including intercalibration type R-C3) using phytobenthos. Most likely range (black broken line), best model upper/lower quartiles of model residuals (red dotted line), possible range (blue solid line). This figure was reproduced from Phillips & Pitt (2015) phosphorus boundary values for AT, BE(W) and FR are for 90th percentiles, those for AT are soluble phosphorus.

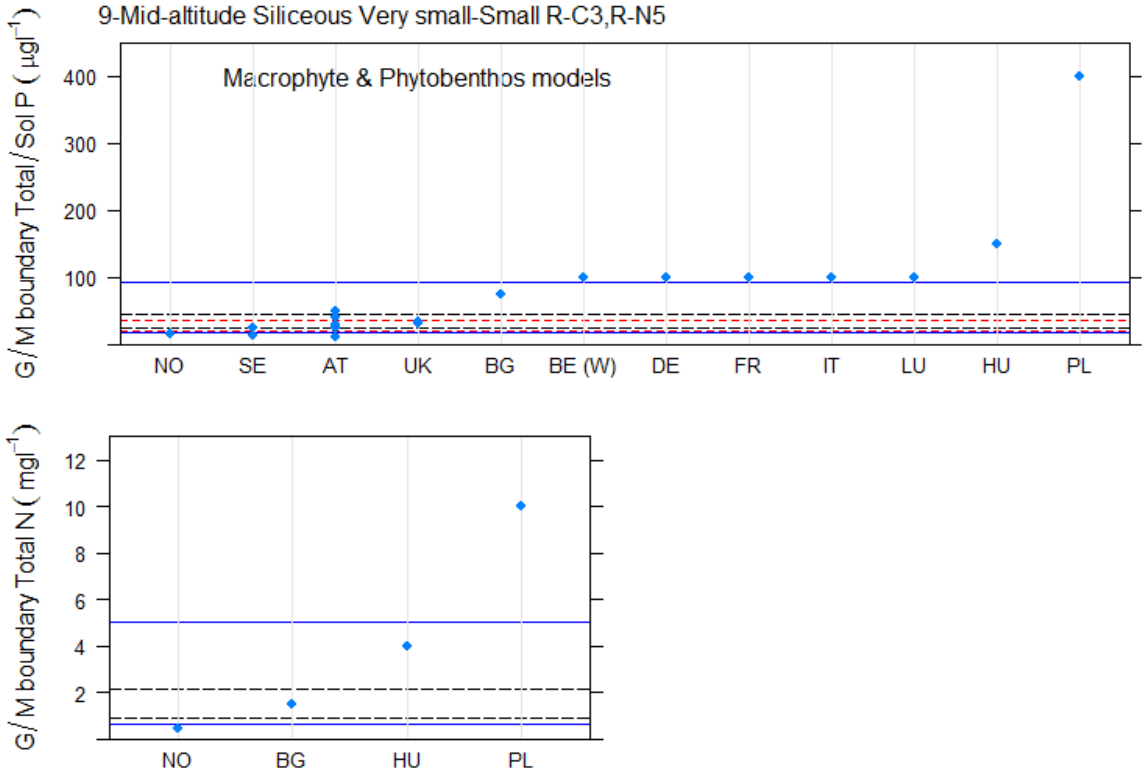

Figure 4-10 Comparison of range of reported good/moderate boundary values for a) ortho-phosphorus and b) total nitrogen for broad type 9 mid-altitude siliceous very small-small rivers in comparison to range of modelled values of ortho- P and TN for low alkalinity upland rivers (including intercalibration type R-C3) using combined macrophytes and phytobenthos. Most likely range (black broken line), best model upper/lower quartiles of model residuals (red dotted line), possible range (blue solid line). This figure was reproduced from Phillips & Pitt (2015) phosphorus boundary values for AT, BE(W) and FR are for 90th percentiles, those for AT are soluble phosphorus.

4.4 Very large rivers (broad type 1)

Phytobenthos and macroinvertebrate data were available for very large rivers. The datasets compiled during the X-GIG intercalibration of large rivers were used for this analysis.

Phytobenthos correlations with nutrients were sufficiently strong to follow the approaches outlined in chapter 3. Macroinvertebrates however, are strongly influenced by hydromorphology and other factors. This causes poor correlations with the nutrients. Hence, other approaches have to be explored to gain an idea about nutrient boundaries in these cases. Consequently, phytobenthos and macroinvertebrates are presented in separate chapters.

4.4.1 Phytobenthos

This included data for 10 countries for medium and high alkalinity rivers and two countries for low alkalinity rivers (Table 4-7). Data for soluble and total phosphorus as well as

nitrate-nitrogen are available. Biological data are expressed as the phytobenthos ICM, the average of IPS and TI values (Kelly et al. 2009), each expressed as an EQR.

Table 4-6 number of phytobenthos samples per water type and country

Subtype	Country	Number of samples with pressure data	Number of water bodies
Low alkalinity rivers (R-L1)	Finland	19	7
	Sweden	4	4
	Total number	**23**	**11**
Medium- to high alkalinity rivers (R-L2)	Austria	14	11
	Belgium (Flanders)	3	3
	Belgium (Wallonia)	4	2
	Czech Republic	24	7
	Estonia	5	2
	Germany	24	12
	Hungary	26	16
	Netherlands[d]	92	10
	Slovakia	37	3
	Slovenia	26	11
	Total number	**255**	**77**

The small quantity of data available for low alkalinity, coupled with the relatively short gradient (most predictions of the good/moderate boundary require extrapolation) mean that only results for medium and high alkalinity rivers are included here. However, it is clear from the results that nutrient boundaries for low alkalinity rivers are much lower than medium and high alkalinity rivers.

The range of boundaries for very large rivers (excluding low alkalinity rivers) is shown in Table 5-6 – Table 5-8. The majority of the reported member state boundary values for broad type 3, lowland siliceous very small-small rivers, were within the most likely range of the predicted boundaries. SE and NO have lower boundaries, because they have predominantly low alkalinity large rivers. For low alkalinity very large rivers the modelled boundary values are quite imprecise, due to the above mentioned reasons. Nevertheless, it is sure that they are much lower than for the medium and high alkalinity very large rivers. Only two countries are above the predicted range for orthophosphate and only one above the range for nitrate.

Table 4-7 Summary of predicted soluble phosphorus (sol-P) boundaries for medium and high alkalinity very large rivers

IC Type	BQE used		GM sol-P μgl^{-1}			HG sol-P μgl^{-1}		
			Pred	range		Pred	Range	
	Min	most likely boundary		40	56		16	39
		best model R^2 0.357	46	27	105	20	12	34
		possible range		27	117		8	39

Table 4-8 Summary of predicted total phosphorus boundaries for medium and high alkalinity very large rivers

IC Type	BQE used		GM TP μgl^{-1}			HG TP μgl^{-1}		
			Pred	range		Pred	Range	
	Phytobenthos	most likely boundary		33	130		11	60
		best model R^2 0.406	37	25	52	18	12	25
		possible range		20	130		7	60

Table 4-9 Summary of predicted nitrate-nitrogen boundaries for medium and high alkalinity very large rivers

IC Type	BQE used		GM TN mgl^{-1}			HG TN mgl^{-1}		
			Pred	range		Pred	Range	
	Phytobenthos	most likely boundary		1.6	2.5		0.52	0.96
		best model R^2 0.236	1.64	1.1	3.3	0.80	0.55	1.4
		possible range		1.1.	3.3		0.21	1.4

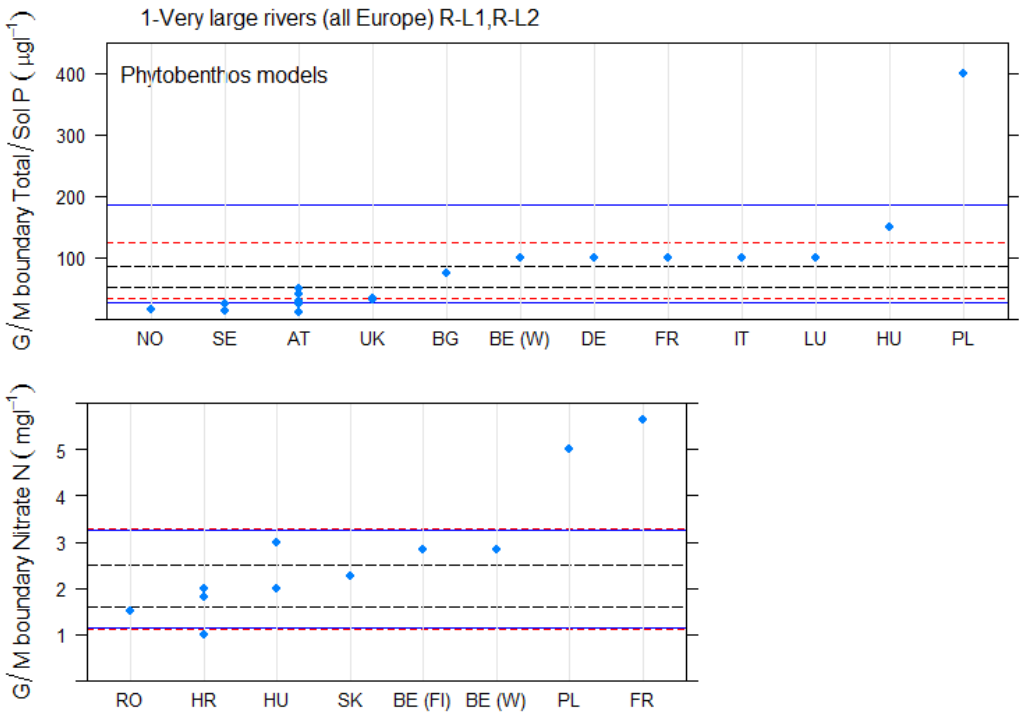

Figure 4-11 Comparison of range of reported good/moderate boundary values for a) total phosphorus (TP) and b) nitrate nitrogen for broad type 1 very large rivers in comparison to range of modelled values of TP and nitrate for medim to high alkallinity very large rivers using phytobenthos. Most likely range (black broken line) best model upper/lower quartiles of model residuals (red dotted line), possible range (blue solid line). No and SE have predominantly low alkalinity rivers, in which modelled nutrient boundaries are much lower. This figure was reproduced from Phillips & Pitt (2015) all phosphorus boundary values are for TP those of BE(W), FR, RO,SK are for 90th percentiles, those for AT are soluble phosphorus. For TN values for RO & SK are for 90th percentiles

4.4.2 Macroinvertebrates

The intercalibration exercise for macroinvertebrates is not fished. Hence the biological boundaries are not finally intercalibrated.

The correlations with the nutrients were generally poor. This was to be expected, because the macroinvertebrate assessment results are even more influenced by hydromorphology. This leads to wedge shaped relationship (Figure 4-12) as described by Kail et al. (2012).

Therefore the regression analysis was not possible, it would lead to much too low boundary values. Similarly, the categorical mismatch approach is also likely to lead to boundary values which are most too low (yellow line in Figure 4-12), because many samples have a moderate macroinvertebrate status as a result of other pressures, despite very low nutrient levels. This illustrates the difficulty of using relationships between a biological quality element and supporting nutrients when the biological status can be significantly lowered by other pressures.

In Germany a method was developed to derive good/moderate status boundaries for chemical parameters from such wedge shaped distributions (umweltbüro essen and chromgruen 2014). For this purpose an upper threshold or borderline concentration was

produced using a categorical approach, the regression line of the maximum P-values without outliers for each biological status class. It can also be derived graphically.

The application of this approach to the very large rivers macroinvertebrates is illustrated by the red line in Figure 4-12. Naturally, this approach gives higher boundary values than the regression approaches, because it uses the maximum values without outliers instead of the confidence limits, but the resulting values are absolutely limiting for sustaining the corresponding ecological status.

Using the borderline upper limit and the concentrations suggested using the mismatch method as lower limits a very rough indication of possible boundary range can be given (Table 4-10). However further work is needed to develop this approach and identify a way to reduce this range.

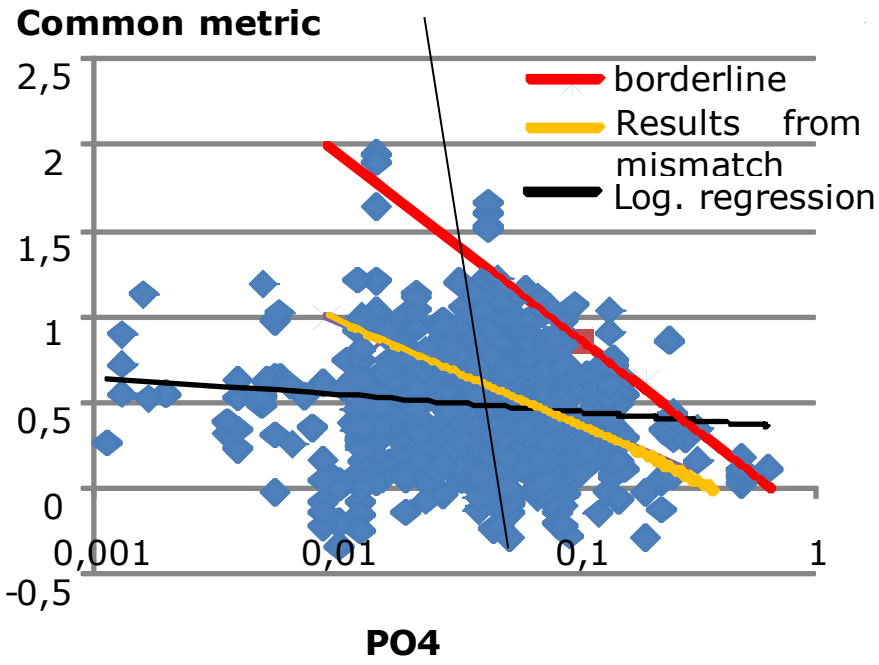

Figure 4-12 Wedge shaped dose response relationship between orthophosphate and macroinvertebrate ICM. The regression lines from x to y as well as the other way round (black lines) are almost perpendicular on each other and hence meaningless. The red "borderline" gives the maximum P-level still supporting the corresponding assessment value. The yellow line resulted from the class mismatch approach, which underestimates the boundaries for such a distribution. Consequently the red and yellow lines give a rough possible range for the P-boundary values.

Table 4-10 Summary of predicted soluble phosphorus (sol-P) boundaries for very large rivers using invertebrates, derived from the "borderline analysis"

IC Type	BQE used		GM sol-P µgl^{-1}			HG sol-P µgl^{-1}		
			Pred	range		Pred	Range	
	Invertebrates	most likely boundary		Not available			Not available	
		best model R^2 0.18	ns					
		possible range		40	170		16	105

5 Discussion

5.1 Uncertainty of relationships

Pressure response relationships provide an objective method of establishing the "levels" of nutrients that would be required to support good ecological status. Fitting regression models allows nutrient concentrations to be determined that are most likely to occur at the biological boundary values of high/good and good/moderate status. The relationships are, however often uncertain, in the case of rivers 54% of the relationships tested were not significant and, of the significant relationships in both lakes and rivers, the majority explain only 35-45% of the variation (Figure 5-1). There were few differences in uncertainty between BQEs, except for phytoplankton which had markedly higher R^2 for phosphorus and lower for nitrogen. This variability is not surprising as many factors are likely to influence ecological status, but it needs to be taken into consideration when comparing the predicted boundary values from the models with those reported by member states.

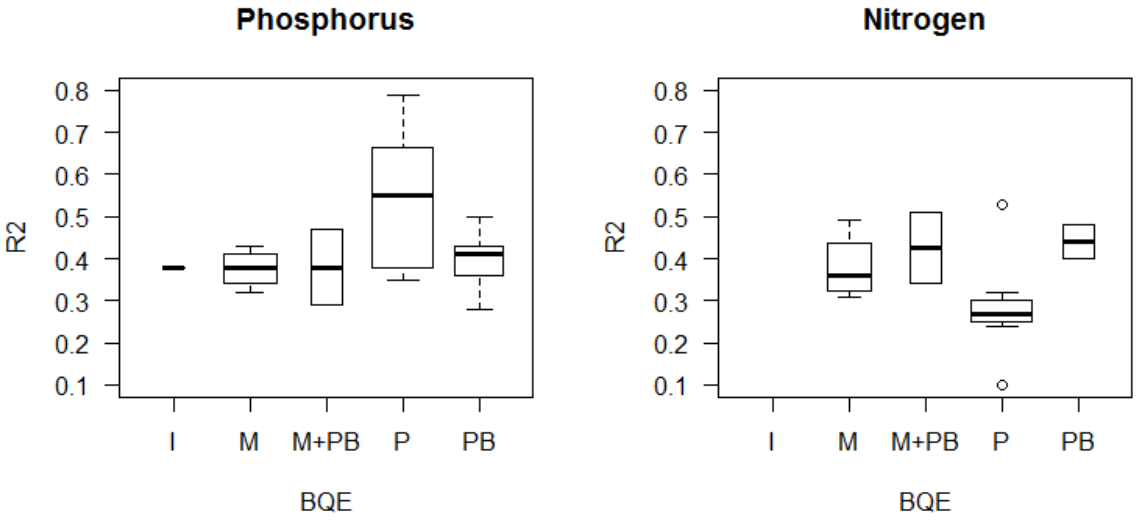

Figure 5-1 Range of R^2 values for regressions between different BQEs and a) Phosphorus or b) Nitrogen in lakes and rivers. (Invertebrates = I, macrophytes = M, macrophytes and phytobenthos = M+PB, phytoplankton = P, phytobenthos = PB)

When variability is high the regression approach used will influence boundary values. Conventional OLS is likely to underestimate slopes which, depending on the mean value of the data used, is likely to overestimate good/moderate and underestimate high/good boundary values. Conversely type I OLS regression where the variation in the nutrient concentration is minimised, over estimates slopes, over and under estimating the good/moderate and high/good boundary values respectively. Type II regression (Reduced Major Axis) which minimises variation in both nutrient and biological variables produces a slope intermediate to the OLS regressions. Thus different regression approaches produce a range of slopes and, as a result, different predicted boundary values from the same data set, with the greatest differences where uncertainty is greatest.

Regression also allows the uncertainty of parameters to be determined and thus for a particular model a range of potential regression lines can be determined. For simplicity in our analysis we present the upper and lower quartiles of regression residuals, which

approximate to the regressions ± 1 standard error. These lines, therefore, represent the range of relationships that might be determined from other similar data sets, as might be used by individual member states. In other words, different member states, even using the same regression approach for a similar water body type are likely to determine different boundary values, depending on the method, the data set and, in particular, the length of gradient available. The range of potential boundary values is often relatively high and is typically similar to the range of the majority of reported boundary values.

5.2 Interpretation of relationships

The above discussion assumes that boundary values for nutrients are determined using the best fit regression line (Figure 5-2a). This will provide values that minimise the mis-match between biological and nutrient classifications, but depending on the purpose of the nutrient boundary value upper or lower lines reflecting uncertainty may be used. The use of the upper line minimises the risk of a water body being wrongly downgraded (Figure 5-2b), the lower line is more protective but will result in more waterbodies being wrongly downgraded (Figure 5-2c).

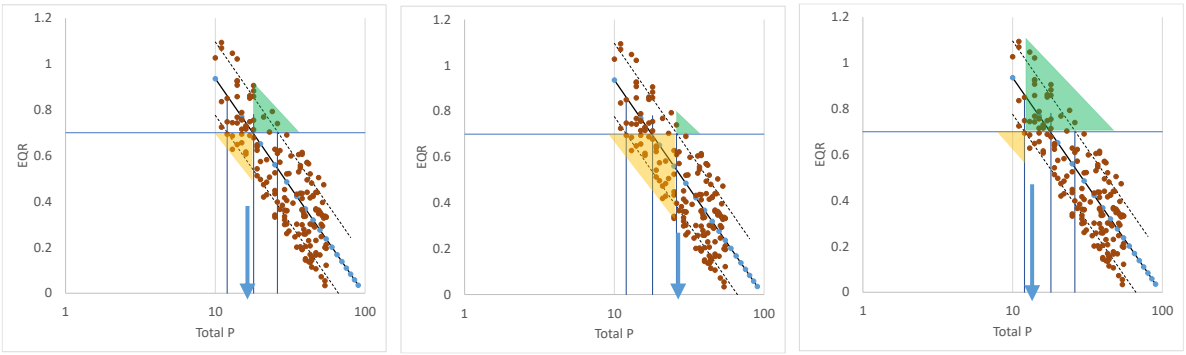

Figure 5-2 Hypothetical relationship between total phosphorus and biological EQR, showing regression line with confidence intervals (dotted lines). Horizontal line shows the biological good/moderate boundary, vertical lines show intersection with regression line ± confidence intervals marking potential good/moderate boundary values for total phosphorus using, a) intersection with best fit line, b) upper confidence line, c) lower confidence line. Triangles mark areas where classification mismatches occur, green (biology Good but phosphorus Moderate) and yellow (biology Moderate or worse but phosphorus Good) using three different approaches to interpretation.

In rivers, many more factors other than nutrients influence biological status, particularly when BQEs such as invertebrates are considered. In these cases relationships between nutrient concentration and biological status have a very high uncertainty. A scatter plot may show a "wedge" type relationship to which an upper quantile line can be fitted which provides an estimate of the highest level of nutrient that is consistent with good status (Figure 5-3). Although this is an extreme example it illustrates the difficulty of identifying an appropriate boundary where multiple pressures exist.

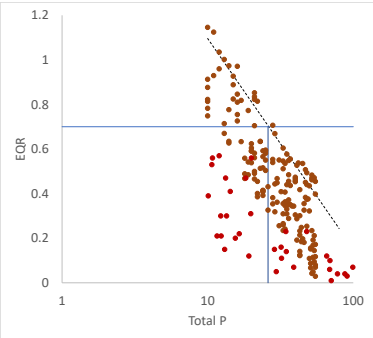

Figure 5-3 Hypothetical relationship between total phosphorus and biological EQR where multiple pressures occur, showing regression of upper quantile value (e.g. 95th percentile). Horizontal line shows the biological good/moderate boundary, vertical lines show intersection with line marking potential good/moderate boundary values for total phosphorus.

The choice of line depends on the purpose of the boundary value but the important point is that, given the uncertainty of even national type specific relationships, there will always be a range of potential boundary values from which a member state can choose. The choice will also reflect how the boundary value is used within the country and, therefore, it is important to recognise this as a further factor influencing variation between national standards. Broadly, two strategies may be adopted:

- **Action** (e.g. programmes of measures) is triggered as soon as the nutrient boundary is exceeded. Under such circumstances, a higher boundary value may be appropriate in order to minimise the instances where biology is at good status despite the elevated nutrient concentrations (i.e. Fig. 22b)
- An exceedance of the nutrient boundary is one of a number of strands of **evidence** that is considered before a programme of measures is triggered. Under such circumstances, a more precautionary (lower levels) boundary value may be selected; however, the country would then check that for a particular water body a BQE was also failing prior to taking action, or that there was other evidence that it might do so in the future, for example if there was evidence of increasing nutrient concentrations.

This, in turn, raises questions about the role of supporting element standards. It is clear from this report that the relationships are rarely sufficiently strong enough to indicate convincing cause-effect relationships between nutrients and BQEs. Indeed, the scale of uncertainty in the relationships is a timely reminder that we are attempting to detect the effect of a single stressor within a multi-stressor environment. There is, nonetheless, a need for regulators to unpick the Gordian knot of ecological interactions in order to identify those stressors most likely to be responsible for BQE failures.

Using the analogy of a car dashboard, the BQEs are equivalent to the speedometer, giving drivers an indication of their performance in relation to ecological status boundaries (equivalent to the "speed limit") whilst the supporting elements allow a quick diagnosis of likely causes for the biological "engine" not running as smoothly as desired you might wish (indicated by low EQRs for BQEs). This also allows broad scale overviews of problems and the likely costs for dealing with these to be established. What the supporting element standards do not do is provide an unambiguous indication that status of any particular water body is compromised by one supporting element and not influenced by another.

5.3 Alternative approaches and high uncertainty

The analysis presented here suggests that categorical methods and the method where classification mismatches are minimised produce similar boundary values to the regression approaches. They may be particularly useful where uncertainty is high. However, several of the data sets used for this report produced very weak relationships. The reasons for this are not clear, in some cases it may be the result of using pan-European data sets with the inevitable range of sampling strategies influencing the values of the summary nutrient metrics, but it is also probably a reflection of the many factors that influence biological status, whether pressure-related, intrinsic or stochastic. In these cases it is very difficult to produce general models that can be used to determine boundary values. One approach is to fit a line to an upper quantile of the data, as was used for large river invertebrates. However, this produces a relatively high boundary value representing the highest nutrient value observed at the biological boundaries. Higher nutrient values will not support the corresponding biological status classes any more. On the other hand this upper boundary may mark the only "relatively stable line" (the limiting effect) within such very common wedge-shaped data patterns. Describing this upper boundary line might be therefore an alternative way to describe the relations between two factors, when the dependent one is affected not only by the independent factor as it is the case in most multi-pressure and multi-factor environments. Although antagonistic pressure-effects might be able to affect the stability and position of this upper boundaries in wedge shaped data distributions, a combination of this approach with the application of safety factors as, it is commonly done for the derivation of ecological quality standards for pollutants might be a conceivable way for the determination of G/M Boundaries under the WFD. Another approach to reduce the uncertainty would be to include further relevant factors into the models, like hydromorphology for macroinvertebrates. However such data are not easily available and it still needs to be investigated, if the combined effects are additive, multiplicative or follow some other principle.

5.4 Wider considerations

Finally it is important to remind ourselves that the WFD makes it clear that the purpose of establishing boundary values for nutrients is to ensure the functioning of the ecosystem, not simply to ensure that BQEs achieve good or better status ("nutrient concentrations do not exceed the levels established so as to ensure the functioning of the ecosystem and the achievement of the values specified for the biological quality elements" WFD Annex V, Section 1.2).

By using pressure response relationships to determine equivalence between a biological good/moderate boundary and nutrient concentration we are assuming that our biological indicators do indeed reflect ecological function. The intercalibrated WFD biological methods are our current best available assessments of ecological function, however they are not perfect and it is important to place the wider ecological literature alongside empirical analysis. For example it is widely reported that the response to phosphorus in lakes by phytoplankton reaches a plateau at concentrations above 100 $\mu g l^{-1}$ (Maberly et al. 2002; Phillips et al. 2008; Reynolds 1992). Although nutrient responses in rivers are more difficult to quantify, significant ecological changes do occur in rivers at similar concentrations (Hilton et al. 2006; Mainstone 2010). Thus, further consideration of whether the functioning of the ecosystem is still ensured might be needed where

good/moderate boundary values are substantially greater than these levels, particularly where they are derived from either weak relationships or where multiple pressures exist.

5.5 Conclusions

- Pressure response relationships provide an objective method for determining nutrient boundary values. The use of regression methods allows uncertainty to be determined and thus provides a method of determining a range of potential boundary values which would represent different levels of precaution of the supporting element. We thus recommend the use of regression over categorical methods when good relationships can be determined from the data. Further work is needed to determine what represents an adequate relationship but currently we suggest that the R^2 value should be ≥ 0.36. Where uncertainty is high and scatter plots suggest a "wedge" shaped relationship (e.g. Figure 4-12) quantile regressions may be used in combination with other methods to provide ranges of potential boundary values, although further work is needed to develop this approach.
- In most cases the most appropriate regression methods are to use either multivariate OLS models with both nitrogen and phosphorus as predictor variables or to use type II (RMA) regression. Relationships with low R^2 values (<0.36) need to be treated with caution (and then require justification).
- Relationships should cover as wide a range of pressure as possible and predicted boundary values beyond the range of the data should not be used, or treated with caution. It is recommended that where national type specific data sets have a limited range of pressures consideration is given to combining water body types prior to analysis or by including data from similar water body types from neighbouring countries.
- Categorical methods provide equally good estimates of boundary values, the method of minimising mismatch of classification is potentially useful as it has a clear and simple objective.
- In this report we present 4 methods, multivariate OLS regression, RMA regression, box plots and minimising mismatch of classifications. We suggest that where strong regression relationships are found (R2 >0.6) univariate or multivariate OLS regression provides a reliable estimate of boundary values. Where there is greater uncertainty it is likely to be more reliable to use type II RMA regression. The box plot and mismatch methods may be particularly useful where pressure gradients are short. At present the mismatch method does not provide any estimate of uncertainty, although it is a method that provides a very clear outcome that is easily understood by non-experts.
- In our analysis we used data collated for the intercalibration exercise. The regressions are derived from data of similar water body types from several countries and should thus represent a general type specific relationship for the water body type. We recommend that the methods should be applied to national data sets to determine national boundary values. For national data sets that are from similar water body types to those we used it is likely that the predicted boundary values will fall within the range of the values we report.

- Recognise limits of nutrient-BQE relationships in terms of indicating causal relationships (i.e. recognise that high uncertainty is inevitable and deal with it by moving to stronger diagnostic tools). This leaves us with a broader question: how far can we go with nutrient standards based on pressure-response, given all that we have shown? Is it thus necessary to reconsider the role of nutrient boundary values and can we develop an approach that could lead to a code of best practice for diagnosing nutrient-based problems?
- We also suggest that it would be useful to develop guidance on the determination of nutrient supporting element boundary values, perhaps as a supplement to the current Eutrophication Guidance. Such guidance could be supported by a statistical tool kit and structured in a way that could lead a member state through the process of determining a potential range of boundary values that would provide for different probabilities of supporting good ecological status.

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6 Appendix containing details of models

6.1 High alkalinity lakes Central Baltic GIG

6.1.1 Phytoplankton IC Type L-CB1 high alkalinity shallow

Note figures for L-CB1 lakes in main text

Table 6-1 Regression parameters for L-CB1 lake phytoplankton v total phosphorus and total nitrogen

```
Call:
lm(formula = CM.EQR ~ log10(total.P) + log10(total.N), data = data.cc.ex,
    subset = total.P > P.minUsed & total.P <= P.maxUsed & total.N >
        N.minUsed & total.N <= N.maxUsed)

Residuals:
     Min       1Q   Median       3Q      Max
-0.44848 -0.11221 -0.02407  0.11574  0.65430

Coefficients:
               Estimate Std. Error t value Pr(>|t|)
(Intercept)     1.59884    0.06741  23.719  < 2e-16 ***
log10(total.P) -0.58616    0.04387 -13.362  < 2e-16 ***
log10(total.N) -0.18662    0.05133  -3.636 0.000332 ***
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 0.1762 on 270 degrees of freedom
Multiple R-squared:  0.5529,    Adjusted R-squared:  0.5495
F-statistic: 166.9 on 2 and 270 DF,  p-value: < 2.2e-16
```

Table 6-2 Regression parameters for L-CB1 lake phytoplankton v total phosphorus

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x =
"interval", nperm = 99)

n = 431   r = -0.7277776   r-square = 0.5296602
Parametric P-values:   2-tailed = 2.850313e-72    1-tailed = 1.425156e-72
Angle between the two OLS regression lines = 17.88685 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS  1.779693 -0.6925837       -34.70584              0.01
2     MA  2.135886 -0.9341840       -43.05110              0.01
3    SMA  2.161625 -0.9516420       -43.58061                NA
4    RMA  2.053641 -0.8783985       -41.29602              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS       1.686812        1.872574 -0.7545172  -0.6306501
2     MA       2.017539        2.264537 -1.0214457  -0.8539109
3    SMA       2.073284        2.255902 -1.0155887  -0.8917216
4    RMA       1.941293        2.173352 -0.9595964  -0.8021944

Eigenvalues: 0.1246232 0.01956908

H statistic used for computing C.I. of MA: 0.001989921
```

Table 6-3 Regression parameters for L-CB1 lake phytoplankton v total nitrogen

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x =
"interval", nperm = 99)

n = 267   r = -0.5289825   r-square = 0.2798225
Parametric P-values:   2-tailed = 1.183349e-20    1-tailed = 5.916746e-21
Angle between the two OLS regression lines = 33.4243 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS 0.6723015 -0.6792272       -34.18542              0.01
2     MA 0.5773780 -1.5857179       -57.76327              0.01
3    SMA 0.6089698 -1.2840258       -52.08852                NA
4    RMA 0.6118840 -1.2561955       -51.47830              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS      0.6413614       0.7032416 -0.8110246  -0.5474299
2     MA      0.5397961       0.6057661 -1.9446141  -1.3146202
3    SMA      0.5944621       0.6220645 -1.4225695  -1.1589749
4    RMA      0.5836108       0.6353484 -1.5261962  -1.0321175

Eigenvalues: 0.09203592 0.02533058

H statistic used for computing C.I. of MA: 0.007664919
```

6.1.2 Macrophytes IC Type L-CB1 high alkalinity shallow

Note figures for L-CB1 lakes in main text

Table 6-4 Regression parameters for L-CB1 lake macrophyte v total phosphorus and total nitrogen

```
Call:
lm(formula = CM.EQR ~ log10(total.P) + log10(total.N), data = data.cc.ex,
    subset = total.P > P.minUsed & total.P <= P.maxUsed & total.N >
        N.minUsed & total.N <= N.maxUsed)

Residuals:
      Min        1Q    Median        3Q       Max
-0.260212 -0.064310  0.006819  0.067945  0.191125

Coefficients:
               Estimate Std. Error t value Pr(>|t|)
(Intercept)     0.81463    0.04265  19.099  < 2e-16 ***
log10(total.P) -0.13541    0.02544  -5.323 3.07e-07 ***
log10(total.N) -0.12063    0.03454  -3.492 0.000605 ***
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 0.09569 on 177 degrees of freedom
Multiple R-squared:  0.3972,    Adjusted R-squared:  0.3903
F-statistic:  58.3 on 2 and 177 DF,  p-value: < 2.2e-16
```

Table 6-5 Regression parameters for L-CB1 lake macrophyte v total phosphorus

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x =
"interval", nperm = 99)

n = 123   r = -0.6581599   r-square = 0.4331744
Parametric P-values:   2-tailed = 1.319785e-16    1-tailed = 6.598927e-17
Angle between the two OLS regression lines = 18.3915 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS  1.141015 -0.3107299       -17.26158              0.01
2     MA  1.218245 -0.3506828       -19.32489              0.01
3    SMA  1.452986 -0.4721192       -25.27289                NA
4    RMA  1.312258 -0.3993175       -21.76769              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS       1.016192        1.265838 -0.3747029  -0.2467568
2     MA       1.081484        1.361329 -0.4247033  -0.2799330
3    SMA       1.337664        1.584988 -0.5404067  -0.4124606
4    RMA       1.160930        1.481031 -0.4866275  -0.3210321

Eigenvalues: 0.07882921 0.008098454

H statistic used for computing C.I. of MA: 0.004133463
```

Table 6-6 Regression parameters for L-CB1 lake macrophyte v total nitrogen

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x =
"interval", nperm = 99)

n = 120   r = -0.5525082   r-square = 0.3052653
Parametric P-values:   2-tailed = 6.055875e-11    1-tailed = 3.027938e-11
Angle between the two OLS regression lines = 29.3008 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS 0.6129207 -0.3399249       -18.77417              0.01
2     MA 0.6327442 -0.4407212       -23.78410              0.01
3    SMA 0.6670665 -0.6152395       -31.60147                NA
4    RMA 0.6402650 -0.4789622       -25.59266              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS       0.5865103        0.6393310 -0.4334089  -0.2464408
2     MA       0.6098142        0.6578331 -0.5682904  -0.3241297
3    SMA       0.6500699        0.6868408 -0.7157854  -0.5288173
4    RMA       0.6157805        0.6681718 -0.6208595  -0.3544660

Eigenvalues: 0.04769424 0.009486798

H statistic used for computing C.I. of MA: 0.01030048
```

6.1.3 Phytoplankton IC Type L-CB2 high alkalinity very shallow

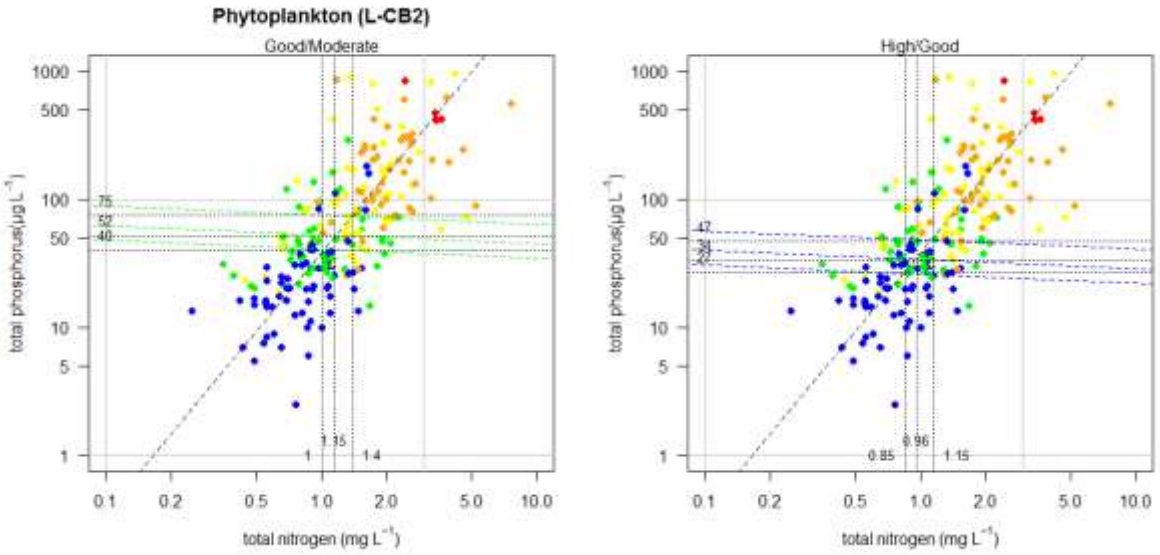


Figure 6-1 Relationship between mean TP and TN, points coloured by WFD class for phytoplankton in high alkalinity shallow CBGIG lakes (Type L-CB2). Dotted lines contours of predicted TN & TP concentration when phytoplankton common metric EQR is at a) good/moderate boundary (green lines) and b) high good boundary, dotted lines show ± 25th & 75th residuals of prediction. Horizontal & vertical lines show intersection with RMA regression of observed TP and TN showing boundary values.

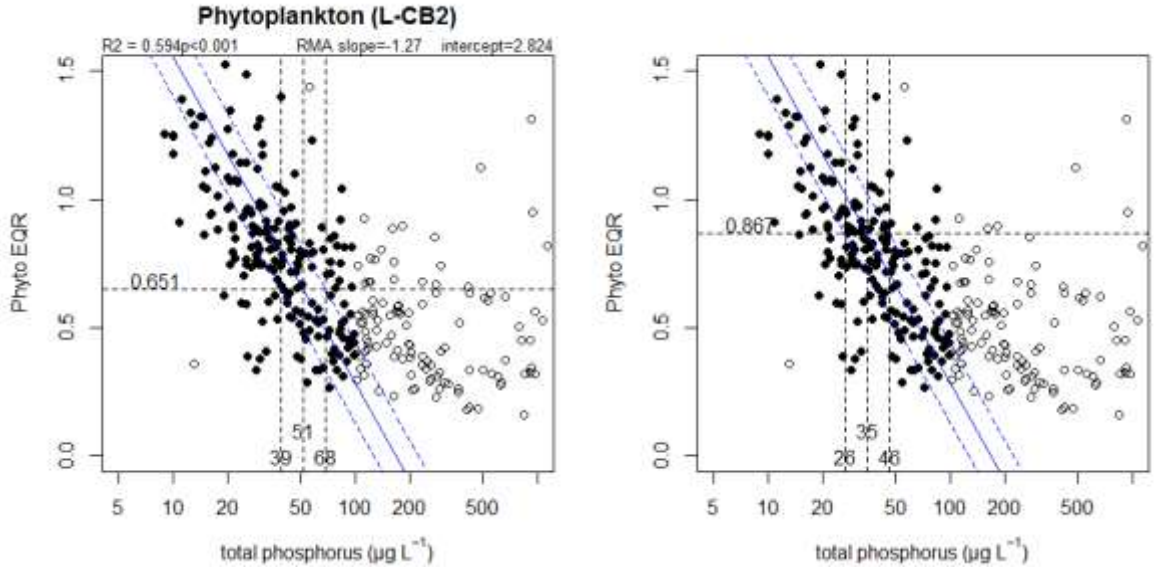


Figure 6-2 Relationship between common metric for phytoplankton and total phosphorus for high alkalinity shallow CBGIG lakes (Type L-CB2) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data, open circles data points excluded from regression.

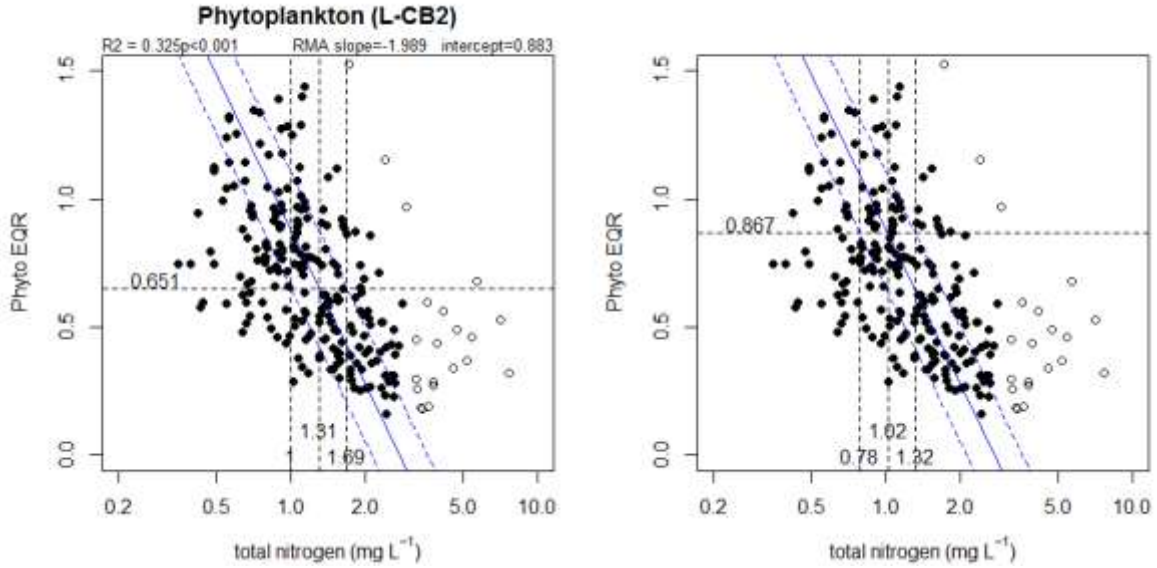


Figure 6-3 Relationship between common metric for phytoplankton and total nitrogen for high alkalinity shallow CBGIG lakes (Type L-CB2) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data, open circles data points excluded from regression.

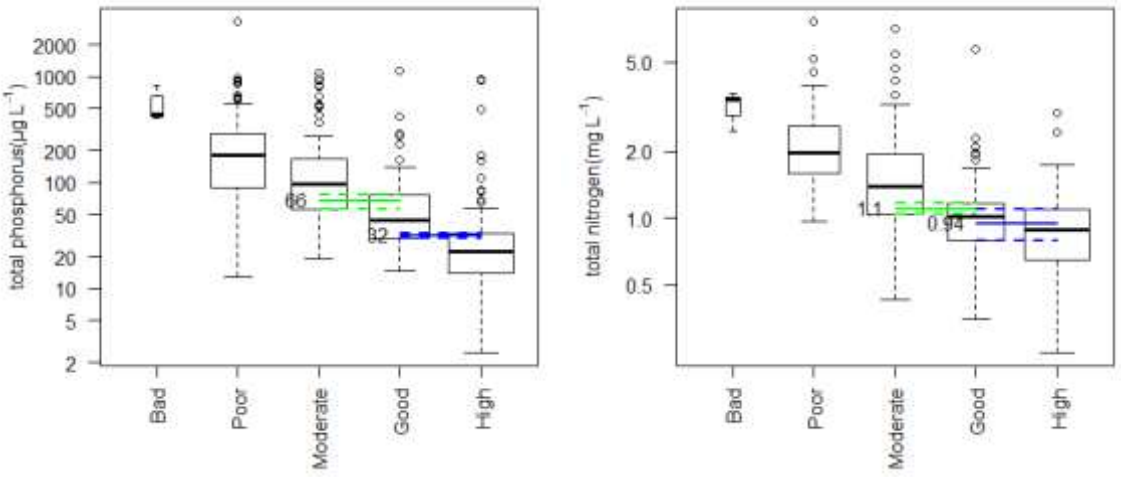


Figure 6-4 Box plots showing range of a) TP and b) TN for high alkalinity shallow CBGIG lakes (Type L-CB2) classified using phytoplankton common metric showing good/moderate boundary & high/good boundary values determined from the average of the upper and lower quartile values

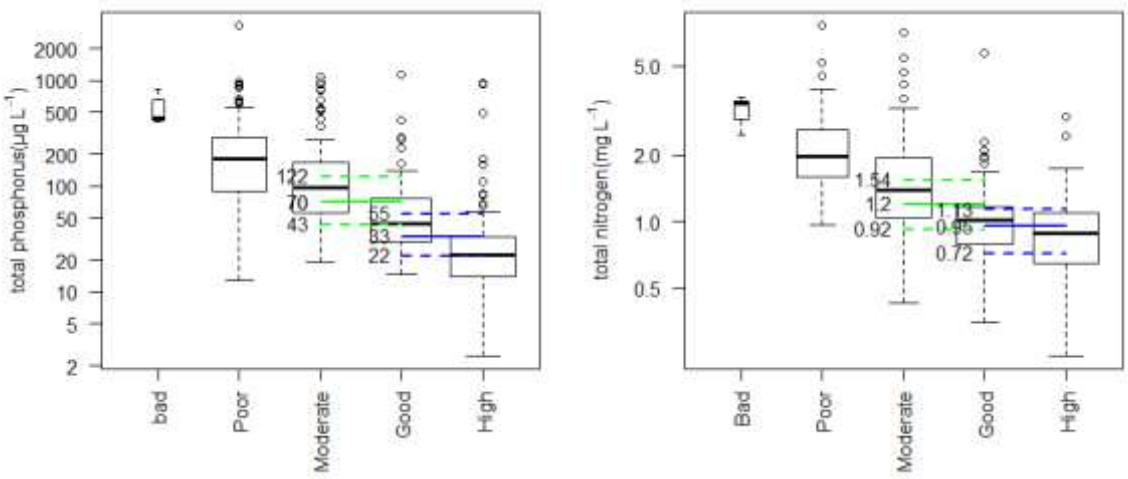


Figure 6-5 Box plots showing range of a) TP and b) TN for high alkalinity shallow CBGIG lakes (Type L-CB2) classified using phytoplankton common metric showing good/moderate boundary & high/good boundary values determined from the average of adjacent classes

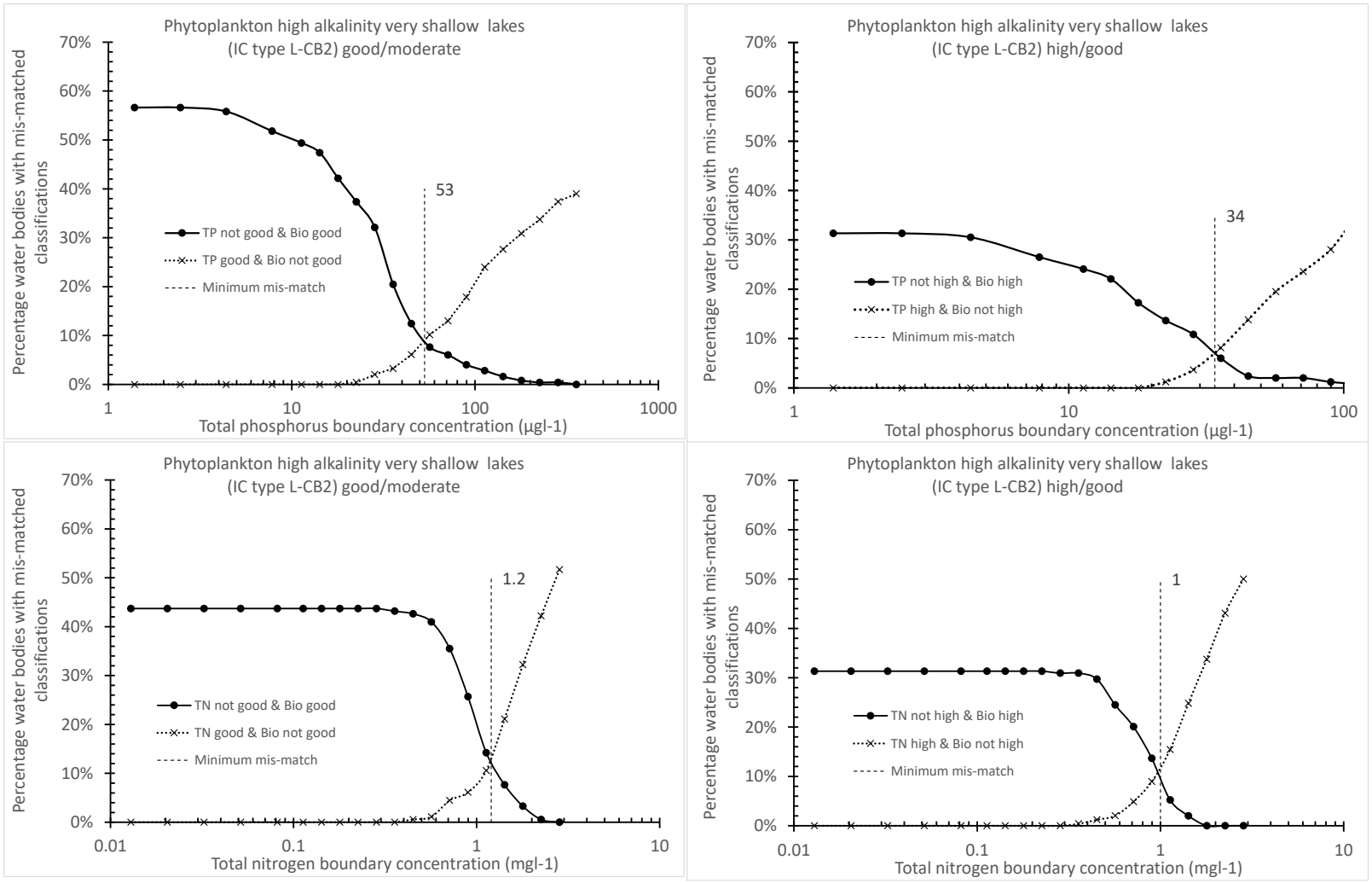


Figure 6-6 Percentage of water bodies where biology or total phosphorus / total nitrogen classifications for good ecological status differ in comparison to the level used to set a) the good/moderate and b) the high/good boundaries. Biological status assessed using the common metric for phytoplankton in high alkalinity very shallow CBGIG lakes (Type L-CB2). Vertical lines mark intersection of curves where mis-match is minimised and equal.

Table 6-7 Regression parameters for L-CB2 lake phytoplankton v total phosphorus and total nitrogen

```
Call:
lm(formula = CM.EQR ~ log10(total.P) + log10(total.N), data = data.cc.ex,
    subset = total.P > P.minUsed & total.P <= P.maxUsed & total.N >
        N.minUsed & total.N <= N.maxUsed)

Residuals:
     Min       1Q   Median       3Q      Max
-0.60460 -0.17345 -0.01684  0.12533  1.11850

Coefficients:
              Estimate Std. Error t value Pr(>|t|)
(Intercept)    2.57861    0.10886  23.688   <2e-16 ***
log10(total.P) -1.12017    0.07097 -15.784   <2e-16 ***
log10(total.N) -0.08054    0.12476  -0.646    0.519
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 0.2536 on 180 degrees of freedom
Multiple R-squared:  0.6761,    Adjusted R-squared:  0.6725
F-statistic: 187.9 on 2 and 180 DF,  p-value: < 2.2e-16
```

Table 6-8 Regression parameters for L-CB2 lake phytoplankton v total phosphorus

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x =
"interval", nperm = 99)

n = 233   r = -0.7705542   r-square = 0.5937537
Parametric P-values:   2-tailed = 4.433742e-47    1-tailed = 2.216871e-47
Angle between the two OLS regression lines = 14.02706 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept     Slope Angle (degrees) P-perm (1-tailed)
1    OLS  2.522617 -1.072451       -47.00218              0.01
2     MA  3.218684 -1.528159       -56.79999              0.01
3    SMA  3.010390 -1.391791       -54.30281                NA
4    RMA  2.823659 -1.269540       -51.77296              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS       2.343802        2.701432  -1.187449  -0.9574521
2     MA       2.985609        3.489647  -1.705557  -1.3755677
3    SMA       2.841981        3.193288  -1.511533  -1.2815357
4    RMA       2.622031        3.038845  -1.410421  -1.1375361

Eigenvalues: 0.2246109 0.02538228

H statistic used for computing C.I. of MA: 0.00241382
```

Table 6-9 Regression parameters for L-CB2 lake phytoplankton v total nitrogen

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x =
"interval", nperm = 99)

n = 257   r = -0.5699522   r-square = 0.3248455
Parametric P-values:   2-tailed = 1.541219e-23    1-tailed = 7.706093e-24
Angle between the two OLS regression lines = 24.61769 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept     Slope Angle (degrees) P-perm (1-tailed)
1    OLS 0.8506234 -1.203559       -50.27786              0.01
2     MA 0.9330267 -3.187843       -72.58379              0.01
3    SMA 0.8883360 -2.111684       -64.65984                NA
4    RMA 0.8832433 -1.989050       -63.30892              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS      0.8065407       0.8947062  -1.417540  -0.9895783
2     MA      0.9127465       0.9611521  -3.865106  -2.6994919
3    SMA      0.8798989       0.8976713  -2.336479  -1.9085174
4    RMA      0.8695528       0.8992139  -2.373624  -1.6593804

Eigenvalues: 0.1977059 0.02544323

H statistic used for computing C.I. of MA: 0.002578088
```

6.1.4 Macrophytes IC Type L-CB2 high alkalinity very shallow

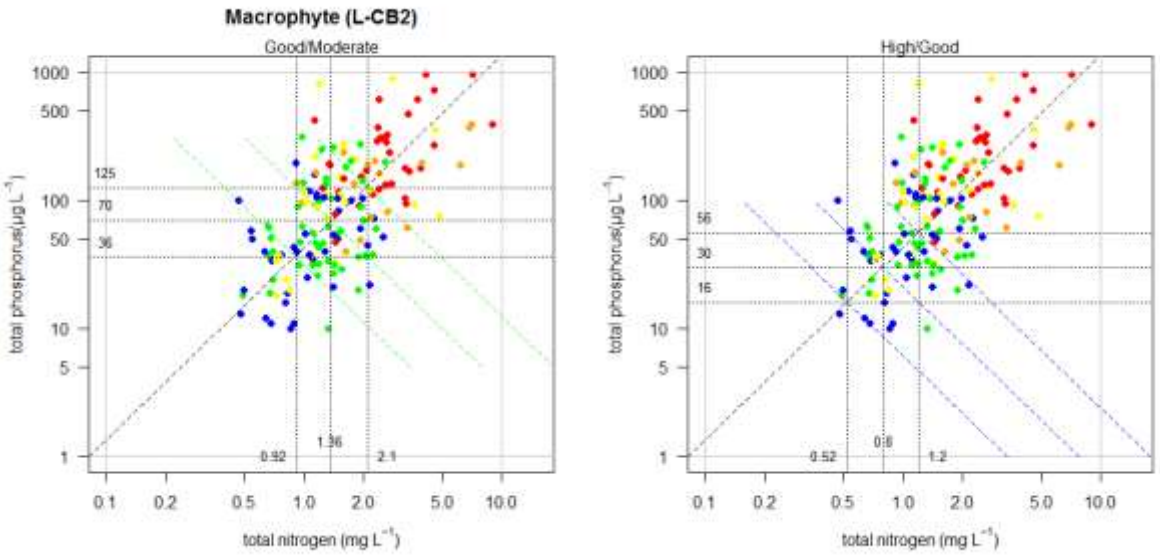


Figure 6-7 Relationship between mean TP and TN, points coloured by WFD class for macrophyte in high alkalinity very shallow CBGIG lakes (Type L-CB2). Dotted lines contours of predicted TN & TP concentration when macrophyte common metric EQR is at a) good/moderate boundary (green lines) and b) high good boundary, dotted lines show ± 25th & 75th residuals of prediction. Horizontal & vertical lines show intersection with RMA regression of observed TP and TN showing boundary values.

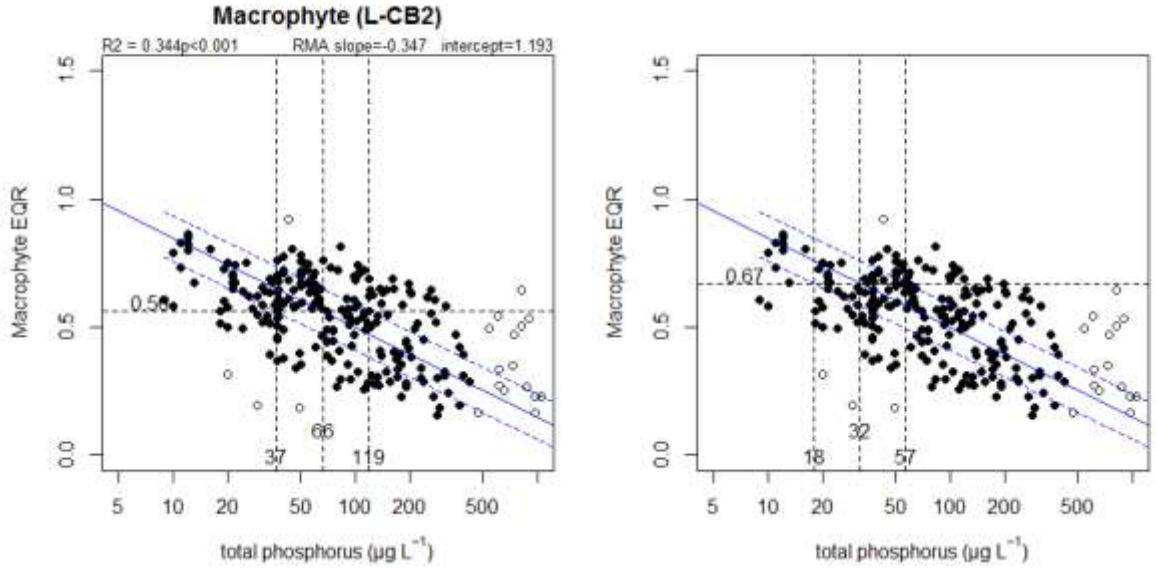


Figure 6-8 Relationship between common metric for macrophytes and total phosphorus for high alkalinity very shallow CBGIG lakes (Type L-CB2) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data, open circles data points excluded from regression. (Details in Table 6-11)

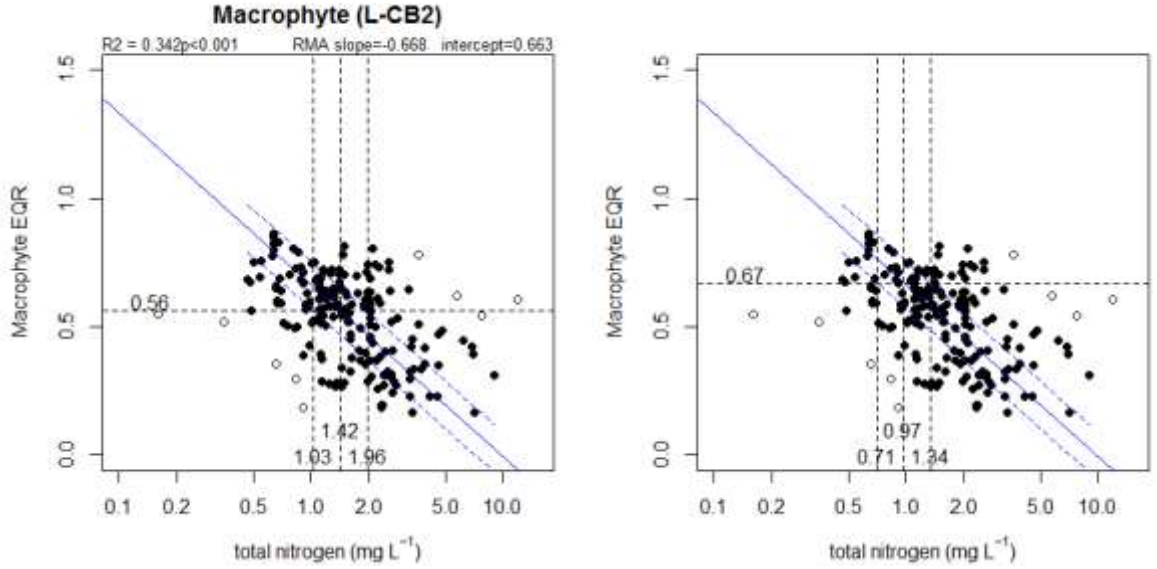


Figure 6-9 Relationship between common metric for macrophytes and total nitrogen for high alkalinity very shallow CBGIG lakes (Type L-CB2) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data, open circles data points excluded from regression (Details in Table 6-12).

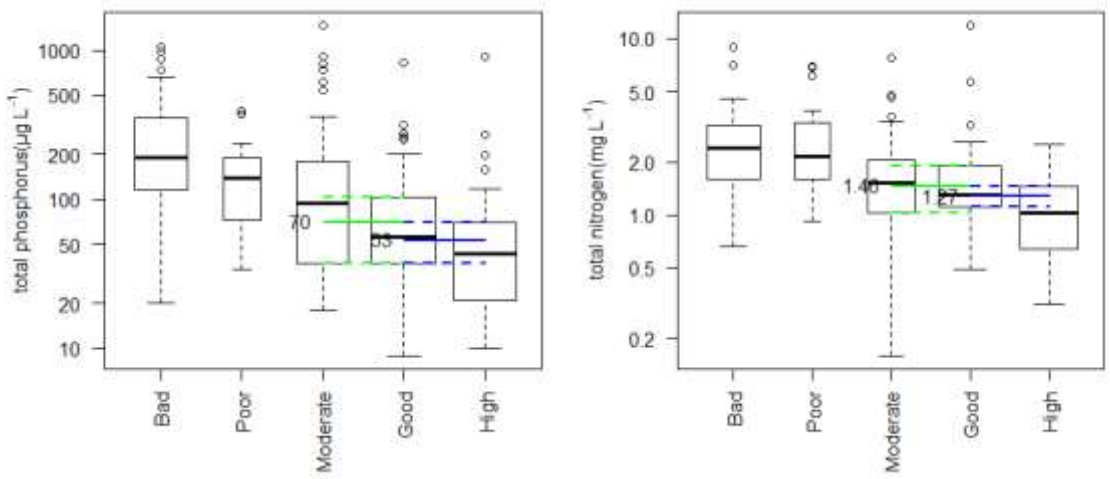


Figure 6-10 Box plots showing range of a) TP and b) TN for high alkalinity very shallow CBGIG lakes (Type L-CB2) classified using macrophyte common metric showing good/moderate boundary & high/good boundary values determined from the average of the upper and lower quartile values of adjacent classes

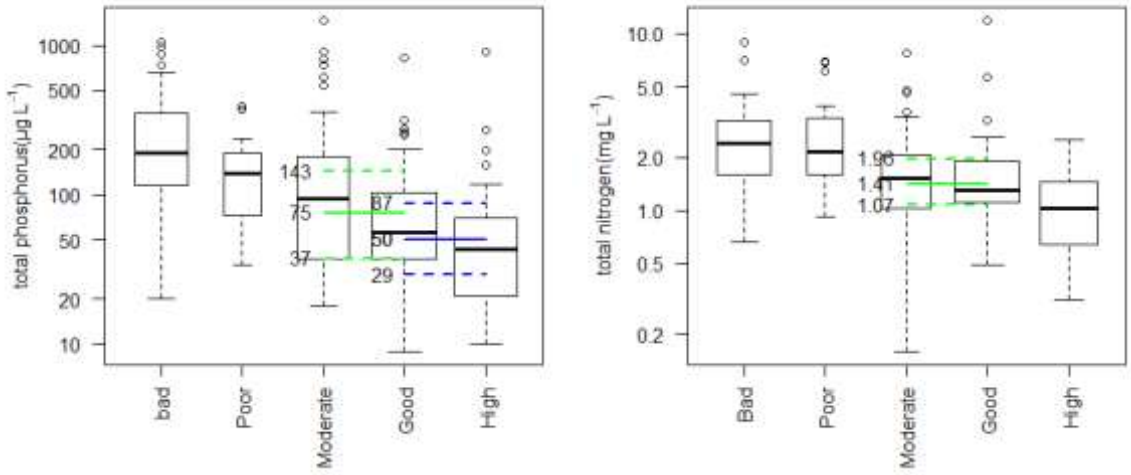


Figure 6-11 Box plots showing range of a) TP and b) TN for high alkalinity very shallow CBGIG lakes (Type L-CB2) classified using macrophyte common metric showing good/moderate boundary & high/good boundary values determined from the average of adjacent classes

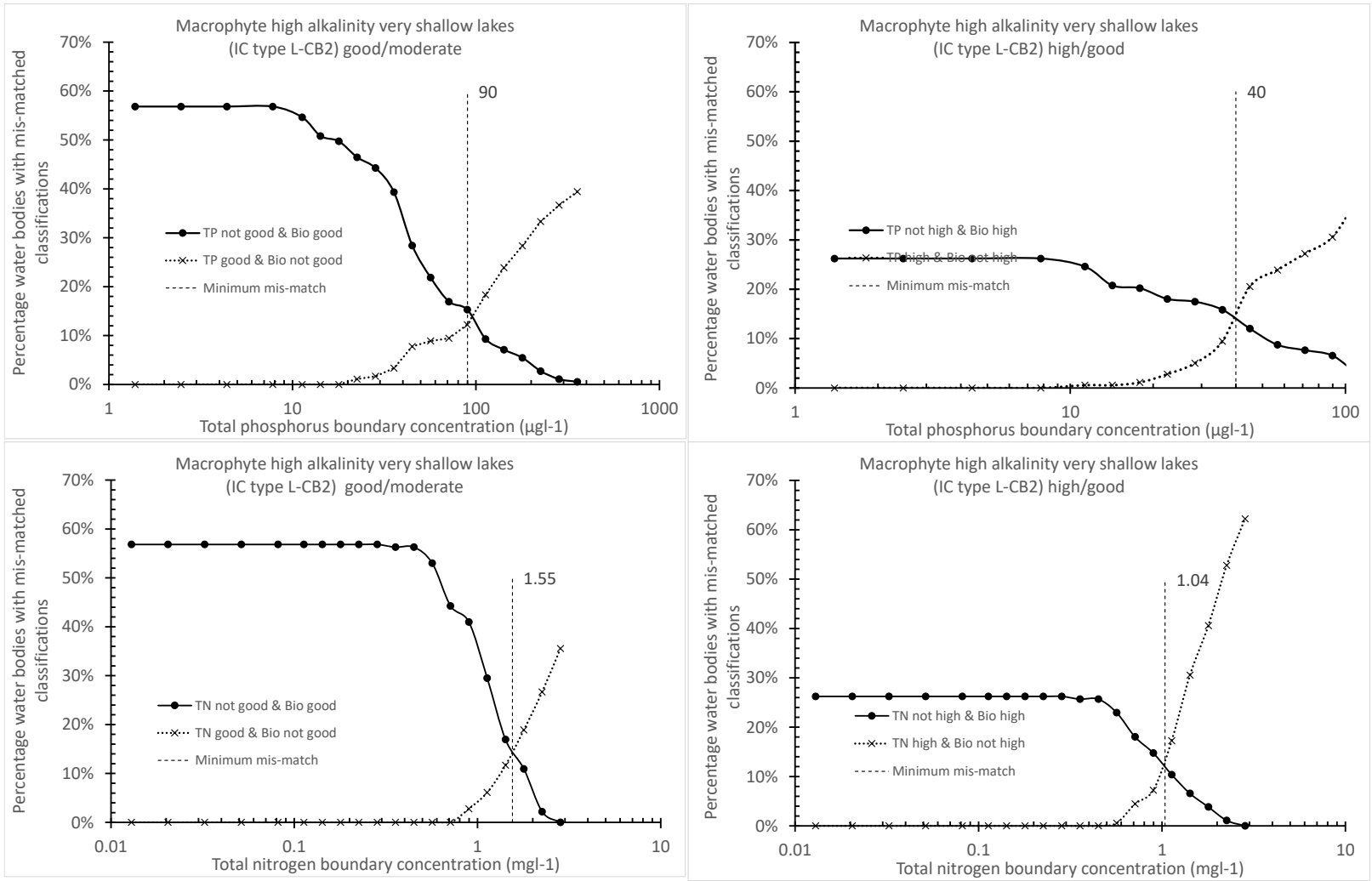


Figure 6-12 Percentage of water bodies where biology or total phosphorus / total nitrogen classifications for good ecological status differ in comparison to the level used to set a) the good/moderate and b) the high/good boundaries. Biological status assessed using the common metric for macrophyte in high alkalinity very shallow CBGIG lakes (Type L-CB2). Vertical lines mark intersection of curves where mismatch is minimised and equal.

Table 6-10 Regression parameters for L-CB2 lake macrophyte v total phosphorus and total nitrogen

```
Call:
lm(formula = CM.EQR ~ log10(total.P) + log10(total.N), data = data.cc.ex,
    subset = total.P > P.minUsed & total.P <= P.maxUsed & total.N >
        N.minUsed & total.N <= N.maxUsed)

Residuals:
     Min       1Q   Median       3Q      Max
-0.27593 -0.08393  0.01441  0.08355  0.27336

Coefficients:
              Estimate Std. Error t value Pr(>|t|)
(Intercept)    0.87468    0.04472  19.560  < 2e-16 ***
log10(total.P) -0.15424    0.02549  -6.051 8.00e-09 ***
log10(total.N) -0.23039    0.04534  -5.082 9.23e-07 ***
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 0.1224 on 182 degrees of freedom
Multiple R-squared:  0.4646,    Adjusted R-squared:  0.4587
F-statistic: 78.96 on 2 and 182 DF,  p-value: < 2.2e-16
```

Table 6-11 Regression parameters for L-CB2 lake macrophyte v total phosphorus

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x =
"interval", nperm = 99)

n = 224   r = -0.5862793   r-square = 0.3437234
Parametric P-values:   2-tailed = 4.499791e-22    1-tailed = 2.249896e-22
Angle between the two OLS regression lines = 21.34054 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS 0.9930275 -0.2384879       -13.41379              0.01
2     MA 1.0428369 -0.2656139       -14.87509              0.01
3    SMA 1.3020523 -0.4067820       -22.13561                NA
4    RMA 1.1925330 -0.3471381       -19.14384              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS      0.9112725        1.074782 -0.2820743  -0.1949015
2     MA      0.9546707        1.133155 -0.3148010  -0.2175989
3    SMA      1.2262937        1.386362 -0.4526969  -0.3655241
4    RMA      1.0819690        1.316577 -0.4146923  -0.2869252

Eigenvalues: 0.1565292 0.01503339

H statistic used for computing C.I. of MA: 0.002056158
```

Table 6-12 Regression parameters for L-CB2 lake macrophyte v total nitrogen

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x =
"interval", nperm = 99)

n = 196   r = -0.5848763   r-square = 0.3420803
Parametric P-values:   2-tailed = 2.233696e-19    1-tailed = 1.116848e-19
Angle between the two OLS regression lines = 27.48763 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS 0.6123594 -0.3921105       -21.41067              0.01
2     MA 0.6355612 -0.5197933       -27.46511              0.01
3    SMA 0.6629316 -0.6704162       -33.83854                NA
4    RMA 0.6625300 -0.6682061       -33.75109              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS      0.5887414       0.6359774 -0.4691115  -0.3151096
2     MA      0.6177011       0.6549838 -0.6266785  -0.4215064
3    SMA      0.6497403       0.6777247 -0.7518246  -0.5978228
4    RMA      0.6407059       0.6890900 -0.8143689  -0.5481051

Eigenvalues: 0.07388125 0.01507563

H statistic used for computing C.I. of MA: 0.006458061
```

6.1.5 Phytobenthos XGIG high alkalinity lakes

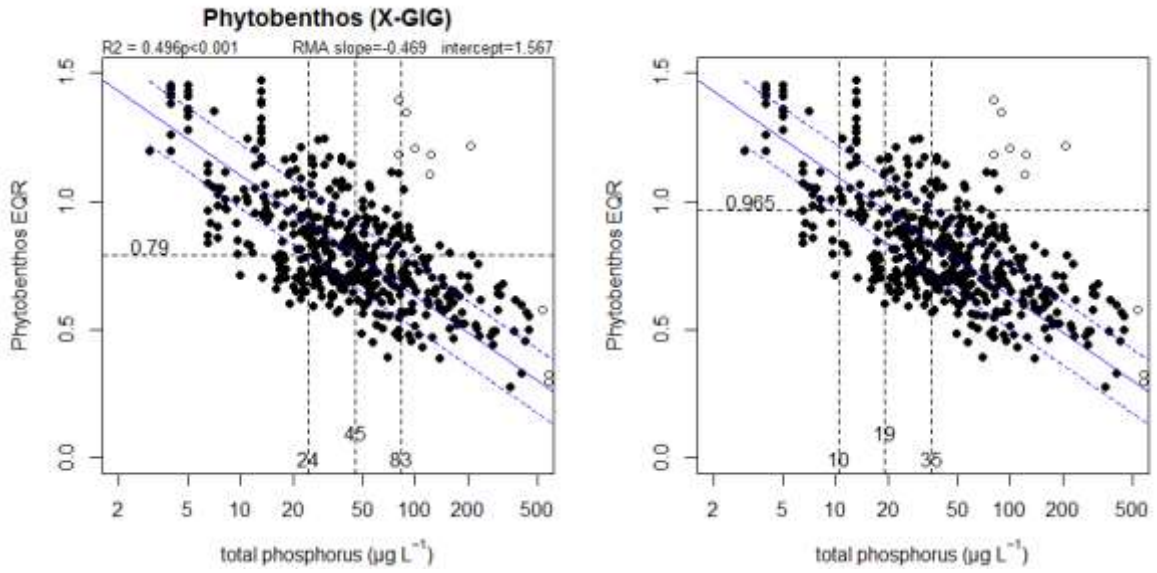


Figure 6-13 Relationship between common metric for phytobenthos and total phosphorus for high alkalinity XGIG lakes showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data, open circles data points excluded from regression.

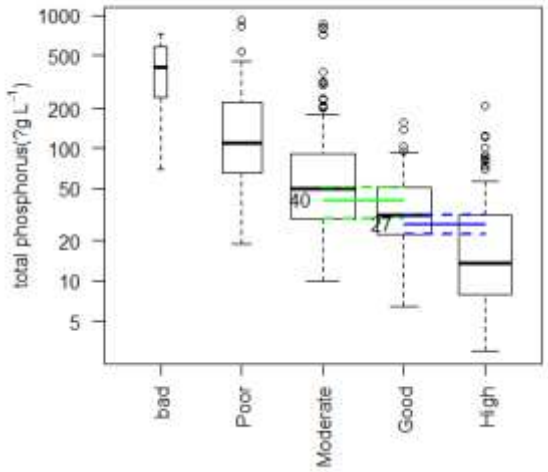


Figure 6-14 Box plots showing range of a)TP for high alkalinity XGIG lakes classified using phytobenthos common metric showing good/moderate boundary & high/good boundary values determined from the average of the upper and lower quartile values of adjacent classes

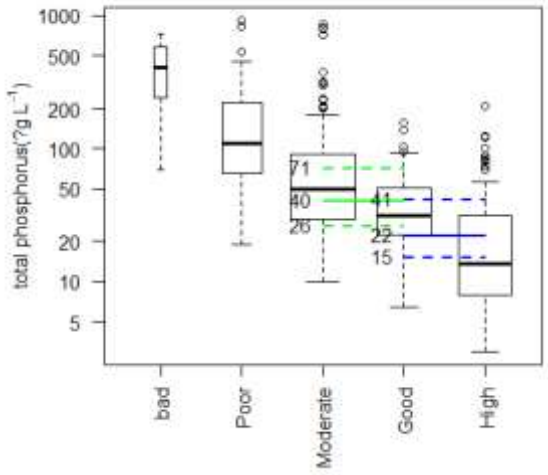


Figure 6-15 Box plots showing range of a)TP for high alkalinity XGIG lakes classified using phytobenthos common metric showing good/moderate boundary & high/good boundary values determined from the average of adjacent classes

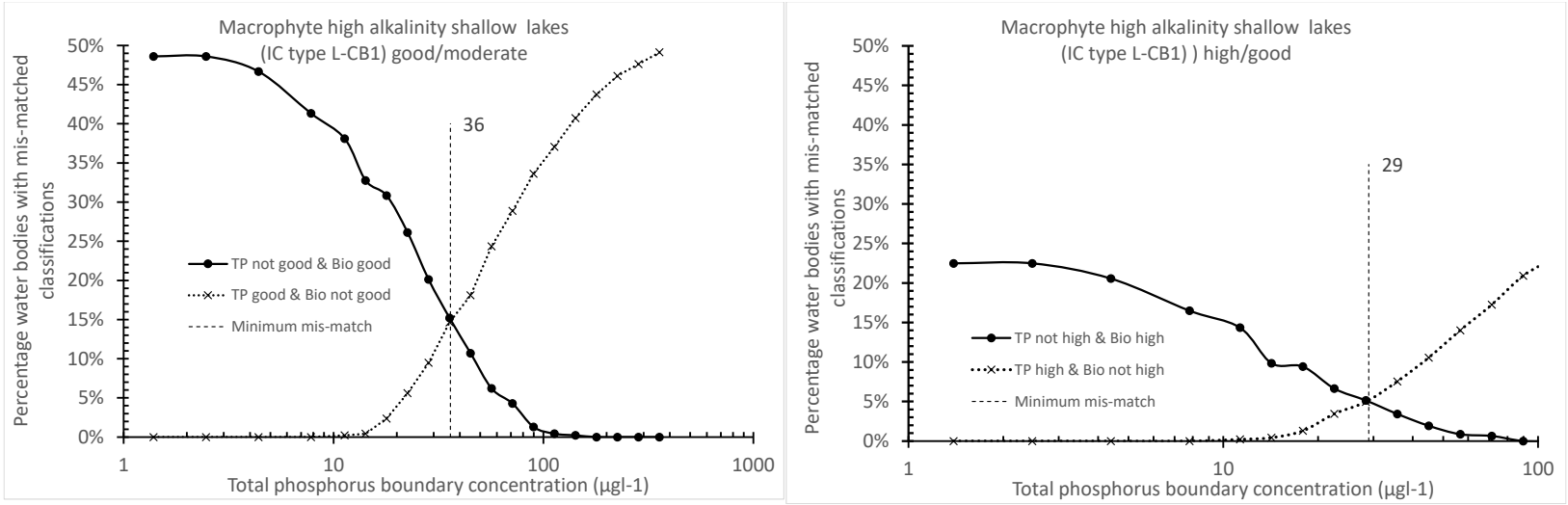


Figure 6-16 Percentage of water bodies where biology or total phosphorus classifications for good ecological status differ in comparison to the level used to set a) the good/moderate and b) the high/good boundaries. Biological status assessed using the common metric for phytobenthos in high alkalinity XGIG lakes. Vertical lines mark intersection of curves where mismatch is minimised and equal.

Table 6-13 Regression parameters for XGIG high alkalinity lake phytobenthos v total phosphorus

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x =
"interval", nperm = 99)

n = 463   r = -0.7041176   r-square = 0.4957816
Parametric P-values:   2-tailed = 1.495768e-70    1-tailed = 7.478838e-71
Angle between the two OLS regression lines = 16.35559 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS  1.404841 -0.3669257       -20.14938              0.01
2     MA  1.483213 -0.4163830       -22.60601              0.01
3    SMA  1.649173 -0.5211142       -27.52466                NA
4    RMA  1.566809 -0.4691372       -25.13302              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS       1.349181        1.460501 -0.4007931  -0.3330583
2     MA       1.423099        1.544990 -0.4553679  -0.3784477
3    SMA       1.597248        1.704583 -0.5560809  -0.4883462
4    RMA       1.500132        1.637619 -0.5138225  -0.4270599

Eigenvalues: 0.2194011 0.02260629

H statistic used for computing C.I. of MA: 0.001072801
```

6.1.6 Invertebrates L-CBGIG all lake types

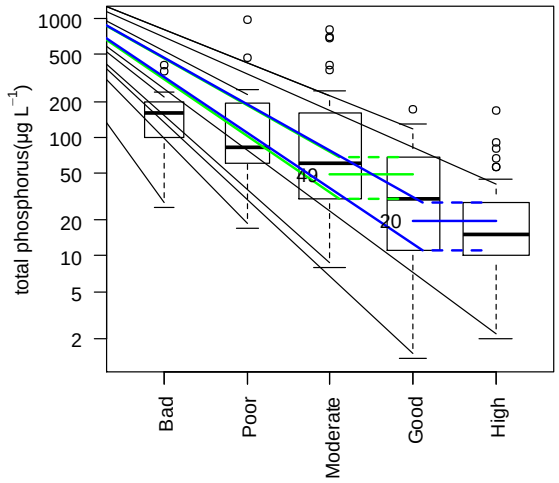


Figure 6-17 Box plots showing range of a)TP for CBGIG lakes (All types) classified using invertebrate common metric showing good/moderate boundary & high/good boundary values determined from the average of the upper and lower quartile values of adjacent classes

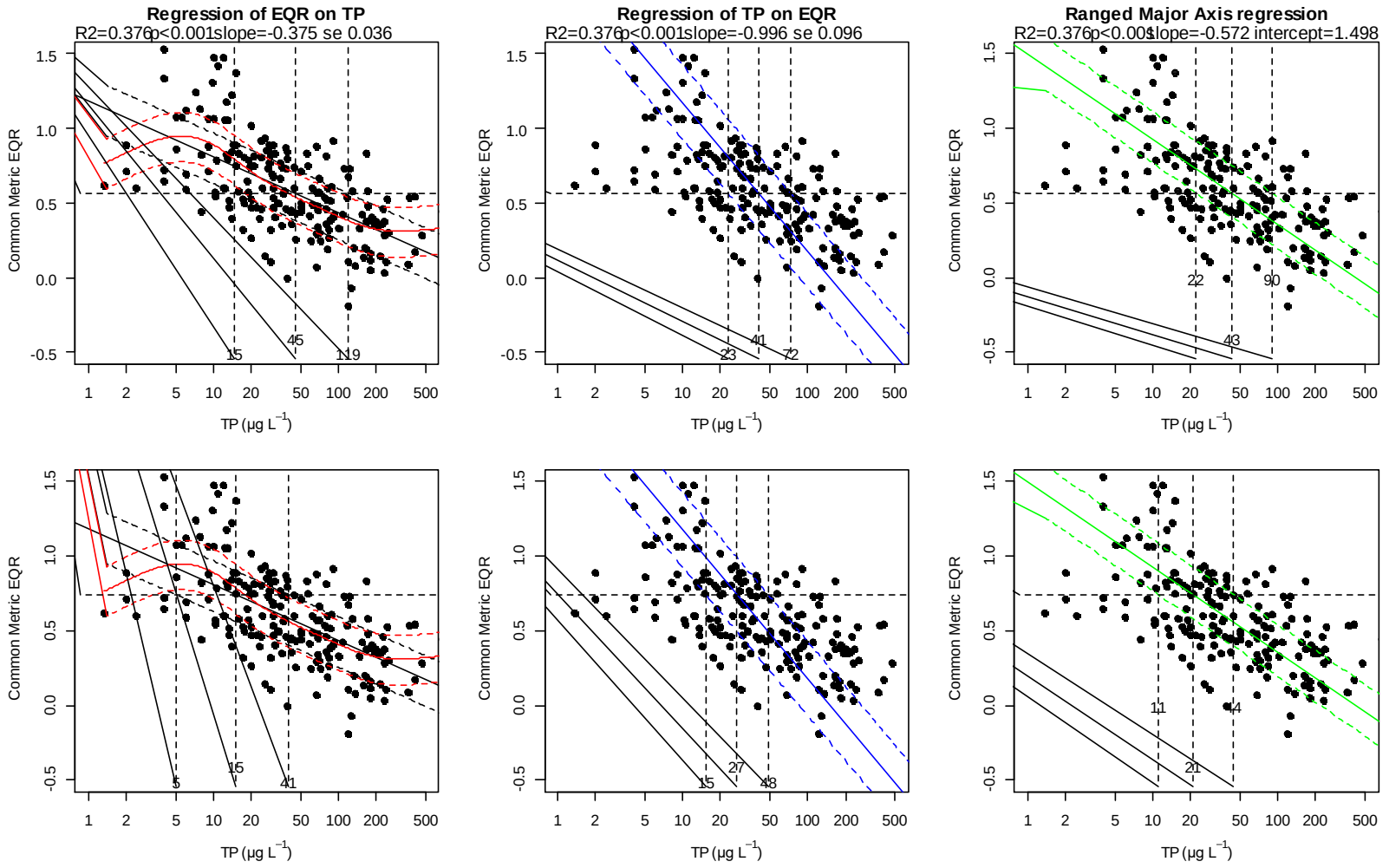


Figure 6-18 Relationship between common metric for invertebrates and total phosphorus for CBGIG lakes (All types) showing a) good/moderate boundary and b) high/good boundary values. Lines show regression, dotted lines show area containing 50% of the data, open circles data points excluded from regression.

6.2 Low and moderate alkalinity clear water lakes Northern GIG

6.2.1 Phytoplankton IC Type L-N2a low alkalinity shallow

Table 6-14. Predicted total phosphorus boundary values for low alkalinity shallow lakes using regression models and categorical methods

IC Type	Phytoplankton Models	R^2	nutrient range TP μgl^{-1}	GM TP μgl^{-1}			HG TP μgl^{-1}		
				Pred	25th	75th	Pred	25th	75th
L-N2a	EQR v TP + TN (OLS)	0.37	2 - 47	20	15	27	9	7	12
	EQR v TP (OLS)	0.35	2 - 47	22	16	31	10	7	13
	TP v EQR (OLS)			11	9	13	8	6	10
	EQR v TP (RMA)			18	13	24	9	7	12
	Average adjacent quartiles			11			8		
	Average adjacent classes			11	13	9	8	7	10
	Minimise class difference			14			10		

IC Type	Phytoplankton Models	R^2	nutrient range TN mgl^{-1}	GM TN mgl^{-1}			HG TN mgl^{-1}		
L-N2a	EQR v TP + TN (OLS)	0.37	0.11 - 1.00	1.30	0.89	1.80	0.47	0.32	0.70
	EQR v TN (OLS)			2.15	1.04	4.26	0.53	0.26	1.05
	TN v EQR (OLS)	0.10	0.11 - 1.12	0.41	0.33	0.52	0.36	0.29	0.45
	EQR v TN (RMA)			1.46	0.83	2.48	0.49	0.28	0.83
	Average adjacent quartiles			0.41			0.35		
	Average adjacent classes			0.41	0.32	0.56	0.36	0.28	0.44
	Minimise class difference			0.65			0.41		

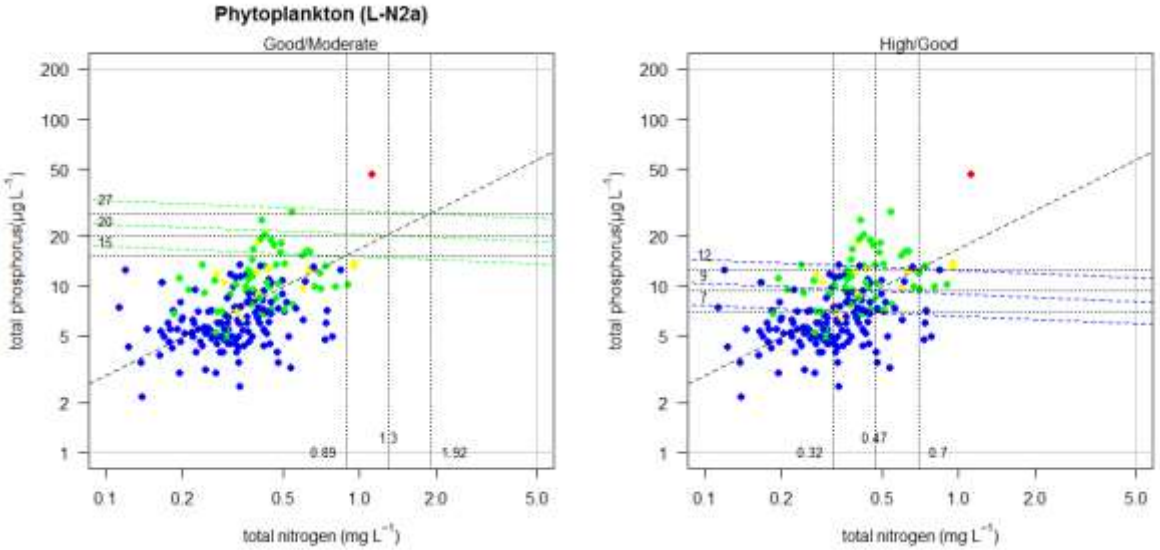


Figure 6-19 Relationship between mean TP and TN, points coloured by WFD class for phytoplankton in low alkalinity shallow NGIG lakes (Type L-N2a). Dotted lines contours of predicted TN & TP concentration when phytoplankton common metric EQR is at a) good/moderate boundary (green lines) and b) high good boundary, dotted lines show ± 25th & 75th residuals of prediction. Horizontal & vertical lines show intersection with RMA regression of observed TP and TN showing boundary values.

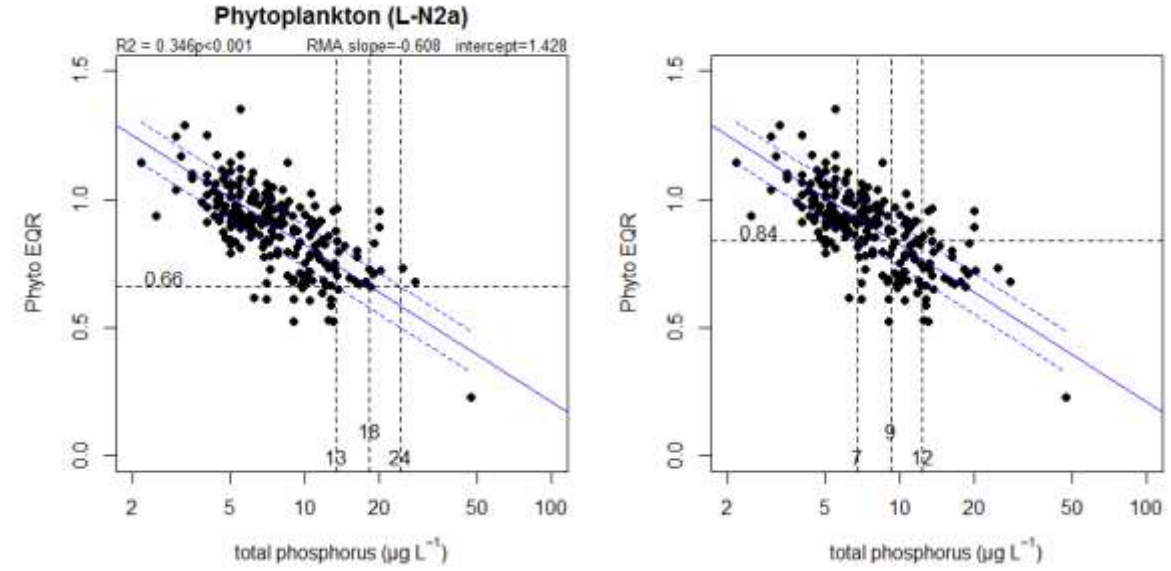


Figure 6-20 Relationship between common metric for phytoplankton and total phosphorus for low alkalinity shallow NGIG lakes (Type L-N2a) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data, open circles data points excluded from regression.

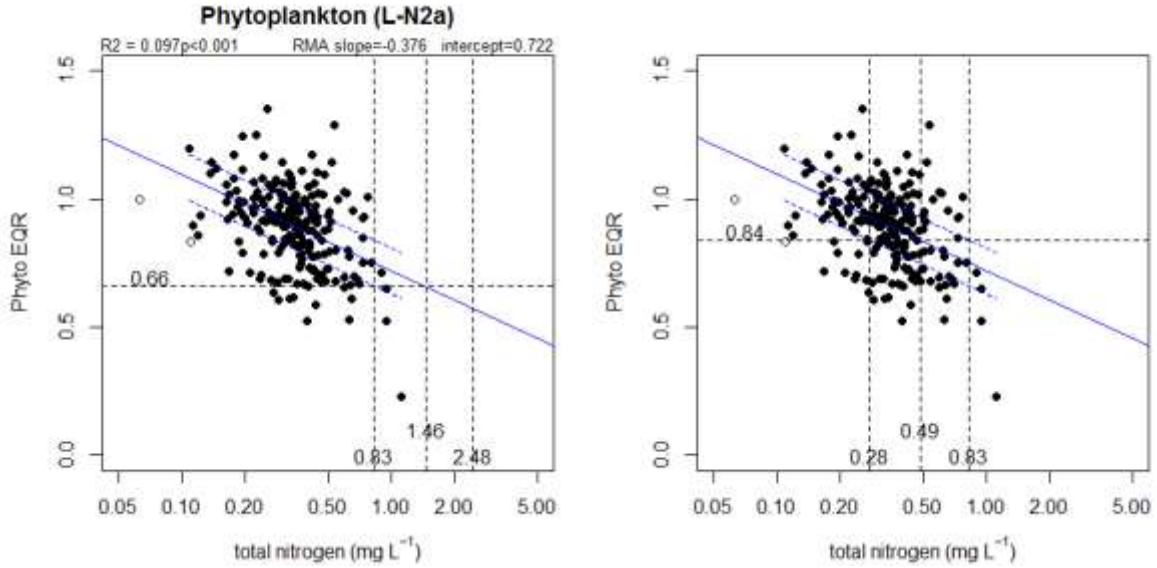


Figure 6-21 Relationship between common metric for phytoplankton and total nitrogen for low alkalinity shallow NGIG lakes (Type L-N2a) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data, open circles data points excluded from regression.

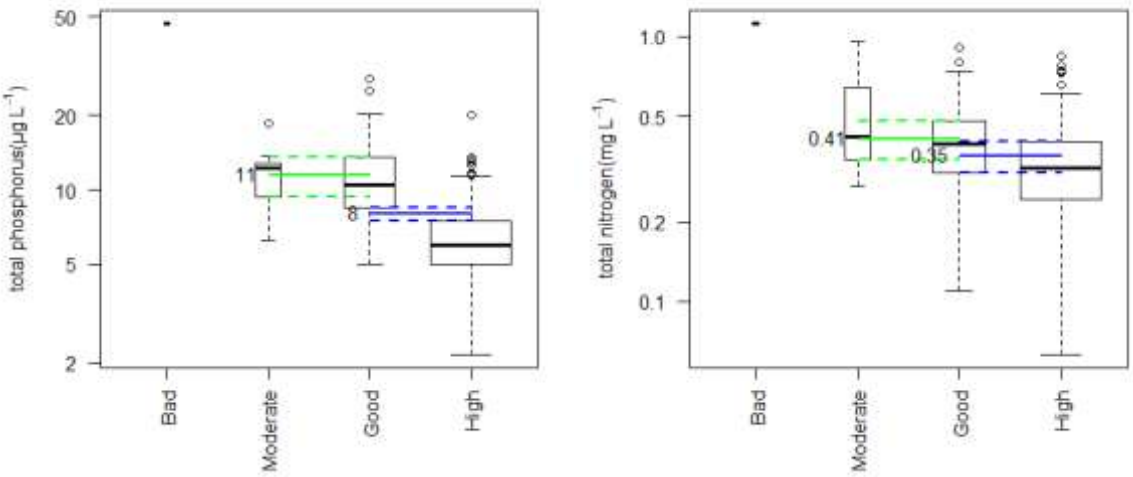


Figure 6-22 Box plots showing range of a) TP and b) TN for low alkalinity shallow NGIG lakes (Type L-N2a) classified using phytoplankton common metric showing good/moderate boundary & high/good boundary values determined from the average of the upper and lower quartile values

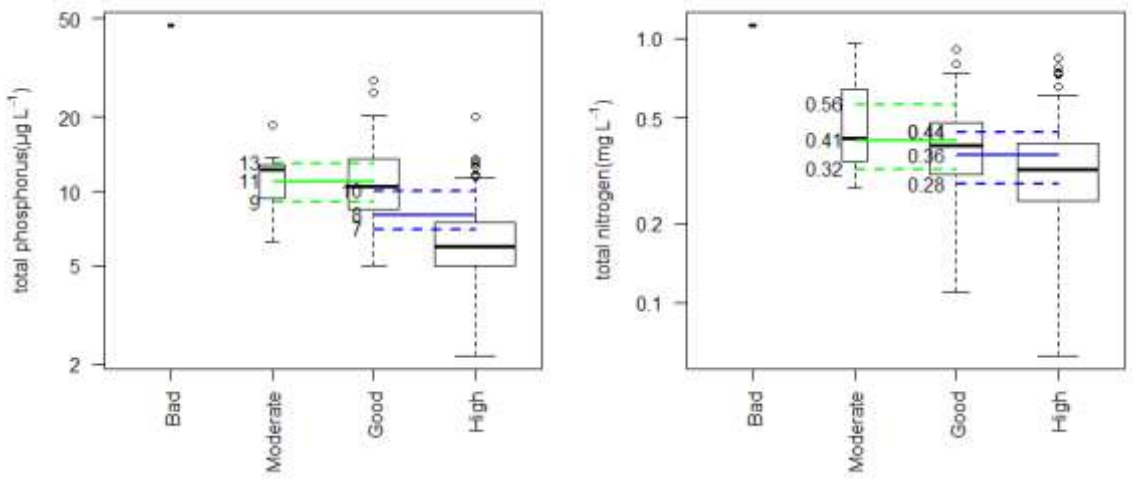


Figure 6-23 Box plots showing range of a) TP and b) TN for low alkalinity shallow NGIG lakes (Type L-N2a) classified using macrophyte common metric showing good/moderate boundary & high/good boundary values determined from the average of adjacent classes

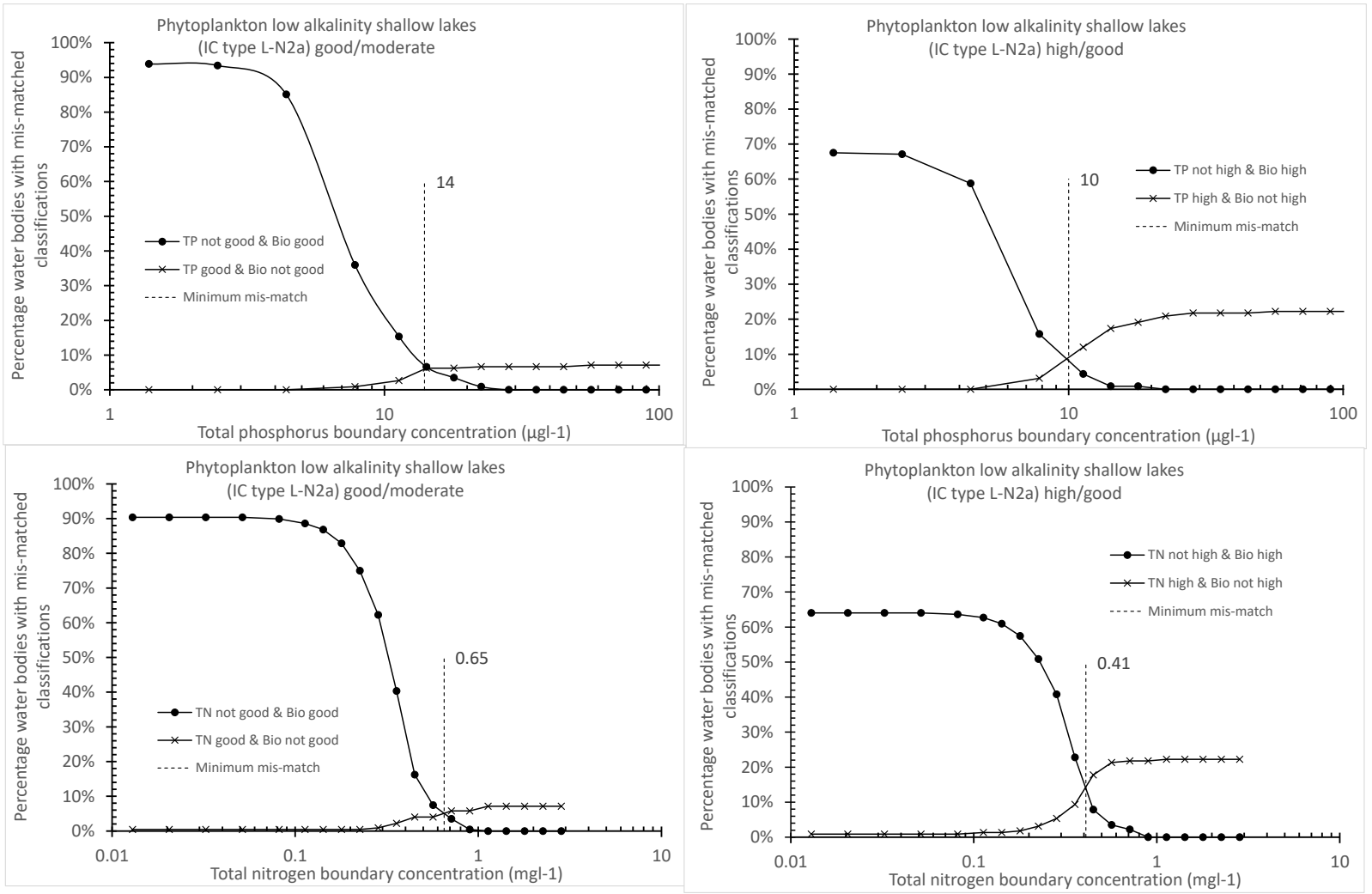


Figure 6-24 Percentage of water bodies where biology or total phosphorus / total nitrogen classifications for good ecological status differ in comparison to the level used to set a) the good/moderate and b) the high/good boundaries.

Biological status assessed using the common metric for phytoplankton in low alkalinity shallow NGIG lakes (Type L-N2a) Vertical lines mark intersection of curves where mis-match is minimised and equal.

Table 6-15 Regression parameters for L-N2a lake phytoplankton v total phosphorus and total nitrogen

```
Call:
lm(formula = CM.EQR ~ log10(total.P) + log10(total.N), data = data.cc.ex,
    subset = total.P > P.minUsed & total.P <= P.maxUsed & total.N >
        N.minUsed & total.N <= N.maxUsed)

Residuals:
     Min       1Q   Median       3Q      Max
-0.32905 -0.07067 -0.00580  0.06606  1.19536

Coefficients:
              Estimate Std. Error t value Pr(>|t|)
(Intercept)    1.31987    0.06267  21.060   <2e-16 ***
log10(total.P) -0.50413    0.05158  -9.774   <2e-16 ***
log10(total.N) -0.03176    0.05876  -0.541    0.589
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 0.1369 on 213 degrees of freedom
Multiple R-squared:  0.3726,    Adjusted R-squared:  0.3667
F-statistic: 63.24 on 2 and 213 DF,  p-value: < 2.2e-16
```

Table 6-16 Regression parameters for L-N2a lake phytoplankton v total phosphorus

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x =
"interval", nperm = 99)

n = 230   r = -0.5879814   r-square = 0.3457221
Parametric P-values:   2-tailed = 8.841474e-23    1-tailed = 4.420737e-23
Angle between the two OLS regression lines = 28.72588 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS  1.328105 -0.4941434       -26.29598              0.01
2     MA  1.548110 -0.7460310       -36.72408              0.01
3    SMA  1.630541 -0.8404066       -40.04391                NA
4    RMA  1.427739 -0.6082161       -31.30864              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS       1.248601        1.407609 -0.5828514  -0.4054355
2     MA       1.437806        1.673606 -0.8897131  -0.6197425
3    SMA       1.557138        1.712098 -0.9337833  -0.7563674
4    RMA       1.334626        1.525875 -0.7205737  -0.5016102

Eigenvalues: 0.05549768 0.01369095

H statistic used for computing C.I. of MA: 0.007402868
```

Table 6-17 Regression parameters for L-N2a lake phytoplankton v total nitrogen

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x =
"interval", nperm = 99)

n = 219   r = -0.3119072   r-square = 0.09728611
Parametric P-values:   2-tailed = 2.509522e-06    1-tailed = 1.254761e-06
Angle between the two OLS regression lines = 55.32204 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS 0.7589401 -0.2970722       -16.54522              0.01
2     MA 0.4993077 -0.8558449       -40.55840              0.01
3    SMA 0.4544260 -0.9524378       -43.60453                NA
4    RMA 0.7221380 -0.3762765       -20.62014              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS      0.6985661       0.8193141 -0.4181484  -0.1759960
2     MA      0.2985231       0.6400177 -1.2879671  -0.5530133
3    SMA      0.3946067       0.5071223 -1.0811790  -0.8390265
4    RMA      0.6496032       0.7925623 -0.5323835  -0.2247116

Eigenvalues: 0.04126414 0.02147977

H statistic used for computing C.I. of MA: 0.04053704
```

6.2.2 Phytoplankton IC Type L-N2b low alkalinity deep

Table 6-18 Predicted total phosphorus boundary values for low alkalinity deep lakes using regression models and categorical methods

IC Type	Phytoplankton Models	R2	nutrient range TP µgl^{-1}	GM TP µgl^{-1}			HG TP µgl^{-1}		
				Pred	25th	75th	Pred	25th	75th
L-N2b	EQR v TP + TN (OLS)	0.37	2 - 19	14	11	19	8	6	10
	EQR v TP (OLS)			15	11	20	8	6	10
	TP v EQR (OLS)	0.37	2 - 19	8	7	10	7	5	8
	EQR v TP (RMA)			13	10	17	7	6	10
	Average adjacent quartiles			9			6		
	Average adjacent classes			9	11	7	7	5	8
	Minimise class difference			11			7		

IC Type	Macrophyte Models	R2	nutrient range TN mgl^{-1}			GM TN mgl^{-1}			HG TN mgl^{-1}		
						Pred	25th	75th	Pred	25th	75th
L-N2b	EQR v TP + TN (OLS)	0.37	0.1	-	1.00	**3.50**	1.70	8.50	**0.64**	0.32	1.55
	EQR v TN (OLS)					**0.70**	**0.55**	**0.82**	**0.52**	**0.42**	**0.62**
	TN v EQR (OLS)	0.26	0.4	-	0.73	**0.56**	**0.51**	**0.63**	**0.52**	**0.47**	**0.59**
	EQR v TN (RMA)					**0.66**	**0.55**	**0.76**	**0.52**	**0.44**	**0.60**
	Average adjacent quartiles					0.39			0.29		
	Average adjacent classes					0.39	0.28	0.53	0.32	0.18	0.40
	Minimise class difference					0.55			0.39		

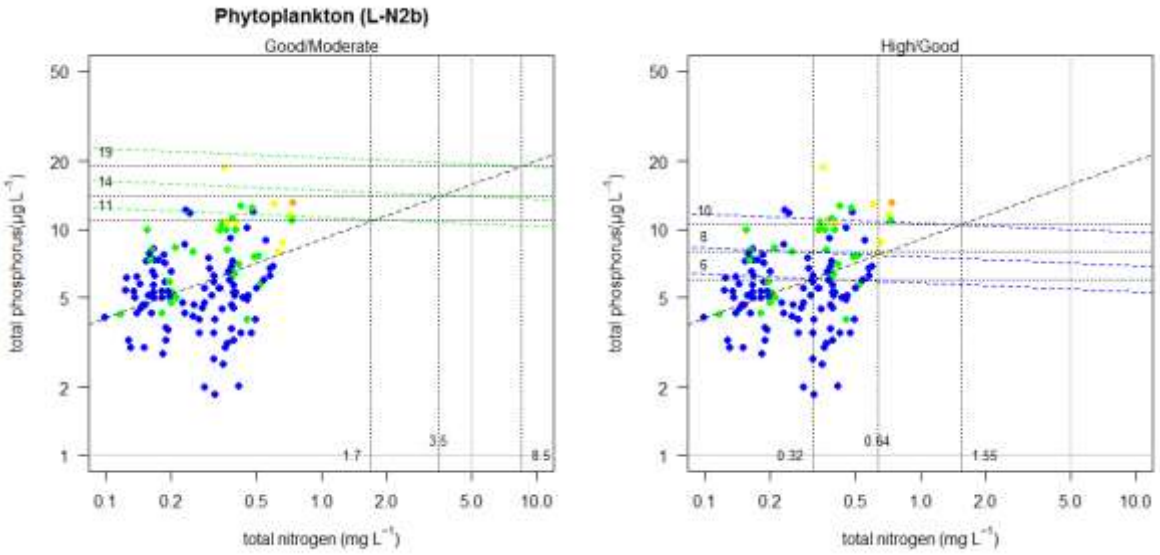


Figure 6-25Relationship between mean TP and TN, points coloured by WFD class for phytoplankton in low alkalinity deep NGIG lakes (Type L-N2b). Dotted lines contours of predicted TN & TP concentration when phytoplankton common metric EQR is at a) good/moderate boundary (green lines) and b) high good boundary, dotted lines show ± 25th & 75th residuals of prediction. Horizontal & vertical lines show intersection with RMA regression of observed TP and TN showing boundary values.

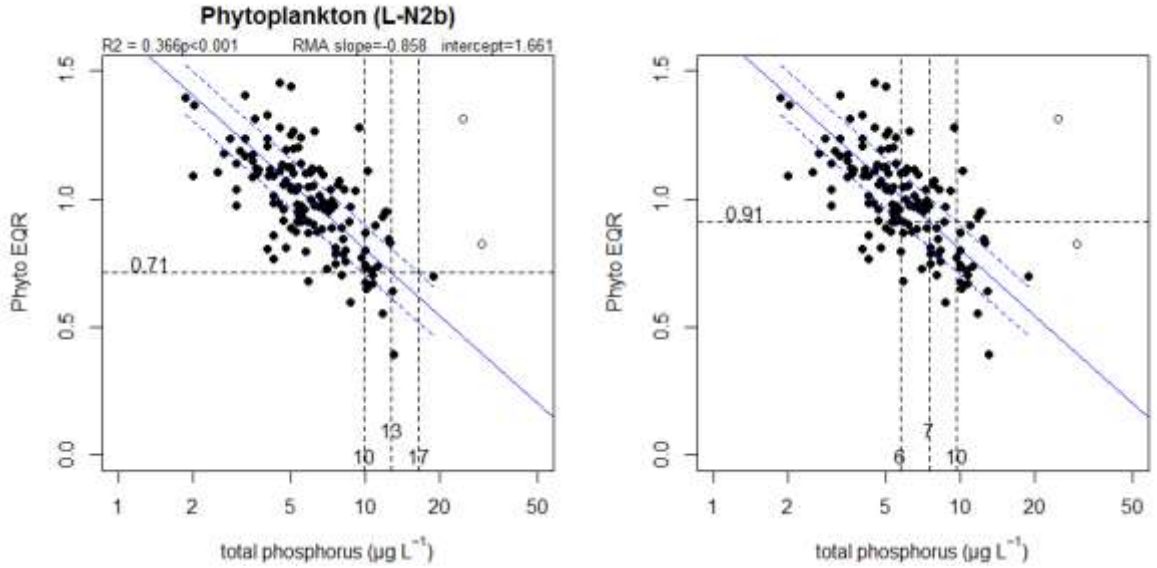


Figure 6-26 Relationship between common metric for phytoplankton and total phosphorus for low alkalinity deep NGIG lakes (Type L-N2b) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data, open circles data points excluded from regression.

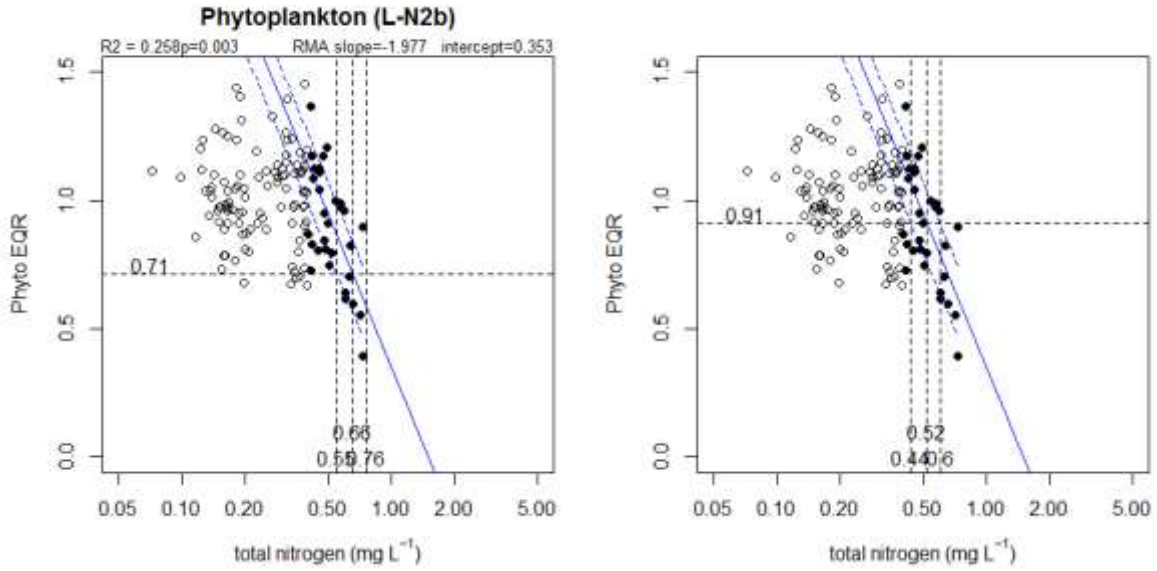


Figure 6-27 Relationship between common metric for phytoplankton and total nitrogen for low alkalinity deep NGIG lakes (Type L-N2b) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data, open circles data points excluded from regression.

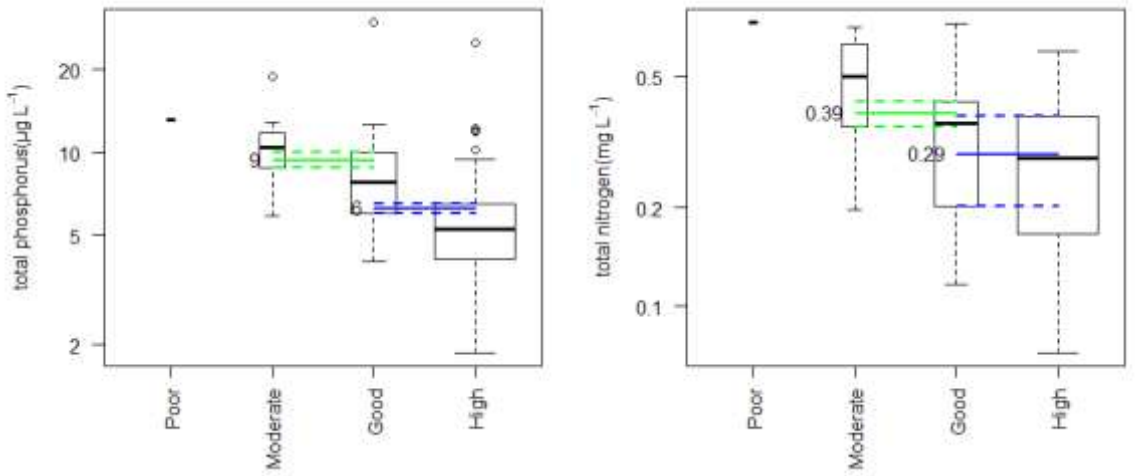


Figure 6-28 Box plots showing range of a) TP and b) TN for low alkalinity deep NGIG lakes (Type L-N2b) classified using phytoplankton common metric showing good/moderate boundary & high/good boundary values determined from the average of the upper and lower quartile values

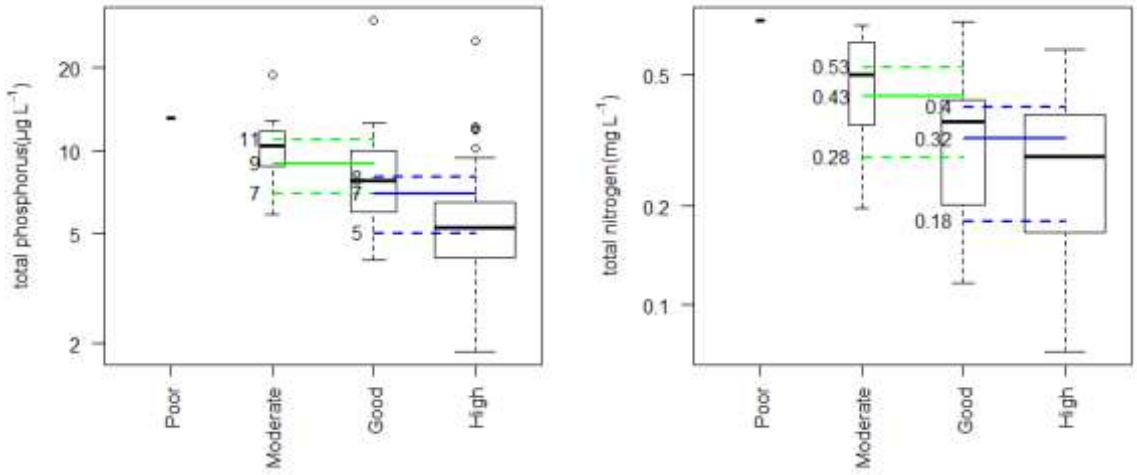


Figure 6-29 Box plots showing range of a) TP and b) TN for low alkalinity deep NGIG lakes (Type L-N2b) classified using macrophyte common metric showing good/moderate boundary & high/good boundary values determined from the average of adjacent classes

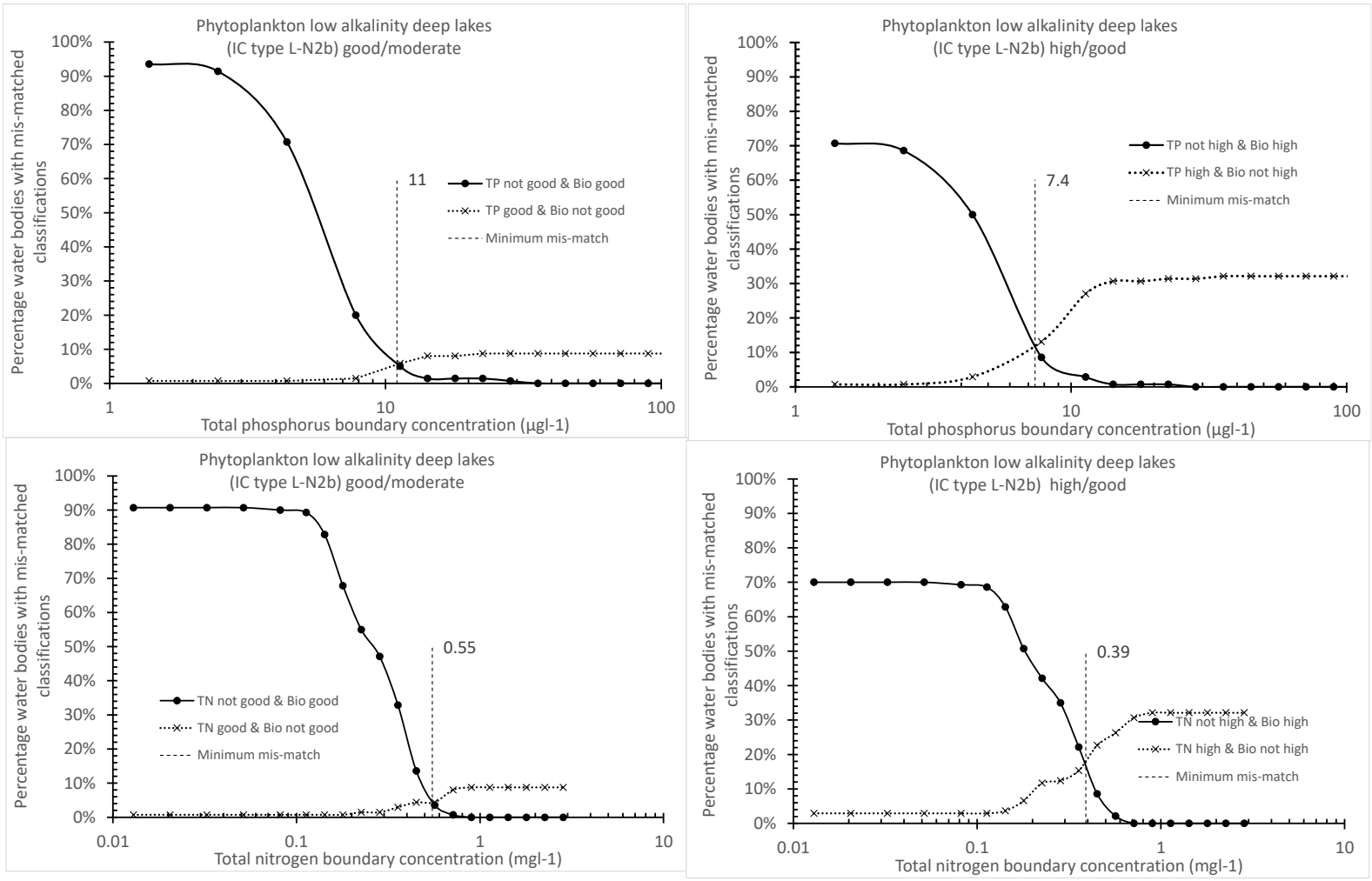


Figure 6-30 Percentage of water bodies where biology or total phosphorus / total nitrogen classifications for good ecological status differ in comparison to the level used to set a) the good/moderate and b) the high/good boundaries. Biological status assessed using the common metric for phytoplankton in low alkalinity deep NGIG lakes (Type L-N2b). Vertical lines mark intersection of curves where mis-match is minimised and equal.

Table 6-19 Regression parameters for L-N2b lake phytoplankton v total phosphorus and total nitrogen

```
Call: lm(formula = CM.EQR ~ log10(total.P), data = data.cc.ex, subset = total.P >
    P.minUsed & total.P <= P.maxUsed & total.N > N.minUsed &
    total.N <= N.maxUsed)

Residuals:
     Min       1Q   Median       3Q      Max
-0.36381 -0.10229 -0.00233  0.07648  0.75270

Coefficients:
              Estimate Std. Error t value Pr(>|t|)
(Intercept)    1.53518    0.06212  24.711  < 2e-16 ***
log10(total.P) -0.69412    0.07905  -8.781 8.09e-15 ***
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 0.1682 on 130 degrees of freedom
Multiple R-squared:  0.3723,    Adjusted R-squared:  0.3675
F-statistic:  77.1 on 1 and 130 DF,  p-value: 8.094e-15
```

Table 6-20 Regression parameters for L-N2b lake phytoplankton v total phosphorus

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x =
"interval", nperm = 99)

n = 140   r = -0.604958   r-square = 0.3659742
Parametric P-values:   2-tailed = 2.452718e-15    1-tailed = 1.226359e-15
Angle between the two OLS regression lines = 27.45276 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS  1.531266 -0.6900465       -34.60748              0.01
2     MA  1.955922 -1.2416854       -51.15353              0.01
3    SMA  1.878146 -1.1406520       -48.75923                NA
4    RMA  1.660595 -0.8580477       -40.63117              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS       1.410241        1.652291 -0.8429229  -0.5371701
2     MA       1.768271        2.202236 -1.5616524  -0.9979221
3    SMA       1.768312        2.003683 -1.3037274  -0.9979746
4    RMA       1.519066        1.813171 -1.0562475  -0.6741990

Eigenvalues: 0.06378869 0.01526219

H statistic used for computing C.I. of MA: 0.01171306
```

Table 6-21 Regression parameters for L-N2b lake phytoplankton v total nitrogen

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x =
"interval", nperm = 99)

n = 32   r = -0.5075676   r-square = 0.2576249
Parametric P-values:   2-tailed = 0.00302385    1-tailed = 0.001511925
Angle between the two OLS regression lines = 22.81232 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method   Intercept     Slope Angle (degrees) P-perm (1-tailed)
1    OLS  0.45956678 -1.604405       -58.06537              0.01
2     MA -0.73380860 -5.777479       -80.18020              0.01
3    SMA  0.01443606 -3.160968       -72.44477                NA
4    RMA  0.35307459 -1.976794       -63.16654              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS      0.1588584       0.7602751  -2.619916  -0.5888935
2     MA     -3.4873504      -0.0838140 -15.406247  -3.5045349
3    SMA     -0.3214735       0.2593387  -4.335599  -2.3045759
4    RMA     -0.0356108       0.6958408  -3.335975  -0.7781862

Eigenvalues: 0.06259905 0.004402909

H statistic used for computing C.I. of MA: 0.01131424
```

6.2.3 Phytoplankton IC Type L-N1 moderate alkalinity shallow

Table 6-22 Predicted total phosphorus boundary values for moderate alkalinity shallow lakes using regression models and categorical methods

IC Type	Phytoplankton Models	R^2	nutrient range TP µgl^{-1}	GM TP µgl^{-1}			HG TP µgl^{-1}		
				Pred	25th	75th	Pred	25th	75th
L-N1	EQR v TP + TN (OLS)	0.81	2 - 100	18	15	22	11	9	13
	EQR v TP (OLS)	0.79	2 - 100	18	15	23	11	9	13
	TP v EQR (OLS)			18	15	21	12	10	14
	EQR v TP (RMA)			18	15	22	11	9	14
	Average adjacent quartiles			19			11		
	Average adjacent classes			19	16	23	12	9	15
	Minimise class difference			20			11		

IC Type	Phytoplankton Models	R^2	nutrient range TN mgl^{-1}	GM TN mgl^{-1}			HG TN mgl^{-1}		
				Pred	25th	75th	Pred	25th	75th
L-N1	EQR v TP + TN (OLS)	0.81	0.09 - 4.00	0.65	0.52	0.79	0.36	0.28	0.44
	EQR v TN (OLS)	0.53	0.09 - 4.44	0.70	0.53	1.04	0.35	0.26	0.52
	TN v EQR (OLS)			0.59	0.45	0.74	0.41	0.31	0.52
	EQR v TN (RMA)			0.63	0.49	0.86	0.38	0.29	0.51
	Average adjacent quartiles			0.52			0.39		
	Average adjacent classes			0.52	0.43	0.91	0.38	0.31	0.47
	Minimise class difference			0.54			0.33		

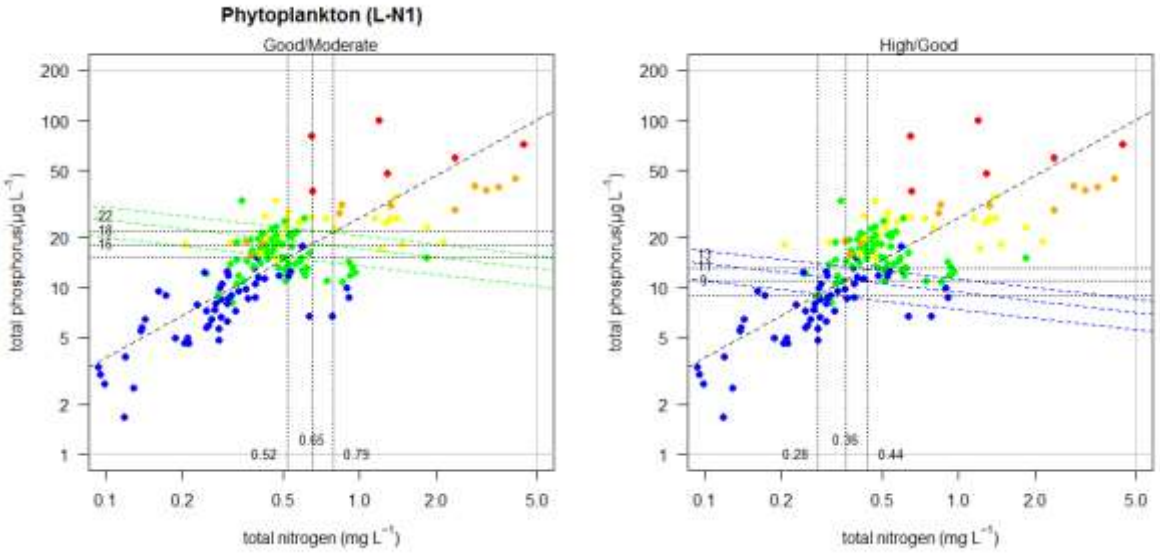


Figure 6-31 Relationship between mean TP and TN, points coloured by WFD class for phytoplankton in moderate alkalinity shallow CBGIG lakes (Type L-N1). Dotted lines contours of predicted TN & TP concentration when phytoplankton common metric EQR is at a) good/moderate boundary (green lines) and b) high/good boundary, dotted lines show ± 25th & 75th residuals of prediction. Horizontal & vertical lines show intersection with RMA regression of observed TP and TN showing boundary values.

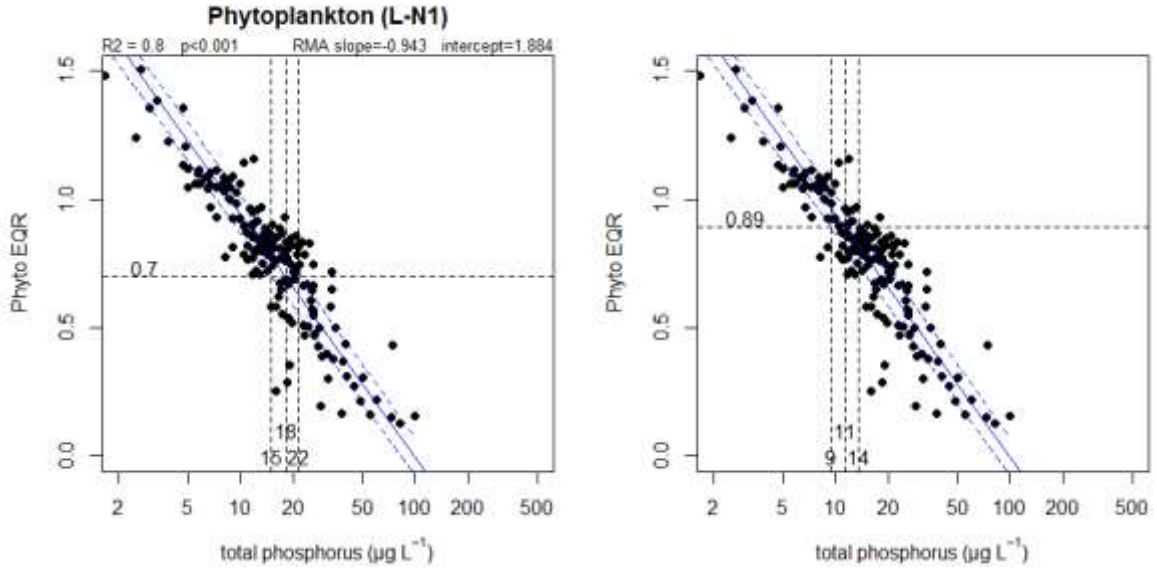


Figure 6-32 Relationship between common metric for phytoplankton and total phosphorus for moderate alkalinity shallow lakes (Type L-N1) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data, open circles data points excluded from regression.

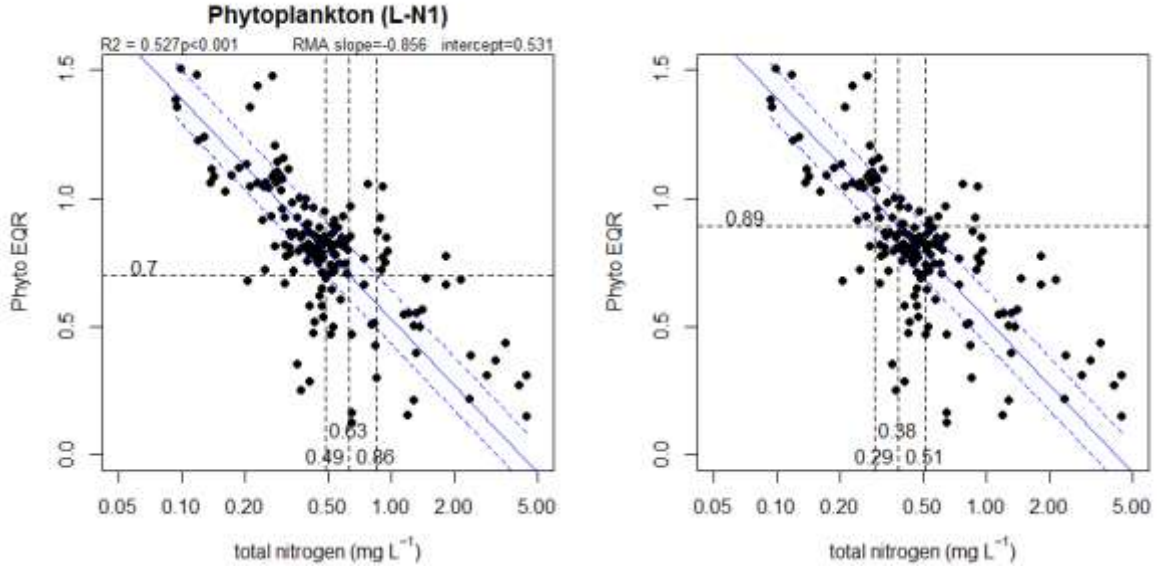


Figure 6-33 Relationship between common metric for phytoplankton and total nitrogen for moderate alkalinity shallow lakes (Type L-N1) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data, open circles data points excluded from regression.

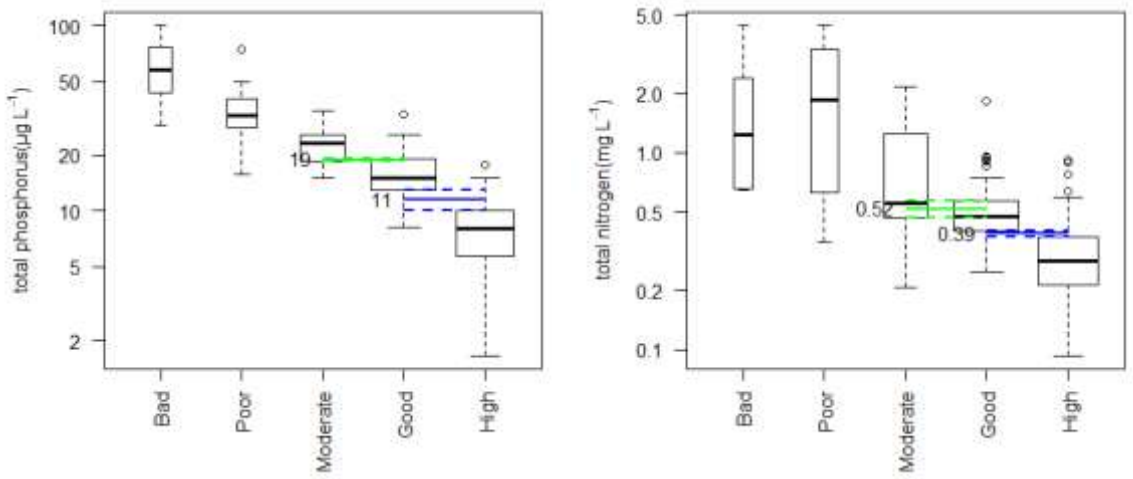


Figure 6-34 Box plots showing range of a) TP and b) TN for moderate alkalinity shallow lakes (Type L-N1) classified using phytoplankton common metric showing good/moderate boundary & high/good boundary values determined from the average of the upper and lower quartile values

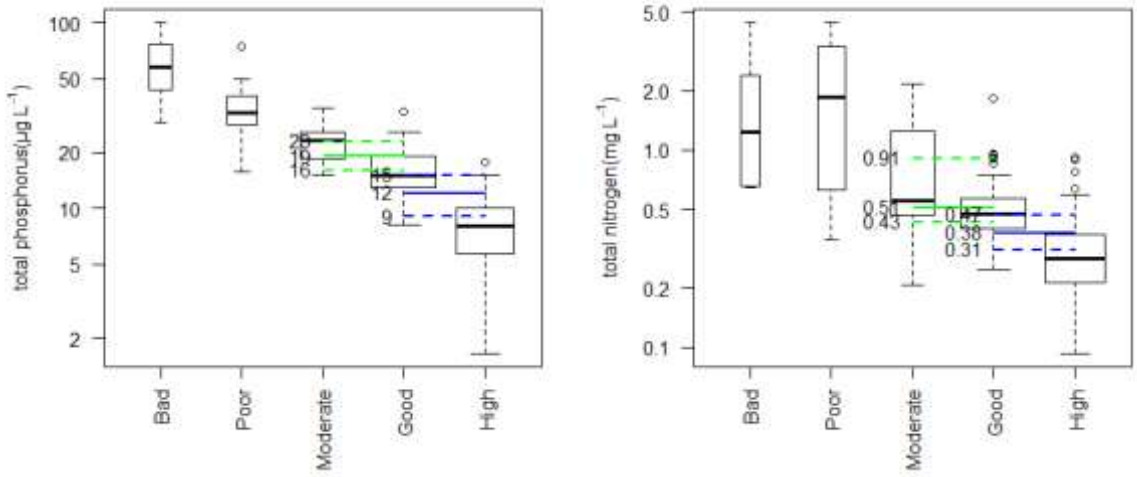


Figure 6-35 Box plots showing range of a) TP and b) TN for moderate alkalinity shallow lakes (Type L-N1) classified using macrophyte common metric showing good/moderate boundary & high/good boundary values determined from the average of adjacent classes

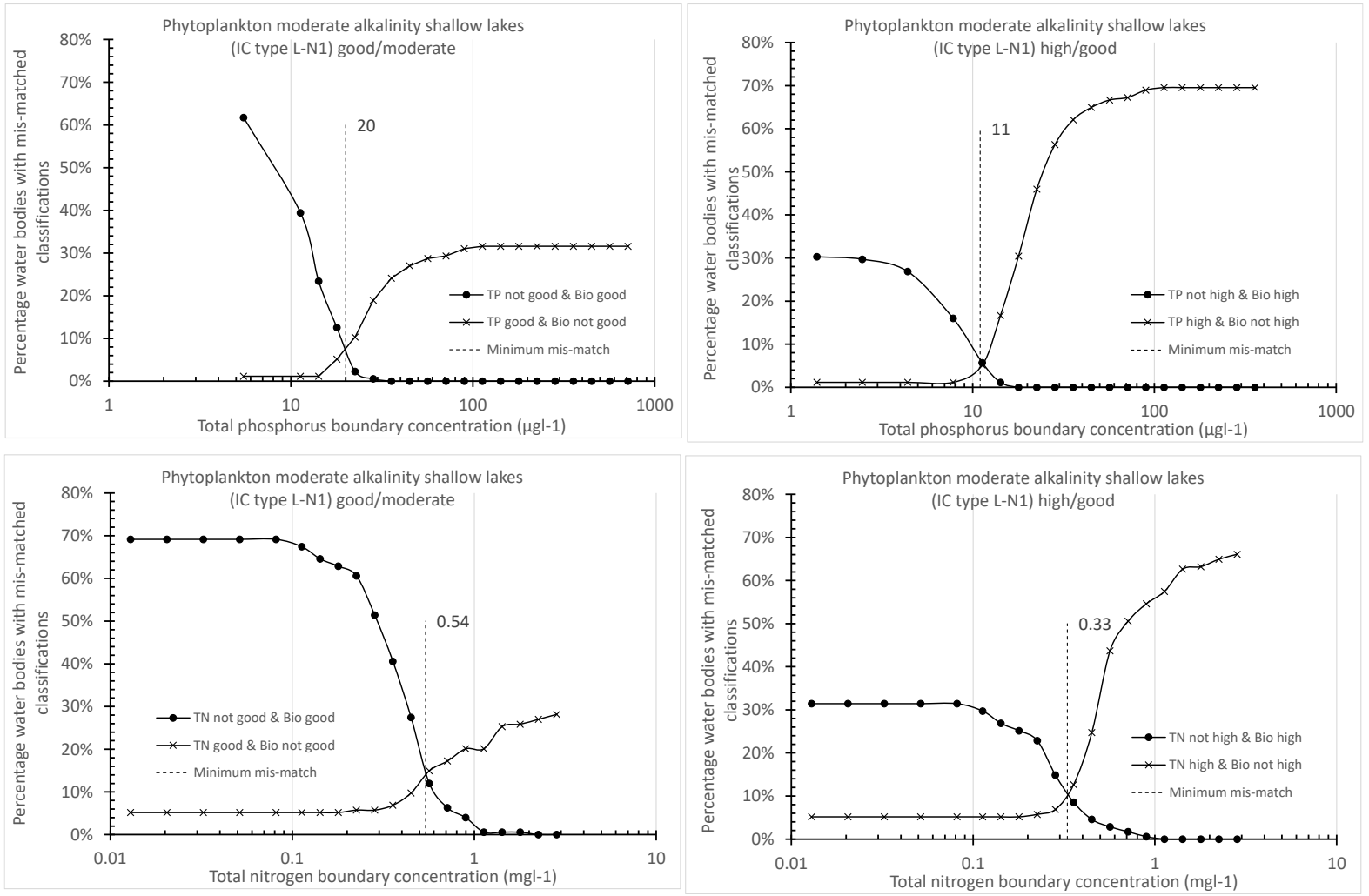


Figure 6-36 Percentage of water bodies where biology or total phosphorus / total nitrogen classifications for good ecological status differ in comparison to the level used to set a) the good/moderate and b) the high/ good boundaries. Biological status assessed using the common metric for phytoplankton in moderate alkalinity shallow (Type L-N1). Vertical lines mark intersection of curves where mis-match is minimised and equal.

Table 6-23 Regression parameters for L-N1 lake phytoplankton v total phosphorus and total nitrogen

```
Call: lm(formula = CM.EQR ~ log10(total.P) + log10(total.N), data = data.cc.ex,
    subset = total.P > P.minUsed & total.P <= P.maxUsed & total.N >
        N.minUsed & total.N <= N.maxUsed)

Residuals:
     Min       1Q   Median       3Q      Max
-0.52131 -0.05706  0.02082  0.07524  0.27520

Coefficients:
               Estimate Std. Error t value Pr(>|t|)
(Intercept)     1.60051    0.06691  23.922  < 2e-16 ***
log10(total.P) -0.73136    0.04799 -15.238  < 2e-16 ***
log10(total.N) -0.12434    0.04402  -2.824  0.00534 **
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 0.1167 on 160 degrees of freedom
Multiple R-squared:  0.8047,    Adjusted R-squared:  0.8023
F-statistic: 329.6 on 2 and 160 DF,  p-value: < 2.2e-16
```

Table 6-24 Regression parameters for L-N1 lake phytoplankton v total phosphorus

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x =
"interval", nperm = 99)

n = 172   r = -0.8944462   r-square = 0.8000341
Parametric P-values:   2-tailed = 2.601099e-61    1-tailed = 1.300549e-61
Angle between the two OLS regression lines = 6.362612 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS  1.756398 -0.8338259       -39.82222              0.01
2     MA  1.862629 -0.9245519       -42.75499              0.01
3    SMA  1.871614 -0.9322258       -42.99113                NA
4    RMA  1.884310 -0.9430684       -43.32172              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS       1.680269        1.832527 -0.8969399  -0.7707119
2     MA       1.783396        1.947594 -0.9971161  -0.8568830
3    SMA       1.800213        1.948013 -0.9974739  -0.8712459
4    RMA       1.804042        1.971620 -1.0176349  -0.8745156

Eigenvalues: 0.1495345 0.008285987

H statistic used for computing C.I. of MA: 0.001423543
```

Table 6-25 Regression parameters for L-N1 lake phytoplankton v total nitrogen

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x =
"interval", nperm = 99)

n = 167   r = -0.7261506   r-square = 0.5272947
Parametric P-values:   2-tailed = 1.210386e-28    1-tailed = 6.05193e-29
Angle between the two OLS regression lines = 17.85939 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS 0.6027383 -0.6296900       -32.19821              0.01
2     MA 0.5417946 -0.8222628       -39.42919              0.01
3    SMA 0.5275854 -0.8671617       -40.93058                NA
4    RMA 0.5310403 -0.8562447       -40.57162              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS      0.5619160       0.6435606 -0.7213328  -0.5380471
2     MA      0.5014514       0.5776528 -0.9497412  -0.7089563
3    SMA      0.4970549       0.5550595 -0.9636335  -0.7803478
4    RMA      0.4885414       0.5680093 -0.9905348  -0.7394282

Eigenvalues: 0.1500522 0.02315371

H statistic used for computing C.I. of MA: 0.005097492
```

6.2.3.1 Macrophyte IC Types 101 & 201 low/moderate alkalinity

For regression analysis types 101 and 201 were combined as the gradient was too short for analysis of type 101 independently.

Table 6-26- Predicted total phosphorus boundary values for low alkalinity deep lakes using regression models and categorical methods

IC Type	Macrophyte Models	R2	nutrient range TP $\mu g l^{-1}$	GM TP			HG TP		
				Pred	25th	75th	Pred	25th	75th
101 201	EQR v TP (OLS)	0.41	10 - 93	22	15	31	13	9	18
	TP v EQR (OLS)			22	16	29	17	13	24
	EQR v TP (RMA)			22	16	29	14	10	19
101	Average adjacent quartiles			10			8		
	Average adjacent classes			8	6	16	6	5	11
	Minimise class difference			19			10		
201	Average adjacent quartiles			20			13		
	Average adjacent classes			20	14	28	15	8	19
	Minimise class difference			21			13		

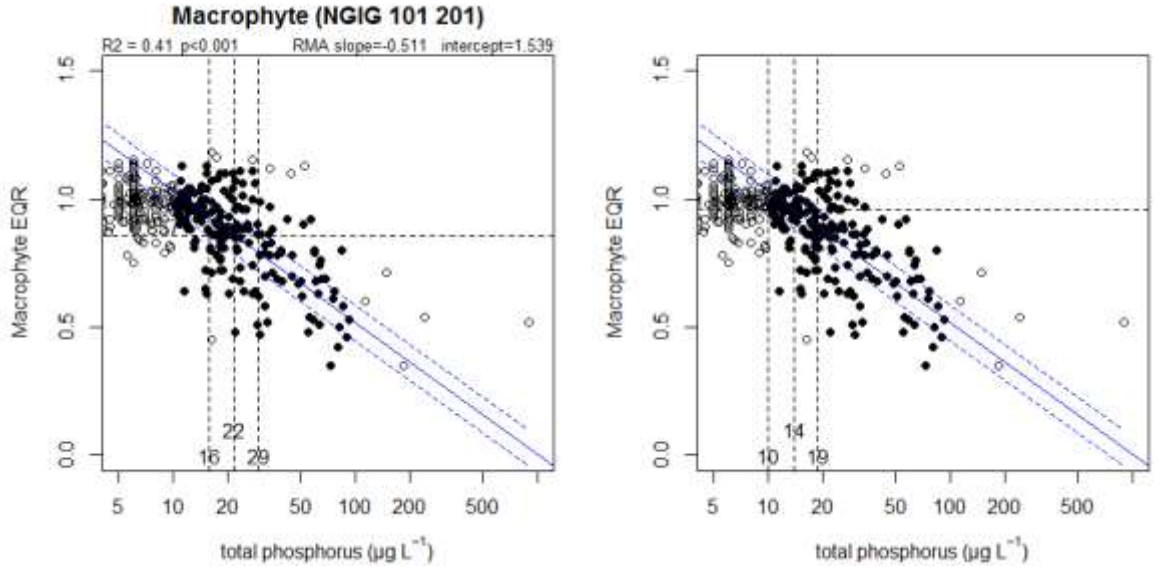


Figure 6-37 Relationship between common metric for macrophytes and total phosphorus for low/moderate alkalinity clear NGIG lakes (Types 101 201) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data, open circles data points excluded from regression.

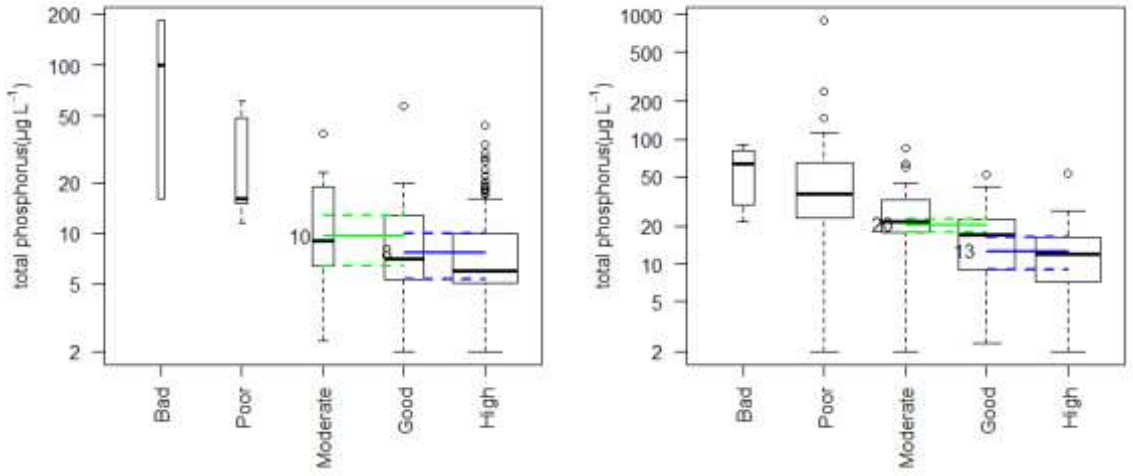


Figure 6-38 Box plots showing range of TP for a) low alkalinity (Type 101) & b) moderate alkalinity (Type 201) clear NGIG lakes classified using macrophytes common metric showing good/moderate boundary & high/good boundary values determined from the average of the upper and lower quartile values

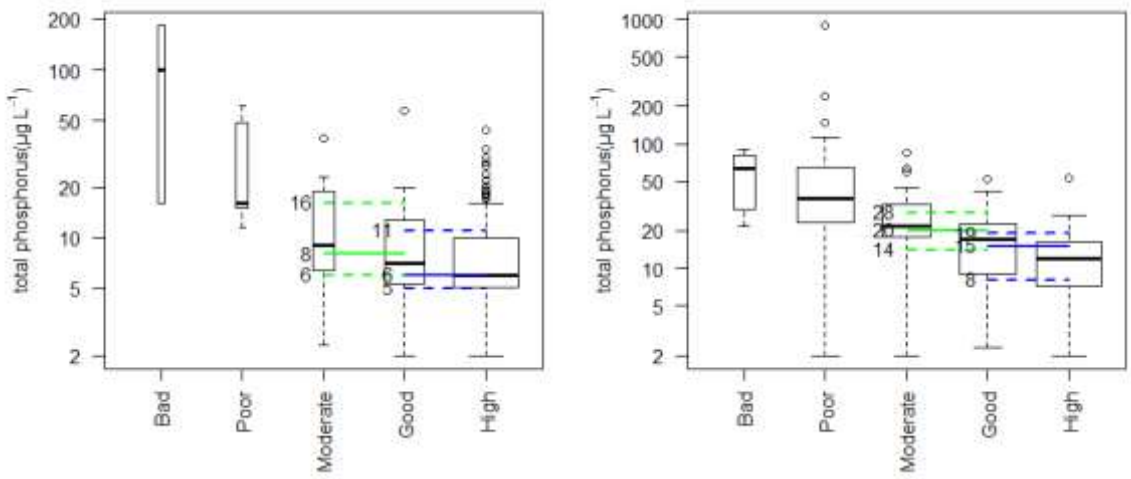


Figure 6-39 Box plots showing range of TP for a) low alkalinity (Type 101) & b) moderate alkalinity (Type 201) clear NGIG lakes classified using macrophyte common metric showing good/moderate boundary & high/good boundary values determined from the average of adjacent classes

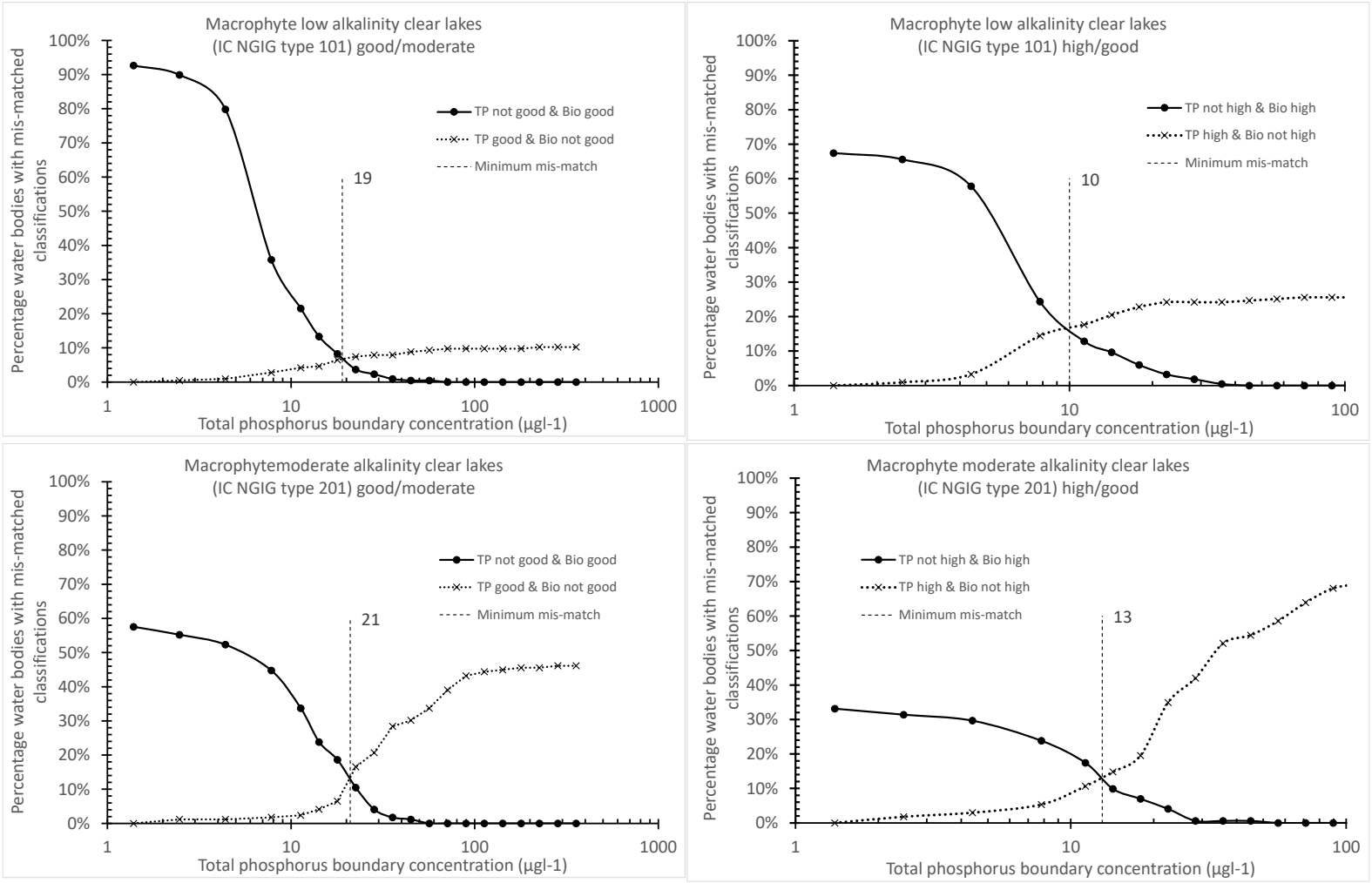


Figure 6-40 Percentage of water bodies where biology or total phosphorus / total nitrogen classifications for good ecological status differ in comparison to the level used to set a) the good/moderate and b) the high/ good boundaries. Biological status assessed using the common metric for macrophytes in low/moderate alkalinity clear NGIG lakes (Types 101 201). Vertical lines mark intersection of curves where mis-match is minimised and equal.

Table 6-27 Regression parameters for L-N2b lake macrophytes v total phosphorus

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x =
"interval", nperm = 99)

n = 197   r = -0.6404629   r-square = 0.4101927
Parametric P-values:   2-tailed = 3.899364e-24    1-tailed = 1.949682e-24
Angle between the two OLS regression lines = 23.0386 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS  1.427333 -0.4275923       -23.15118              0.01
2     MA  1.582314 -0.5435384       -28.52578              0.01
3    SMA  1.748183 -0.6676301       -33.72827                NA
4    RMA  1.539219 -0.5112977       -27.08055              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS       1.328958        1.525709 -0.5000068  -0.3551778
2     MA       1.463560        1.710635 -0.6395392  -0.4546947
3    SMA       1.656623        1.850211 -0.7439603  -0.5991313
4    RMA       1.426293        1.658390 -0.6004530  -0.4268143

Eigenvalues: 0.07292652 0.01262278

H statistic used for computing C.I. of MA: 0.005049183
```

6.3 Low and moderate alkalinity humic water lakes Northern GIG

6.3.1 Phytoplankton IC Type L-N3a low alkalinity shallow humic

Table 6-28- Predicted total phosphorus boundary values for low alkalinity deep lakes using regression models and categorical methods

IC Type	Phytoplankton Models	R^2	nutrient range TP μgl^{-1}			GM TP μgl^{-1}			HG TP μgl^{-1}		
						Pred	25th	75th	Pred	25th	75th
L-N3a	EQR v TP + TN (OLS)	0.61	4	-	77	22	18	27	12	10	15
	EQR v TP (OLS)	0.57	4	-	77	24	19	31	12	9	16
	TP v EQR (OLS)					17	14	22	12	10	15
	EQR v TP (RMA)					21	17	26	12	9	15
	Average adjacent quartiles					19			11		
	Average adjacent classes					19	15	23	12	9	15
	Minimise class difference					22			14		

IC Type	Phytoplankton Models	R^2	nutrient range TN mgl^{-1}			GM TN mgl^{-1}			HG TN mgl^{-1}		
L-N3a	EQR v TP + TN (OLS)	0.61	0.22	-	1.00	0.72	0.61	0.84	0.46	0.40	0.53
	EQR v TN (OLS)	0.28	0.22	-	1.23	0.83	0.64	1.07	0.47	0.36	0.60
	TN v EQR (OLS)					0.52	0.46	0.60	0.44	0.39	0.51
	EQR v TN (RMA)					0.66	0.55	0.8	0.46	0.38	0.55
	Average adjacent quartiles					0.53			0.43		
	Average adjacent classes					0.56	0.47	0.63	0.43	0.37	0.49
	Minimise class difference					0.63			0.41		

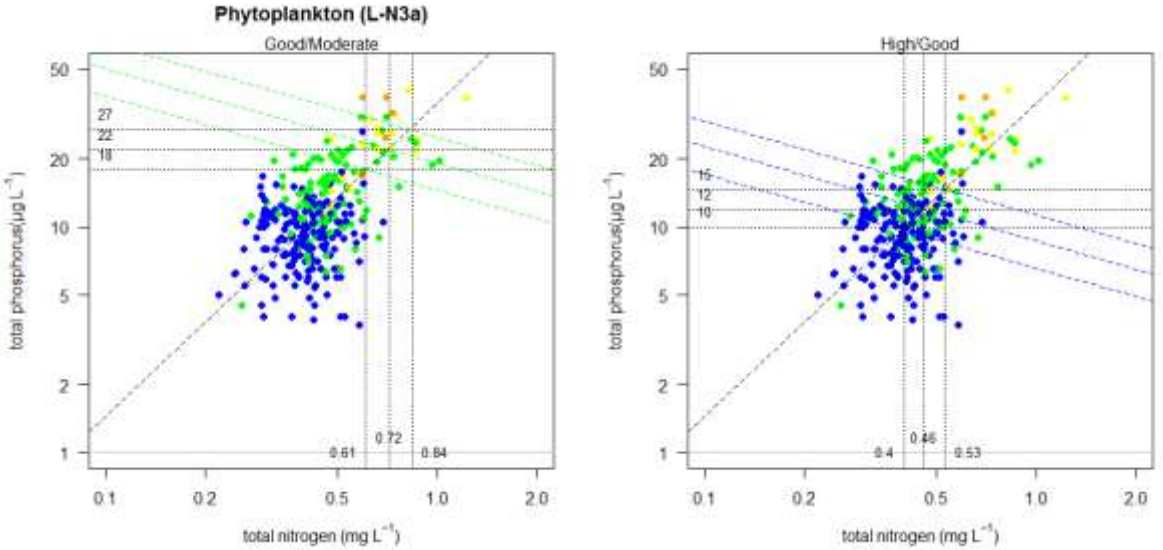


Figure 6-41 Relationship between mean TP and TN, points coloured by WFD class for phytoplankton in low alkalinity shallow humic NGIG lakes. Dotted lines contours of predicted TN & TP concentration when phytoplankton common metric EQR is at a) good/moderate boundary (green lines) and b) high/good boundary, dotted lines show ± 25th & 75th residuals of prediction. Horizontal & vertical lines show intersection with RMA regression of observed TP and TN showing boundary values.

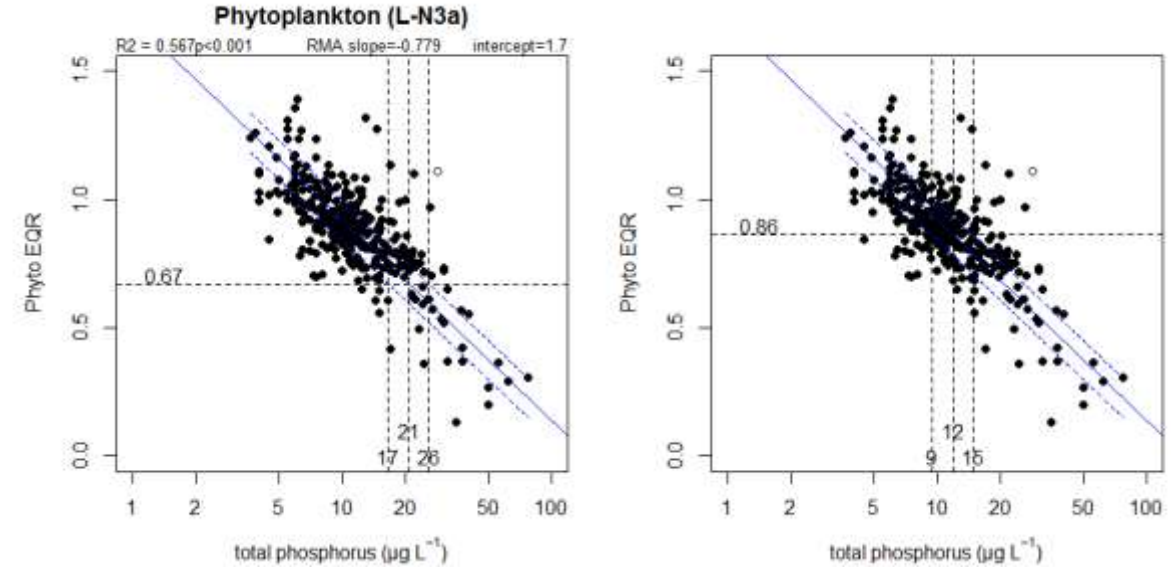


Figure 6-42 Relationship between common metric for phytoplankton and total phosphorus for low alkalinity shallow humic NGIG lakes, showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data, open circles data points excluded from regression.

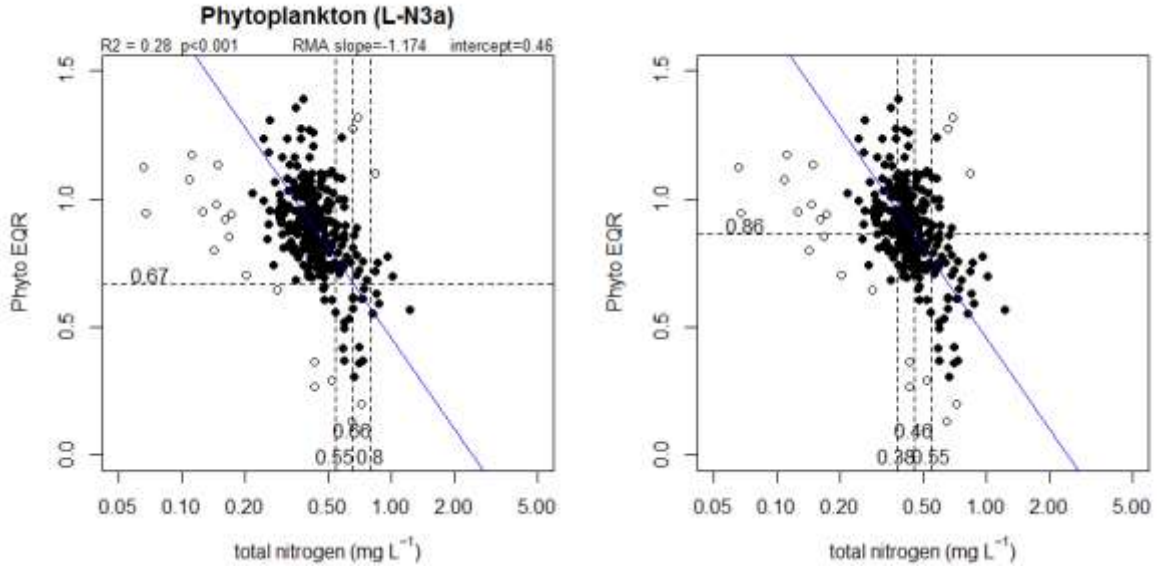


Figure 6-43 Relationship between common metric for phytoplankton and total nitrogen for low alkalinity shallow humic NGIG lakes, showing a) good/moderate boundary and b) high/ good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data, open circles data points excluded from regression.

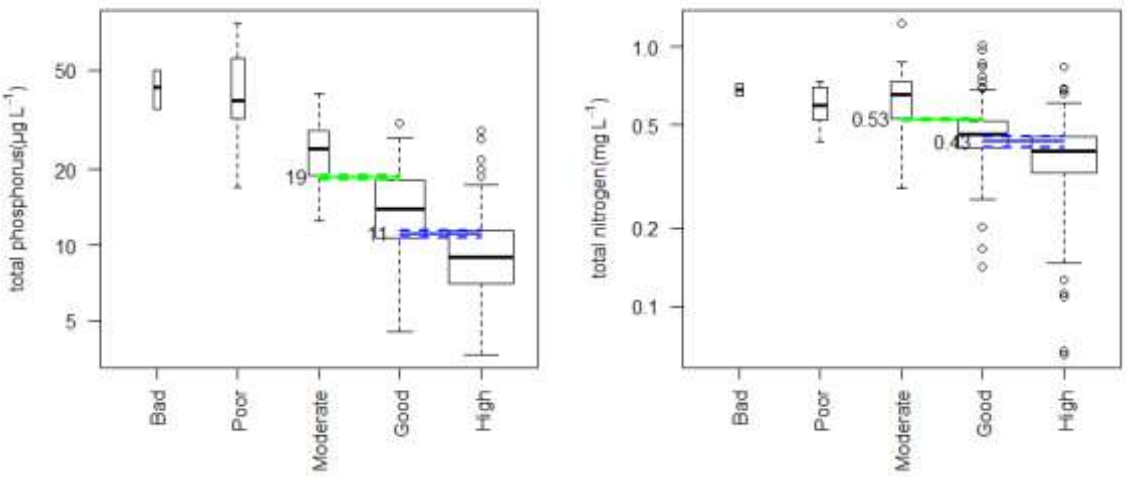


Figure 6-44 Box plots showing range of a) TP and b) TN for low alkalinity shallow humic NGIG lakes classified using phytoplankton common metric showing good/moderate boundary and high/good boundary values determined from the average of the upper and lower quartile values.

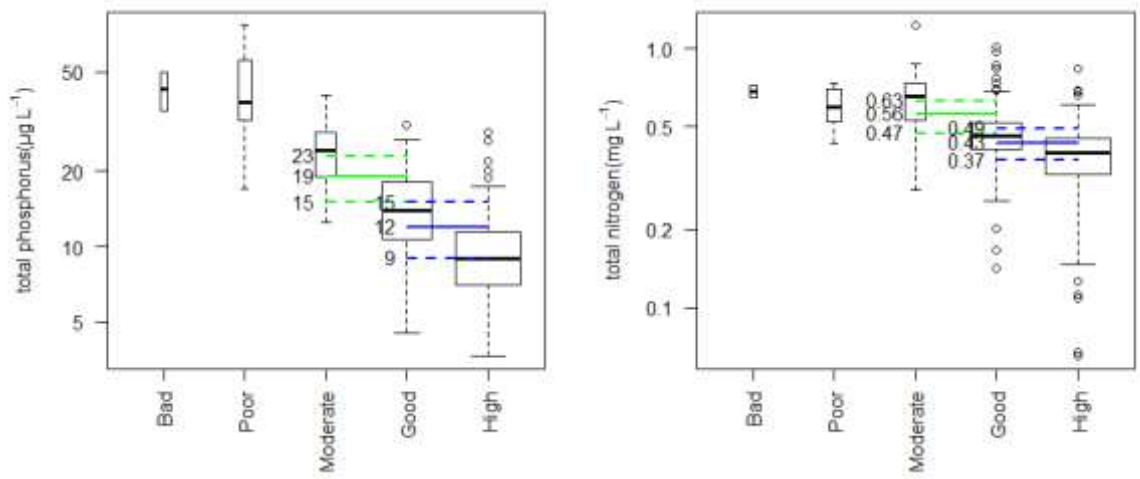


Figure 6-45 Box plots showing range of of a) TP and b) TN for low alkalinity shallow humic NGIG lakes classified using macrophyte common metric showing good/moderate boundary and high/good boundary values determined from the average of adjacent classes

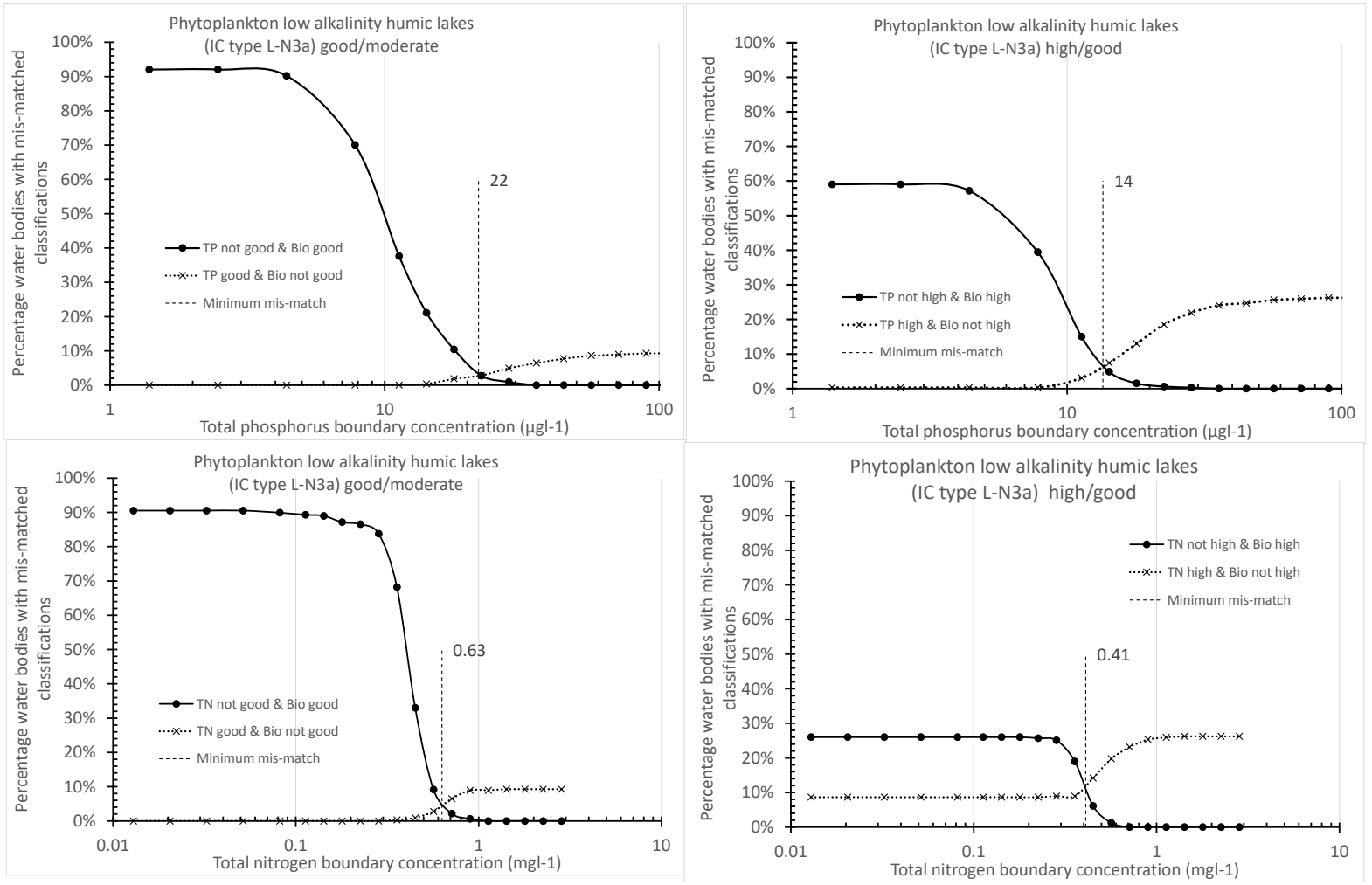


Figure 6-46 Percentage of water bodies where biology or total phosphorus / total nitrogen classifications for good ecological status differ in comparison to the level used to set a) the good/moderate and b) the high/ good boundaries. Biological status assessed using the common metric for phytoplankton in low alkalinity shallow humic NGIG lakes. Vertical lines mark intersection of curves where mis-match is minimised and equal.

Table 6-29 Regression parameters for L-N2b lake phytoplankton v total phosphorus and total nitrogen

```
Call:
lm(formula = CM.EQR ~ log10(total.P) + log10(total.N), data = data.cc.ex,
    subset = total.P > P.minUsed & total.P <= P.maxUsed & total.N >
        N.minUsed & total.N <= N.maxUsed)

Residuals:
     Min       1Q   Median       3Q      Max
-0.33639 -0.06212  0.00015  0.06653  0.35099

Coefficients:
              Estimate Std. Error t value Pr(>|t|)
(Intercept)    1.37726    0.05151  26.739  < 2e-16 ***
log10(total.P) -0.55062    0.03412 -16.137  < 2e-16 ***
log10(total.N) -0.22903    0.06158  -3.719 0.000238 ***
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 0.1035 on 301 degrees of freedom
Multiple R-squared:  0.6107,    Adjusted R-squared:  0.6081
F-statistic: 236.1 on 2 and 301 DF,  p-value: < 2.2e-16
:
```

Table 6-30 Regression parameters for L-N2b lake phytoplankton v total phosphorus

```
Model II regression Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x =
"interval", nperm = 99)

n = 330   r = -0.7531031   r-square = 0.5671643
Parametric P-values:   2-tailed = 1.326782e-61    1-tailed = 6.633912e-62
Angle between the two OLS regression lines = 15.79683 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS  1.544058 -0.6307689       -32.24245              0.01
2     MA  1.713098 -0.7910002       -38.34400              0.01
3    SMA  1.762218 -0.8375597       -39.94819                NA
4    RMA  1.700125 -0.7787026       -37.90798              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS       1.479561        1.608554 -0.6906230  -0.5709148
2     MA       1.636624        1.795376 -0.8689902  -0.7185117
3    SMA       1.701326        1.827616 -0.8995498  -0.7798416
4    RMA       1.624628        1.780874 -0.8552433  -0.7071403

Eigenvalues: 0.07241528 0.009786274

H statistic used for computing C.I. of MA: 0.002131718
```

Table 6-31 Regression parameters for L-N2b lake phytoplankton v total nitrogen

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x =
"interval", nperm = 99)

n = 305   r = -0.5287854   r-square = 0.279614
Parametric P-values:   2-tailed = 2.265311e-23    1-tailed = 1.132655e-23
Angle between the two OLS regression lines = 32.49877 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS 0.6067705 -0.7656217       -37.43845              0.01
2     MA 0.1816928 -1.9459088       -62.80143              0.01
3    SMA 0.3610541 -1.4478874       -55.36866                NA
4    RMA 0.4598457 -1.1735786       -49.56586              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS      0.5542729       0.6592681 -0.9045473  -0.6266960
2     MA      0.0327393       0.2931617 -2.3594989  -1.6363998
3    SMA      0.3086254       0.4086929 -1.5934628  -1.3156115
4    RMA      0.3786727       0.5329955 -1.3989668  -0.9704682

Eigenvalues: 0.03269714 0.007965357

H statistic used for computing C.I. of MA: 0.005441694
```

6.3.2 Phytoplankton IC Type L-N8a moderate alkalinity humic

Table 6-32- Predicted total phosphorus boundary values for moderate alkalinity shallow humic lakes using regression models and categorical methods

IC Type	Phytoplankton Models	R2	nutrient range TP µgl^{-1}	GM TP µgl^{-1}			HG TP µgl^{-1}		
				Pred	25th	75th	Pred	25th	75th
L-N8a	EQR v TP + TN (OLS)	0.80	4 - 127	27	23	32	16	13	19
	EQR v TP (OLS)	0.74	4 - 127	27	22	38	14	11	20
	TP v EQR (OLS)			26	20	34	16	13	21
	EQR v TP (RMA)			27	21	35	15	12	20
	Average adjacent quartiles			27			16		
	Average adjacent classes			28	20	35	19	12	23
	Minimise class difference			27			16		

IC Type	Phytoplankton	R2	nutrient range TN mgl^{-1}	GM TN mgl^{-1}			HG TN mgl^{-1}		
				Pred	25th	75th	Pred	25th	75th
L-N8a	EQR v TP + TN (OLS)	0.80	-	0.85	0.72	1.07	0.47	0.39	0.58
	EQR v TN (OLS)	0.24	0.22 - 1.91	0.90	0.53	1.55	0.40	0.24	0.70
	TN v EQR (OLS)			0.78	0.60	1.04	0.65	0.50	0.86
	EQR v TN (RMA)			0.83	0.55	1.12	0.53	0.35	0.71
	Average adjacent quartiles			0.86			0.68		
	Average adjacent classes			0.82	0.68	1.03	0.65	0.53	0.87
	Minimise class difference			0.80			0.55		

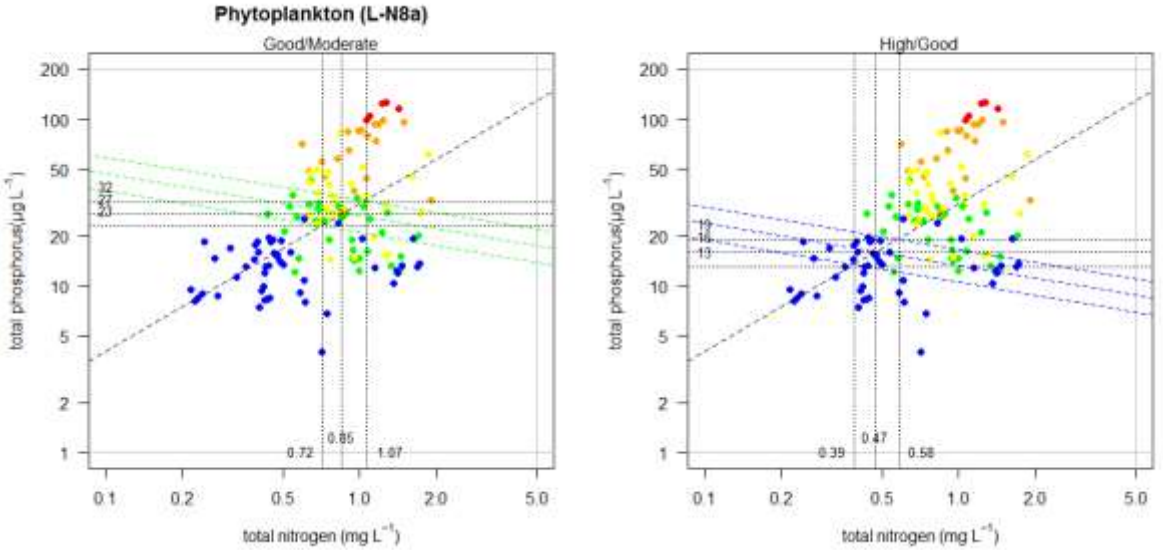


Figure 6-47 Relationship between mean TP and TN, points coloured by WFD class for phytoplankton in moderate alkalinity humic NGIG lakes (Type L-N8a). Dotted lines contours of predicted TN & TP concentration when phytoplankton common metric EQR is at a) good/moderate boundary (green lines) and b) high/good boundary, dotted lines show ± 25th & 75th residuals of prediction. Horizontal & vertical lines show intersection with RMA regression of observed TP and TN showing boundary values.

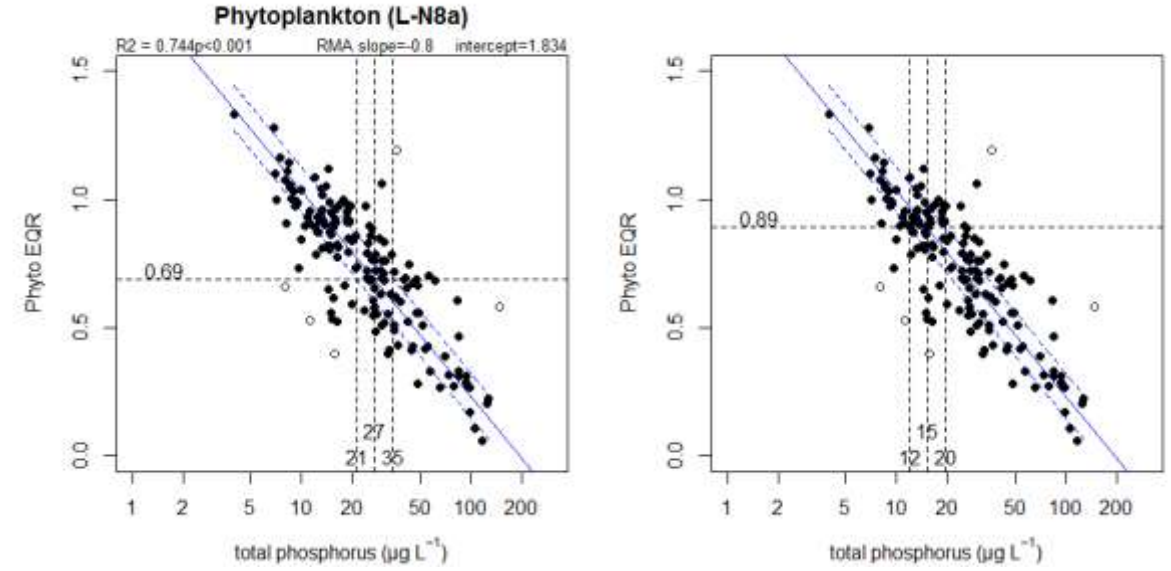


Figure 6-48 Relationship between common metric for phytoplankton and total phosphorus for moderate alkalinity humic NGIG lakes (Type L-N8a) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data, open circles data points excluded from regression.

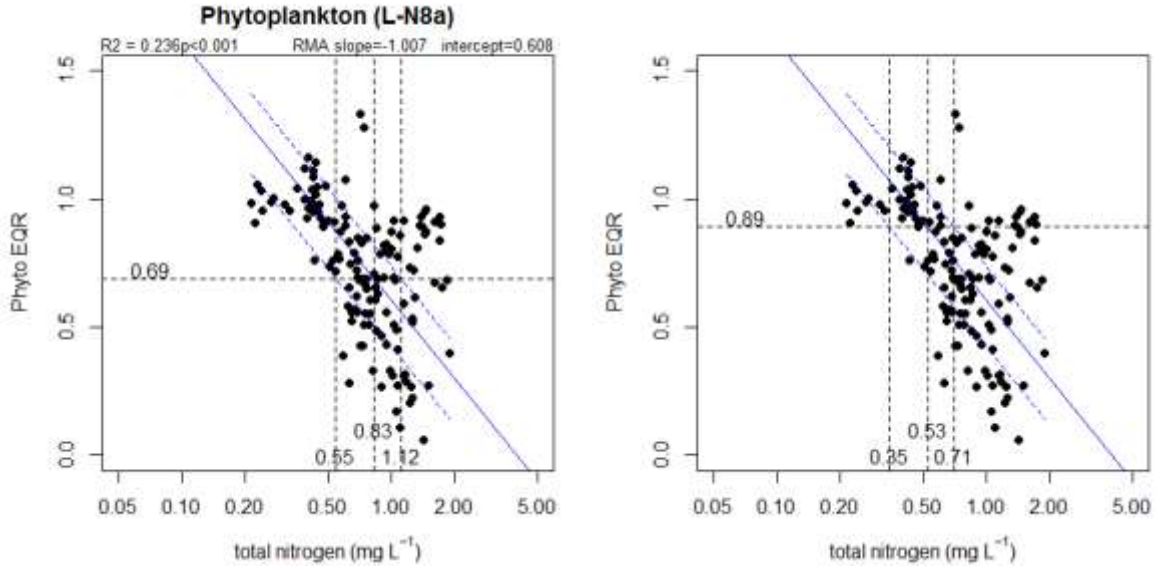


Figure 6-49 Relationship between common metric for phytoplankton and total nitrogen for moderate alkalinity humic NGIG lakes (Type L-N8a) showing a) good/moderate boundary and b) high/ good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data, open circles data points excluded from regression.

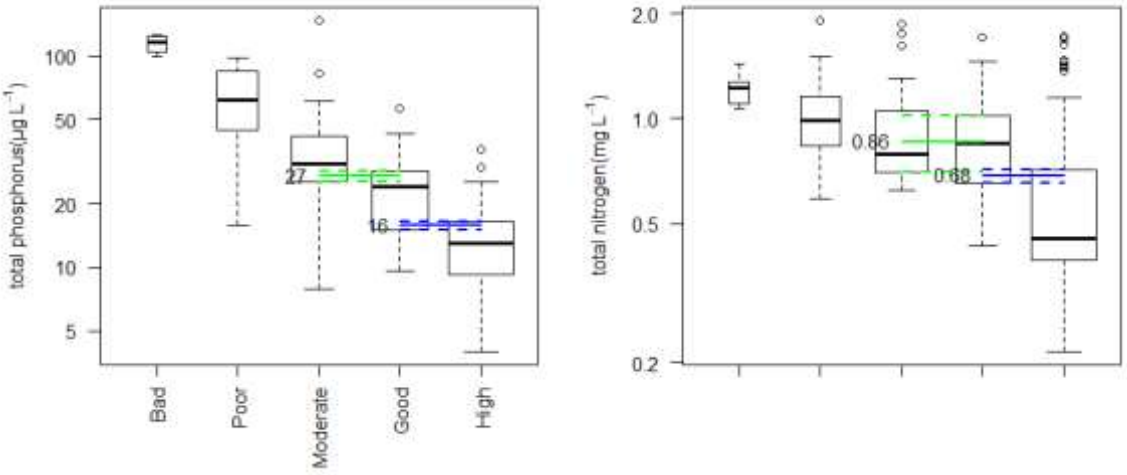


Figure 6-50 Box plots showing range of total phosphorus for moderate alkalinity humic NGIG lakes (Type L-N8a) classified using phytoplankton common metric showing good/moderate boundary & high/good boundary values determined from the average of the upper and lower quartile values

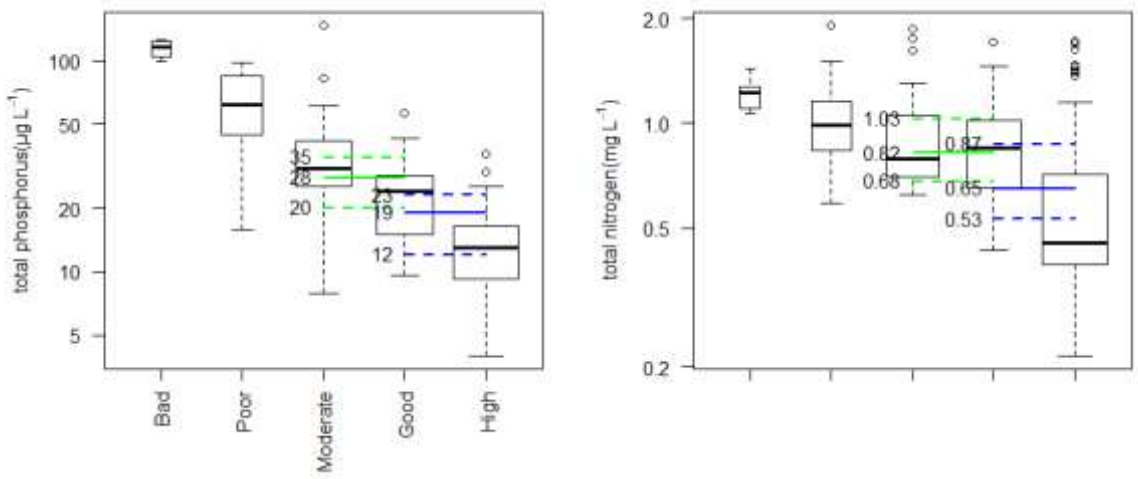


Figure 6-51 Box plots showing range of total phosphorus for moderate alkalinity humic NGIG lakes (Type L-N8a) classified using macrophyte common metric showing good/moderate boundary & high/good boundary values determined from the average of adjacent classes

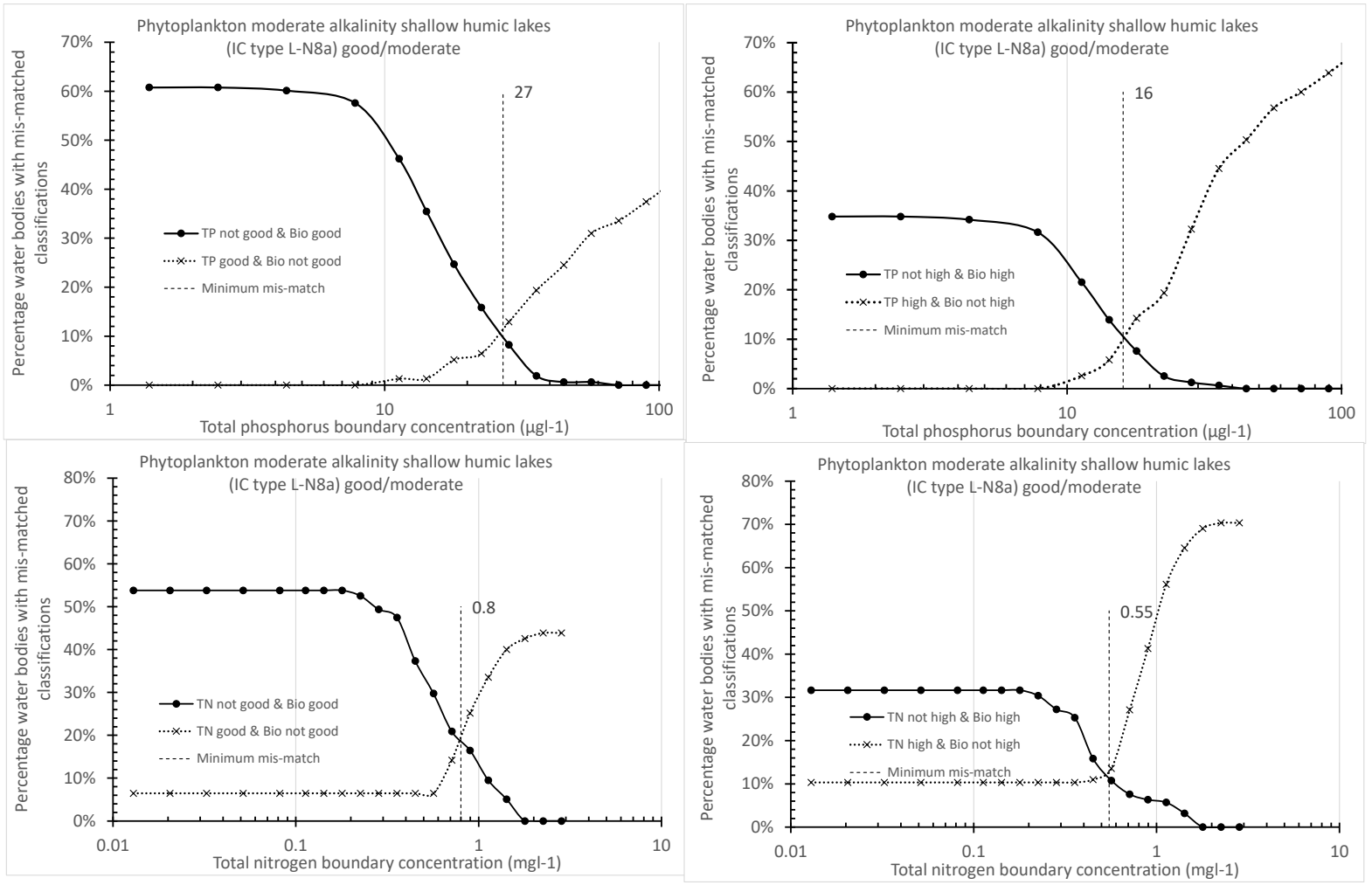


Figure 6-52 Percentage of water bodies where biology or total phosphorus / total nitrogen classifications for good ecological status differ in comparison to the level used to set a) the good/moderate and b) the high/ good boundaries. Biological status assessed using the common metric for phytoplankton in moderate alkalinity humic NGIG lakes (Type L-N8a). Vertical lines mark intersection of curves where mis-match is minimised and equal.

Table 6-33 Regression parameters for L-N8a lake phytoplankton v total phosphorus and total nitrogen

```
Call:
lm(formula = CM.EQR ~ log10(total.P) + log10(total.N), data = data.cc.ex,
    subset = total.P > P.minUsed & total.P <= P.maxUsed & total.N >
        N.minUsed & total.N <= N.maxUsed)

Residuals:
     Min       1Q   Median       3Q      Max
-0.30364 -0.06552 -0.00231  0.06798  0.28965

Coefficients:
              Estimate Std. Error t value Pr(>|t|)
(Intercept)     1.65143    0.05084  32.485  < 2e-16 ***
log10(total.P) -0.67822    0.03405 -19.920  < 2e-16 ***
log10(total.N) -0.17244    0.04943  -3.489 0.000651 ***
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 0.1145 on 139 degrees of freedom
Multiple R-squared:  0.8024,    Adjusted R-squared:  0.7996
F-statistic: 282.3 on 2 and 139 DF,  p-value: < 2.2e-16
:
```

Table 6-34 Regression parameters for L-N8a lake phytoplankton v total phosphorus

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x =
"interval", nperm = 99)

n = 159   r = -0.8623878   r-square = 0.7437127
Parametric P-values:   2-tailed = 2.829647e-48    1-tailed = 1.414823e-48
Angle between the two OLS regression lines = 8.272317 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS  1.696428 -0.6994209       -34.96975              0.01
2     MA  1.813654 -0.7848384       -38.12619              0.01
3    SMA  1.849597 -0.8110283       -39.04303                NA
4    RMA  1.833778 -0.7995019       -38.64240              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS       1.605395        1.787460 -0.7641439  -0.6346980
2     MA       1.717283        1.917080 -0.8602002  -0.7146171
3    SMA       1.764310        1.941961 -0.8783297  -0.7488838
4    RMA       1.736185        1.939823 -0.8767716  -0.7283906

Eigenvalues: 0.147711 0.01037879

H statistic used for computing C.I. of MA: 0.002019913
```

Table 6-35 Regression parameters for L-N8a lake phytoplankton v total nitrogen

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x =
"interval", nperm = 99)

n = 143   r = -0.4858969   r-square = 0.2360958
Parametric P-values:   2-tailed = 7.672302e-10    1-tailed = 3.836151e-10
Angle between the two OLS regression lines = 37.73876 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS 0.6622749 -0.5797786       -30.10424              0.01
2     MA 0.5550223 -1.4301318       -55.03736              0.01
3    SMA 0.5849041 -1.1932131       -50.03453                NA
4    RMA 0.6083746 -1.0071278       -45.20347              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS      0.6192806       0.7052693 -0.7534065  -0.4061507
2     MA      0.4865516       0.6003704 -1.9730018  -1.0705886
3    SMA      0.5614200       0.6052184 -1.3794074  -1.0321516
4    RMA      0.5655566       0.6432306 -1.3466101  -0.7307720

Eigenvalues: 0.08364596 0.02719046

H statistic used for computing C.I. of MA: 0.0197794
```

6.3.3 Phytoplankton IC Type L-N6a mid-altitude low alkalinity shallow humic

Table 6-36- Predicted total phosphorus boundary values for mid-altitude low alkalinity shallow lakes using regression models and categorical methods

IC Type	Phytoplankton Models	R^2	nutrient range TP $\mu g l^{-1}$	GM TP $\mu g l^{-1}$			HG TP $\mu g l^{-1}$		
				Pred	25th	75th	Pred	25th	75**th**
L-N6a	EQR v TP + TN (OLS)	0.41	-	25	19	34	14	10	19
	EQR v TP (OLS)	0.39	2 - 74	31	21	44	15	10	21
	TP v EQR (OLS)			14	10	17	10	8	13
	EQR v TP (RMA)			25	18	33	14	9	18
	Average adjacent quartiles			16			11		
	Average adjacent classes			18	15	23	10	9	12
	Minimise class difference			20			13		

IC Type	Phytoplankton Models	R^2	nutrient range TN $mg l^{-1}$	GM TN $mg l^{-1}$			HG TN $mg l^{-1}$		
				Pred	25th	75th	Pred	25th	75^{th}
L-N6a	EQR v TP + TN (OLS)	0.41	-	0.60	0.50	0.72	0.41	0.34	0.50
	EQR v TN (OLS)	0.26	0.13 - 0.80	0.70	0.55	0.89	0.44	0.34	0.56
	TN v EQR (OLS)			0.37	0.31	0.43	0.33	0.28	0.38
	EQR v TN (RMA)			0.6	0.47	0.75	0.41	0.32	0.51
	Average adjacent quartiles			0.37			0.33		
	Average adjacent classes			0.36	0.32	0.45	0.31	0.27	0.37
	Minimise class difference			0.46			0.39		

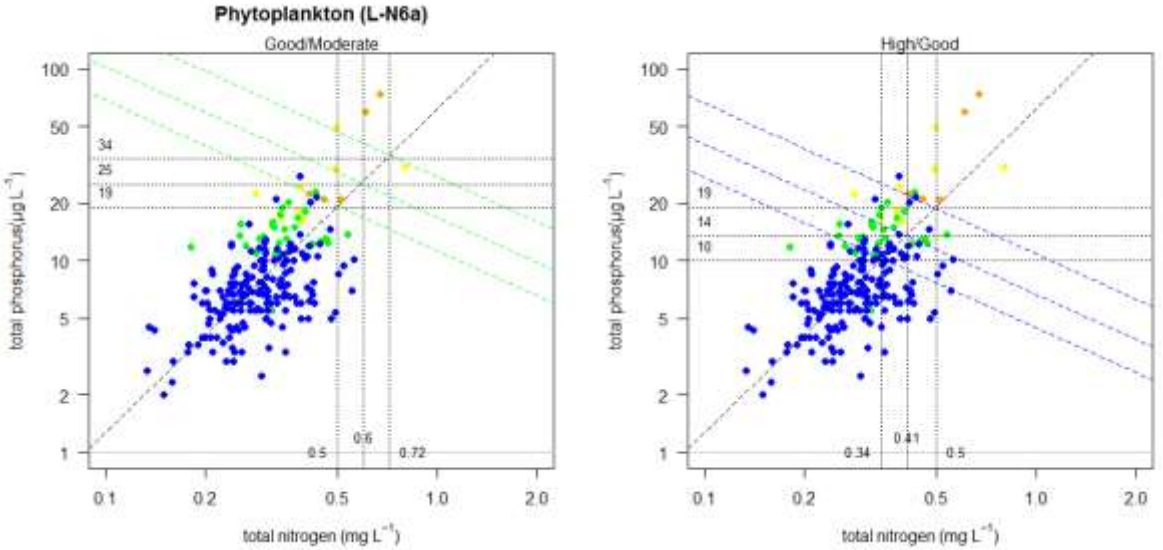


Figure 6-53 Relationship between mean TP and TN, points coloured by WFD class for phytoplankton in mid-altitude low alkalinity shallow humic NGIG (Type L-N6a). Dotted lines contours of predicted TN & TP concentration when phytoplankton common metric EQR is at a) good/moderate boundary (green lines) and b) high/good boundary, dotted lines show ± 25th & 75th residuals of prediction. Horizontal & vertical lines show intersection with RMA regression of observed TP and TN showing boundary values.

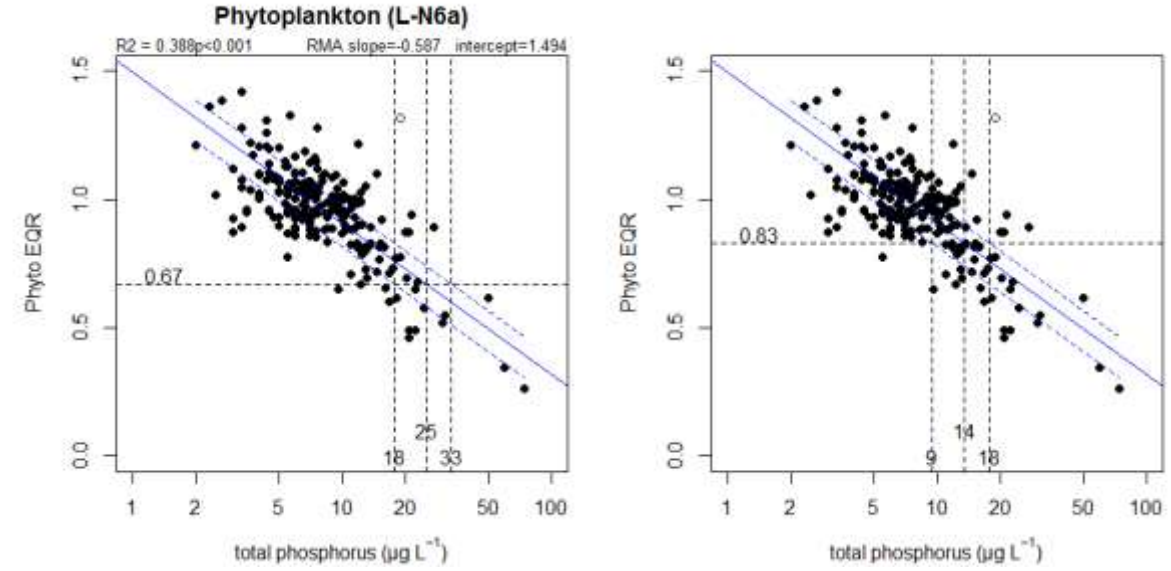


Figure 6-54 Relationship between common metric for phytoplankton and total phosphorus for mid-altitude low alkalinity shallow humic NGIG (Type L-N6a) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data, open circles data points excluded from regression.

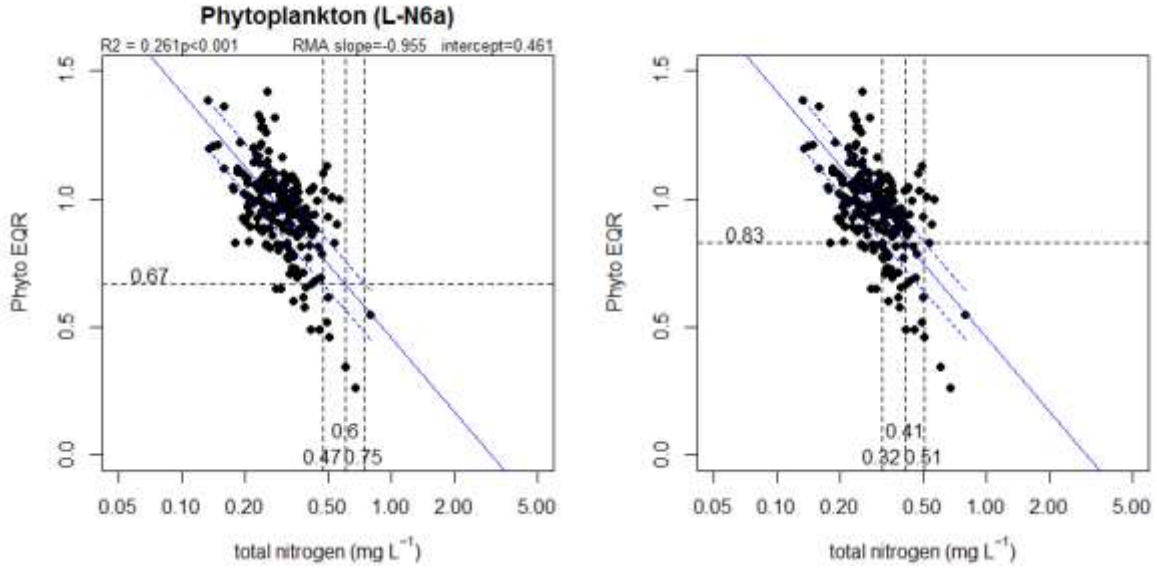


Figure 6-55 Relationship between common metric for phytoplankton and total nitrogen for mid-altitude low alkalinity shallow humic NGIG (Type L-N6a) showing a) good/moderate boundary and b) high/ good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data, open circles data points excluded from regression.

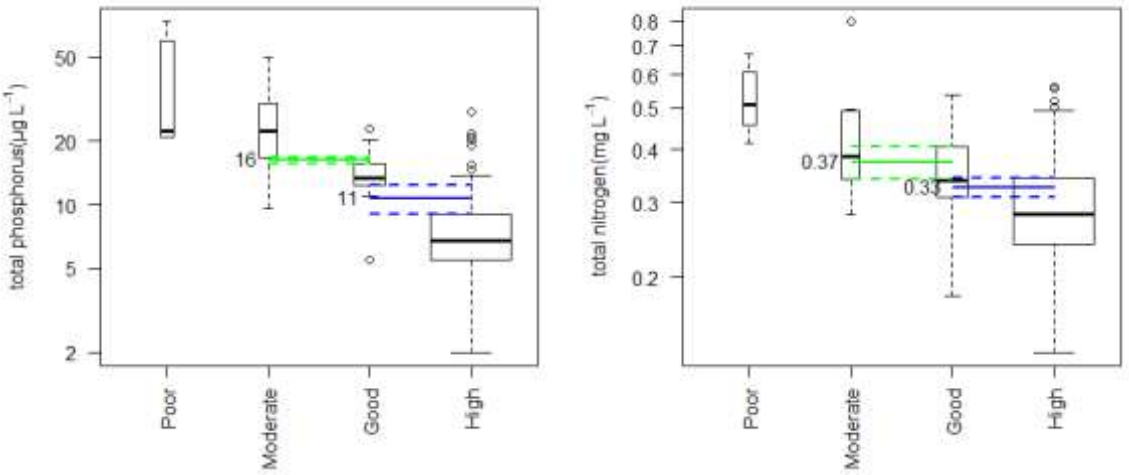


Figure 6-56 Box plots showing range of a) total phosphorus and b) total nitrogen for mid-altitude low alkalinity shallow humic NGIG lakes(Type L-N6a) classified using phytoplankton common metric showing good/moderate boundary & high/good boundary values determined from the average of the upper and lower quartile values

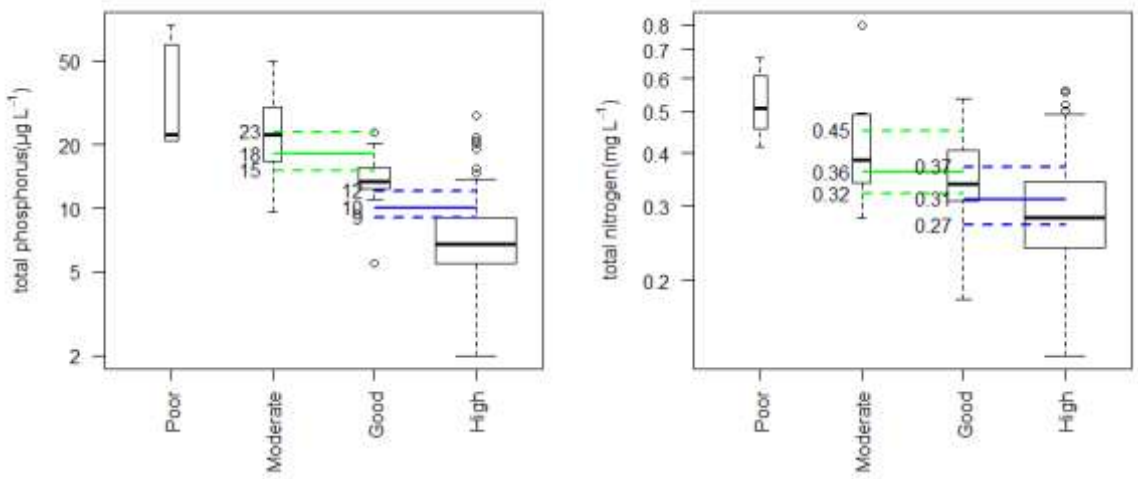


Figure 6-57 Box plots showing range of a) total phosphorus and b) total nitrogen for mid-altitude low alkalinity shallow humic NGIG (Type L-N6a) classified using macrophyte common metric showing good/moderate boundary & high/good boundary values determined from the average of adjacent classes

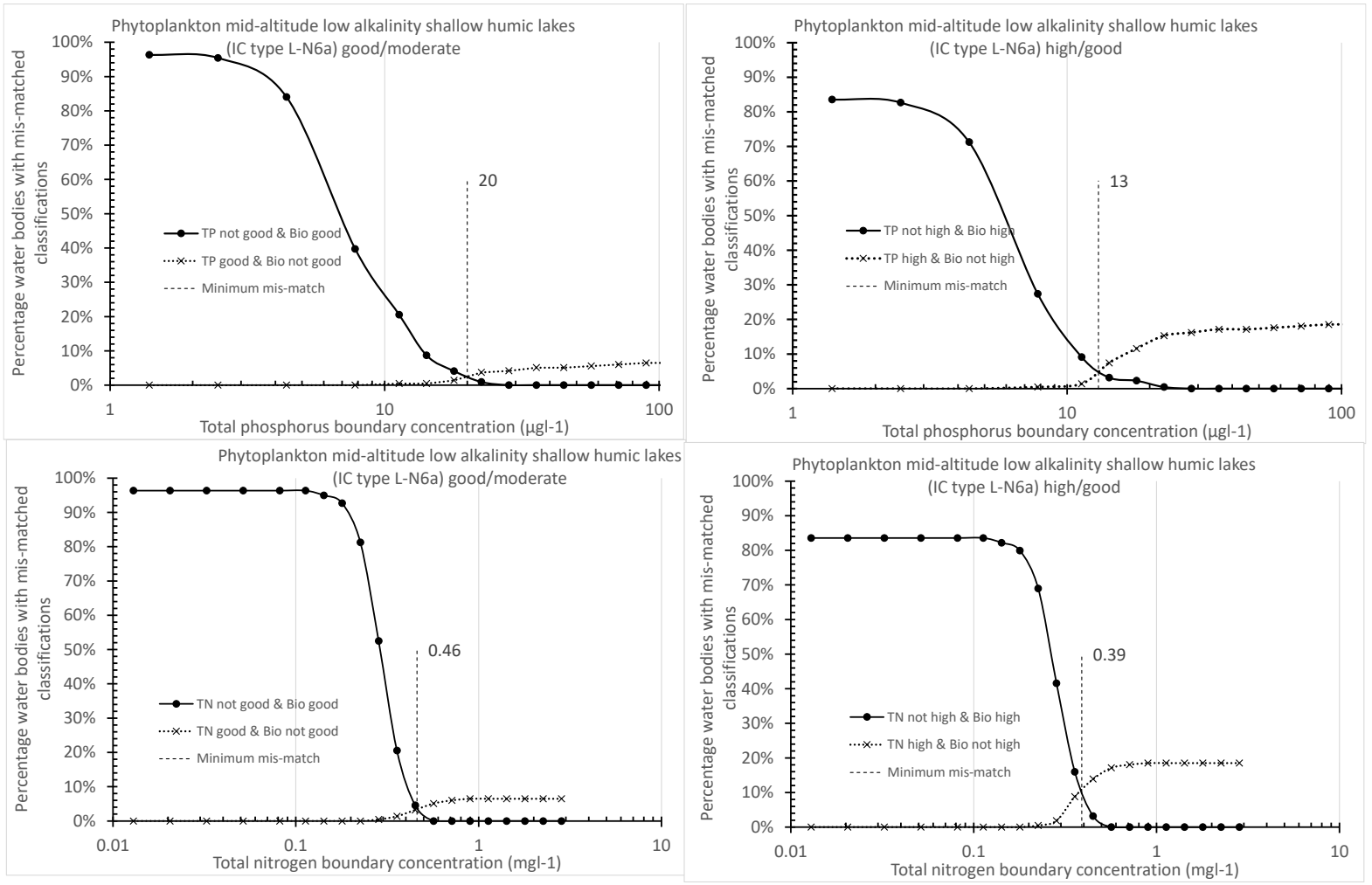


Figure 6-58 Percentage of water bodies where biology or total phosphorus / total nitrogen classifications for good ecological status differ in comparison to the level used to set a) the good/moderate and b) the high/good boundaries. Biological status assessed using the common metric for phytoplankton in mid-altitude low alkalinity shallow humic NGIG (Type L-N6a). Vertical lines mark intersection of curves where mis-match is minimised and equal.

Table 6-37 Regression parameters for L-N2b lake phytoplankton v total phosphorus and total nitrogen

```
Call:
lm(formula = CM.EQR ~ log10(total.P) + log10(total.N), data = data.cc.ex,
    subset = total.P > P.minUsed & total.P <= P.maxUsed & total.N >
        N.minUsed & total.N <= N.maxUsed)

Residuals:
     Min       1Q   Median       3Q      Max
-0.28889 -0.08458 -0.00471  0.06879  1.48473

Coefficients:
              Estimate Std. Error t value Pr(>|t|)
(Intercept)    1.15910    0.09192  12.610  < 2e-16 ***
log10(total.P) -0.39788    0.05294  -7.516 1.39e-12 ***
log10(total.N) -0.31085    0.10144  -3.064  0.00245 **
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 0.1513 on 221 degrees of freedom
Multiple R-squared:  0.4131,    Adjusted R-squared:  0.4078
F-statistic: 77.77 on 2 and 221 DF,  p-value: < 2.2e-16
```

Table 6-38 Regression parameters for L-N2b lake phytoplankton v total phosphorus

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x =
"interval", nperm = 99)

n = 224   r = -0.623015   r-square = 0.3881476
Parametric P-values:   2-tailed = 1.772343e-25    1-tailed = 8.861714e-26
Angle between the two OLS regression lines = 25.60656 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS  1.414506 -0.4993440       -26.53497              0.01
2     MA  1.600039 -0.7041015       -35.14943              0.01
3    SMA  1.688289 -0.8014959       -38.71203                NA
4    RMA  1.493756 -0.5868055       -30.40465              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS       1.336678        1.492334 -0.5822661  -0.4164219
2     MA       1.499327        1.712520 -0.8282385  -0.5929544
3    SMA       1.617029        1.767301 -0.8886961  -0.7228519
4    RMA       1.407167        1.584114 -0.6865264  -0.4912447

Eigenvalues: 0.08129021 0.01749034

H statistic used for computing C.I. of MA: 0.006110663
```

Table 6-39 Regression parameters for L-N2b lake phytoplankton v total nitrogen

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x =
"interval", nperm = 99)

n = 225   r = -0.5109485   r-square = 0.2610684
Parametric P-values:   2-tailed = 2.306726e-16    1-tailed = 1.153363e-16
Angle between the two OLS regression lines = 33.40017 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method  Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS  0.5479232 -0.7903008       -38.31935              0.01
2     MA -0.2007194 -2.2135930       -65.68873              0.01
3    SMA  0.1500448 -1.5467329       -57.11636                NA
4    RMA  0.4614712 -0.9546603       -43.67122              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS     0.45296317      0.64288328 -0.9657602  -0.6148415
2     MA    -0.51901602      0.01818036 -2.8187271  -1.7974288
3    SMA     0.05253638      0.23711736 -1.7321124  -1.3811937
4    RMA     0.34755172      0.57104437 -1.1712399  -0.7463438

Eigenvalues: 0.04484177 0.01048674

H statistic used for computing C.I. of MA: 0.006938419
```

6.3.4 Macrophyte IC Types 102 & 202 low/moderate alkalinity humic lakes

Table 6-40- Predicted total phosphorus boundary values for low/moderate alkalinity humic lakes using regression models and categorical methods

IC Type	Macrophyte Models	R2	nutrient range TP µgl^{-1}	GM TP			HG TP		
				Pred	25th	75th	Pred	25th	75th
102	EQR v TP (OLS)	ns	10 - 100						
	TP v EQR (OLS)								
	EQR v TP (RMA)								
202	EQR v TP (OLS)	0.31	11 - 363	36	18	61	18	9	30
	TP v EQR (OLS)			35	21	49	28	17	39
	EQR v TP (RMA)			36	19	54	20	10	29
102	Average adjacent quartiles			23			18		
	Average adjacent classes			24	16	33	19	11	25
	Minimise class difference			37			24		
202	Average adjacent quartiles			30			20		
	Average adjacent classes			24	18	46	18	13	32
	Minimise class difference			31			22		

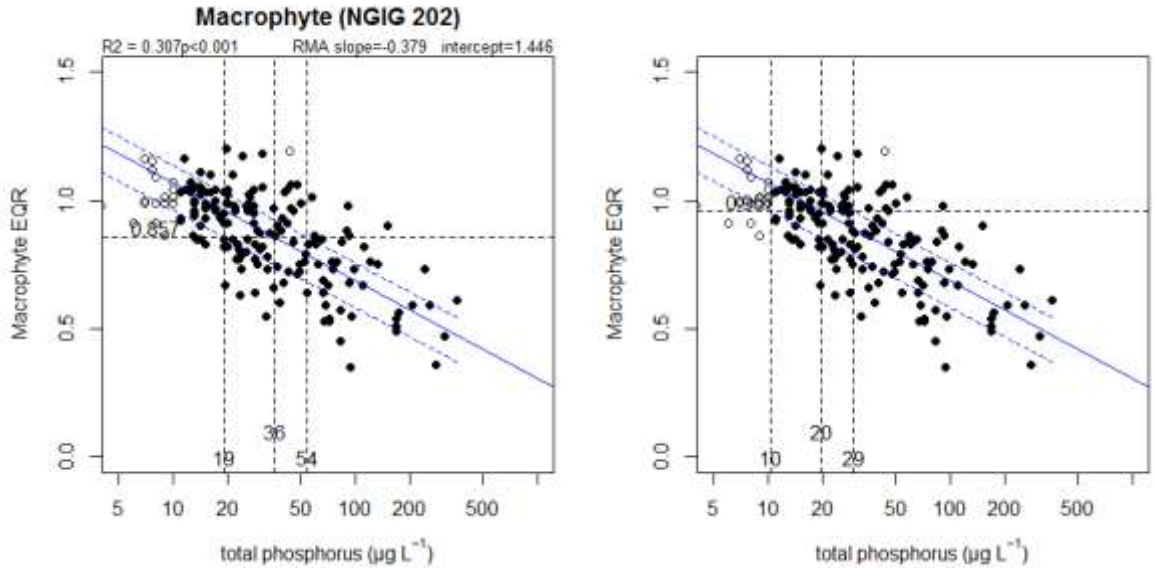


Figure 6-59 Relationship between common metric for macrophytes and total phosphorus for moderate alkalinity humic NGIG lakes (Types 202) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data, open circles data points excluded from regression.

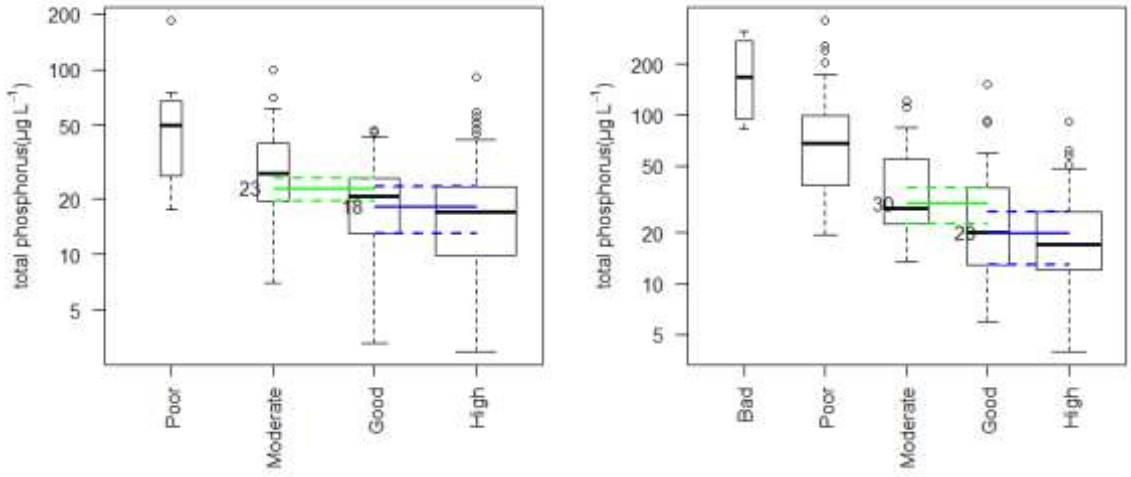


Figure 6-60 Box plots showing range of total phosphorus for a)low alkalinity (Type 102) & b) moderate alkalinity (Type 202) humic NGIG lakes classified using macrophytes common metric showing good/moderate boundary & high/good boundary values determined from the average of the upper and lower quartile values

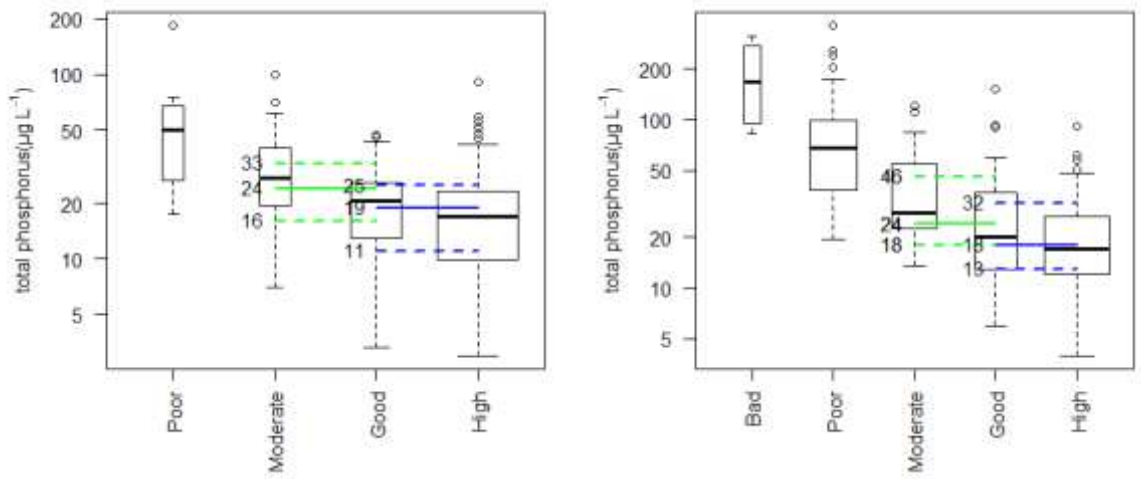


Figure 6-61 Box plots showing range of total phosphorus for a)low alkalinity (Type 102) & b) moderate alkalinity (Type 202) humic NGIG lakes classified using macrophyte common metric showing good/moderate boundary & high/good boundary values determined from the average of adjacent classes

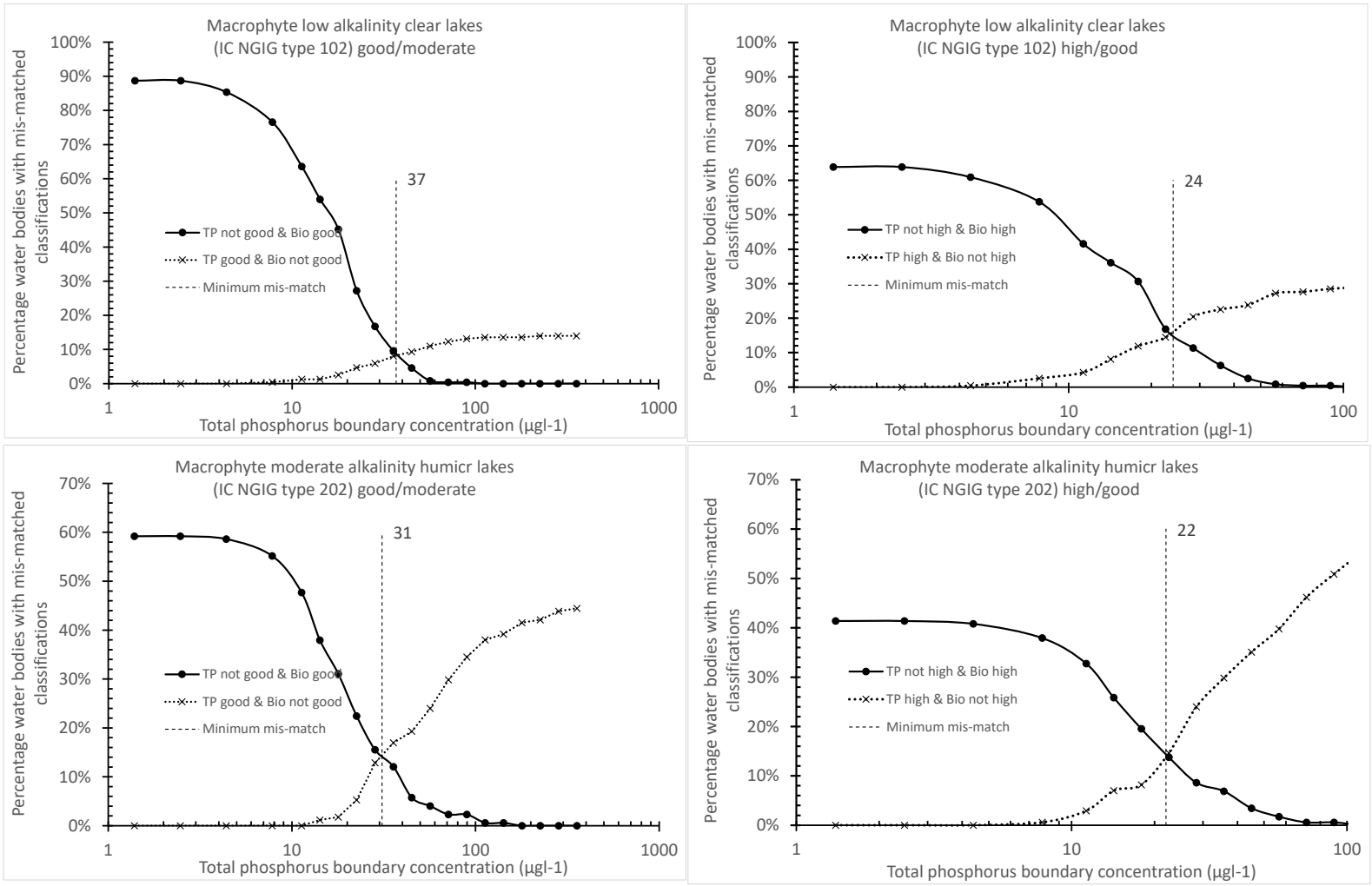


Figure 6-62 Percentage of water bodies where biology or total phosphorus / total nitrogen classifications for good ecological status differ in comparison to the level used to set a) the good/moderate and b) the high/ good boundaries. Biological status assessed using the common metric for macrophytes in low/moderate alkalinity humic NGIG lakes (Types 102 202) Vertical lines mark intersection of curves where mis-match is minimised and equal.

Table 6-41 Regression parameters for type 202 lake macrophytes v total phosphorus

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x =
"interval", nperm = 99)

n = 162   r = -0.5538745   r-square = 0.306777
Parametric P-values:   2-tailed = 2.088171e-14    1-tailed = 1.044086e-14
Angle between the two OLS regression lines = 28.52303 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS  1.357555 -0.3215866       -17.82710              0.01
2     MA  1.486601 -0.4053989       -22.06758              0.01
3    SMA  1.756378 -0.5806128       -30.14000                NA
4    RMA  1.445911 -0.3789722       -20.75532              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS       1.238462        1.476647 -0.3970626  -0.2461107
2     MA       1.344340        1.638643 -0.5041470  -0.3130042
3    SMA       1.647689        1.880110 -0.6609738  -0.5100220
4    RMA       1.311549        1.586203 -0.4700883  -0.2917069

Eigenvalues: 0.1354107 0.02476607

H statistic used for computing C.I. of MA: 0.006677601
```

6.4 Low alkalinity lowland rivers (R-C1)

6.4.1 Macrophytes: Low alkalinity lowland rivers (R-C1)

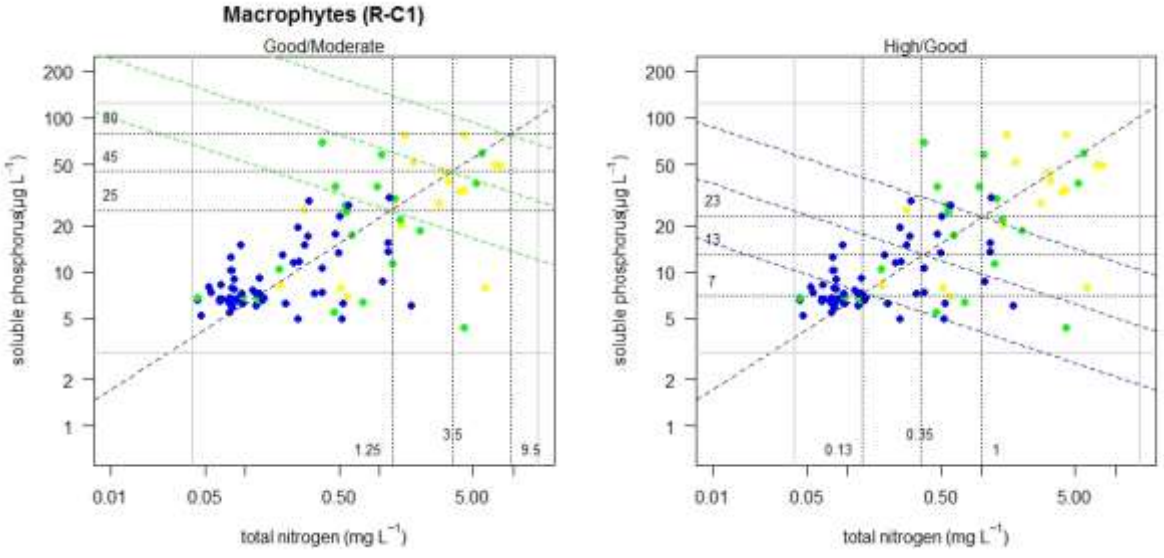


Figure 6-63 Relationship between mean ortho-P and TN, points coloured by WFD class for macrophytes in low alkalinity lowland rivers (Type R-C1). Dotted lines contours of predicted TN & ortho-P concentration when national macrophyte metric EQR is at a) good/moderate boundary (green lines) and b)

high good boundary, dotted lines show ± 25th & 75th residuals of prediction. Horizontal & vertical lines show intersection with RMA regression of observed ortho-P and TN showing boundary values.

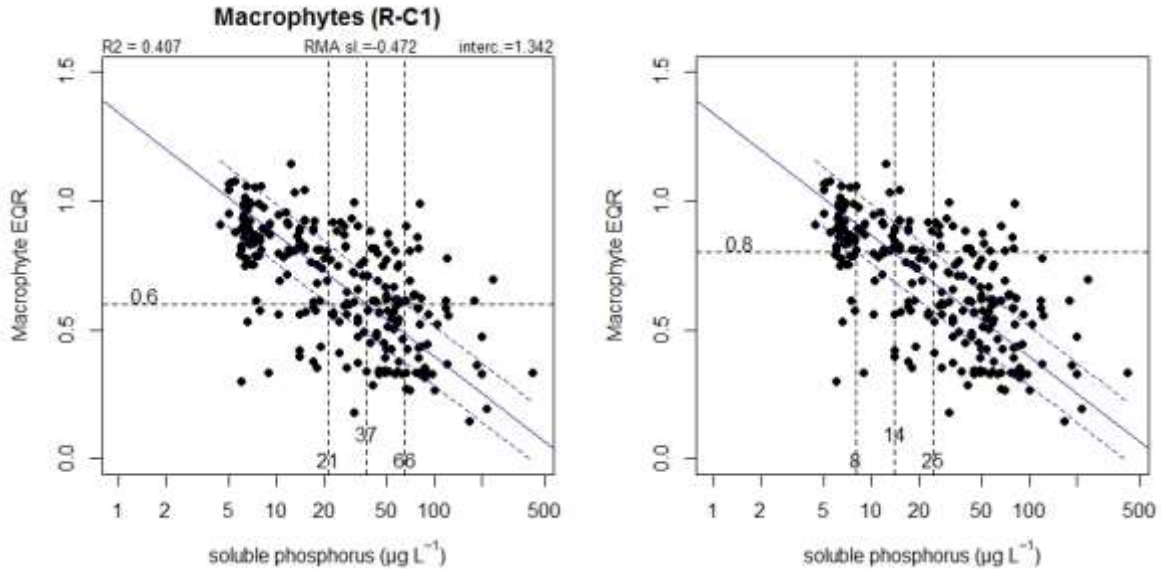


Figure 6-64 Relationship between common metric for macrophytes and ortho-P for low alkalinity lowland rivers (Type R-C1) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data.

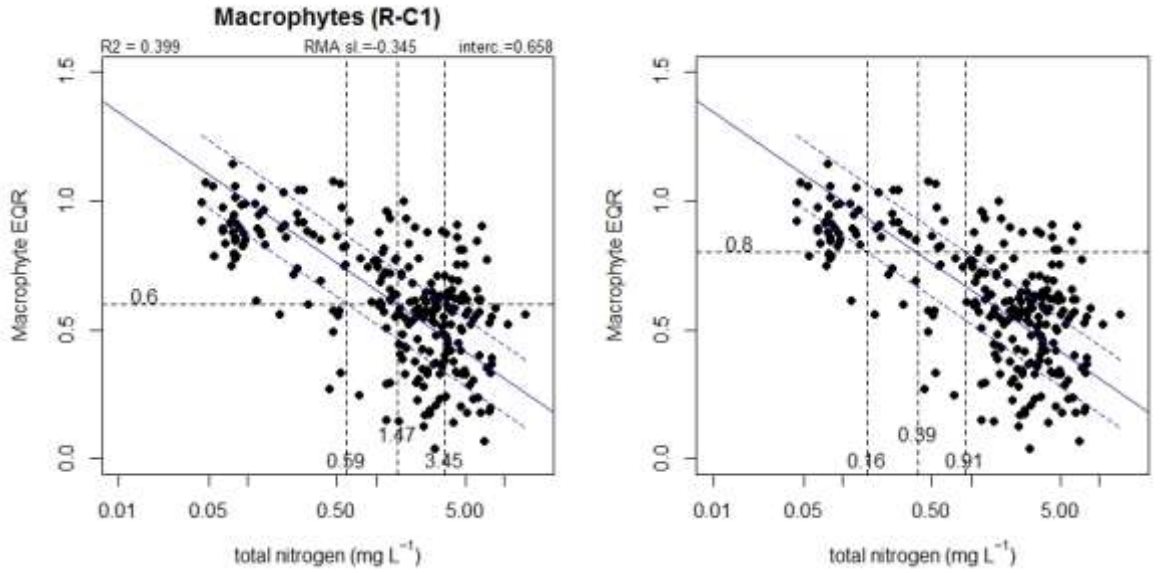


Figure 6-65 Relationship between common metric for macrophytes and total nitrogen for low alkalinity lowland rivers (Type R-C1) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data.

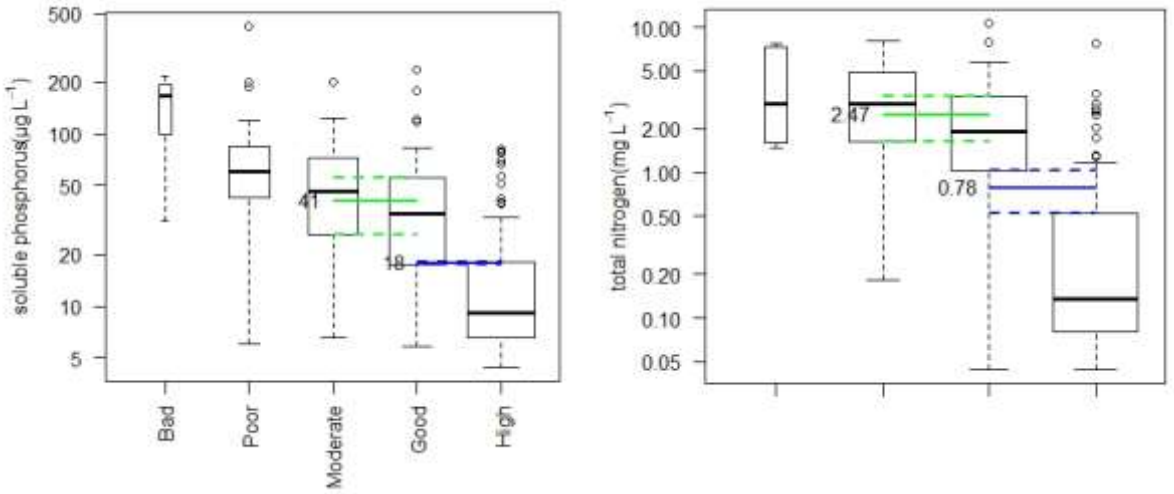


Figure 6-66 Box plots showing range of a) ortho-P and b) TN for low alkalinity lowland rivers (Type R-C1) classified using national macrophyte metrics showing good/moderate boundary & high/good boundary values determined from the average of the upper and lower quartile values

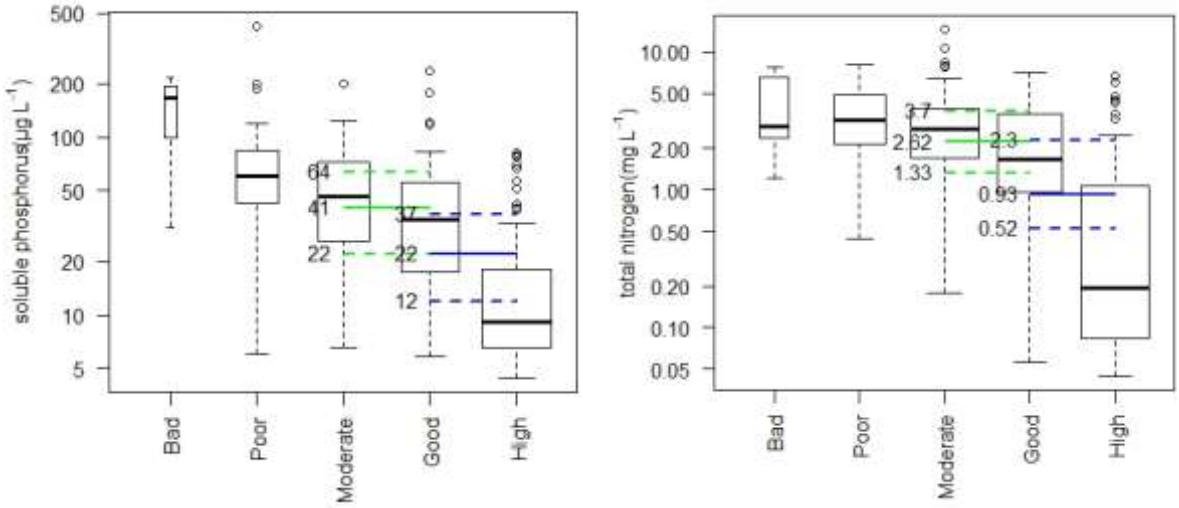


Figure 6-67 Box plots showing range of a) ortho-P and b) TN for low alkalinity lowland rivers (Type R-C1) classified using national macrophyte metrics showing good/moderate boundary & high/good boundary values determined from the average of adjacent classes.

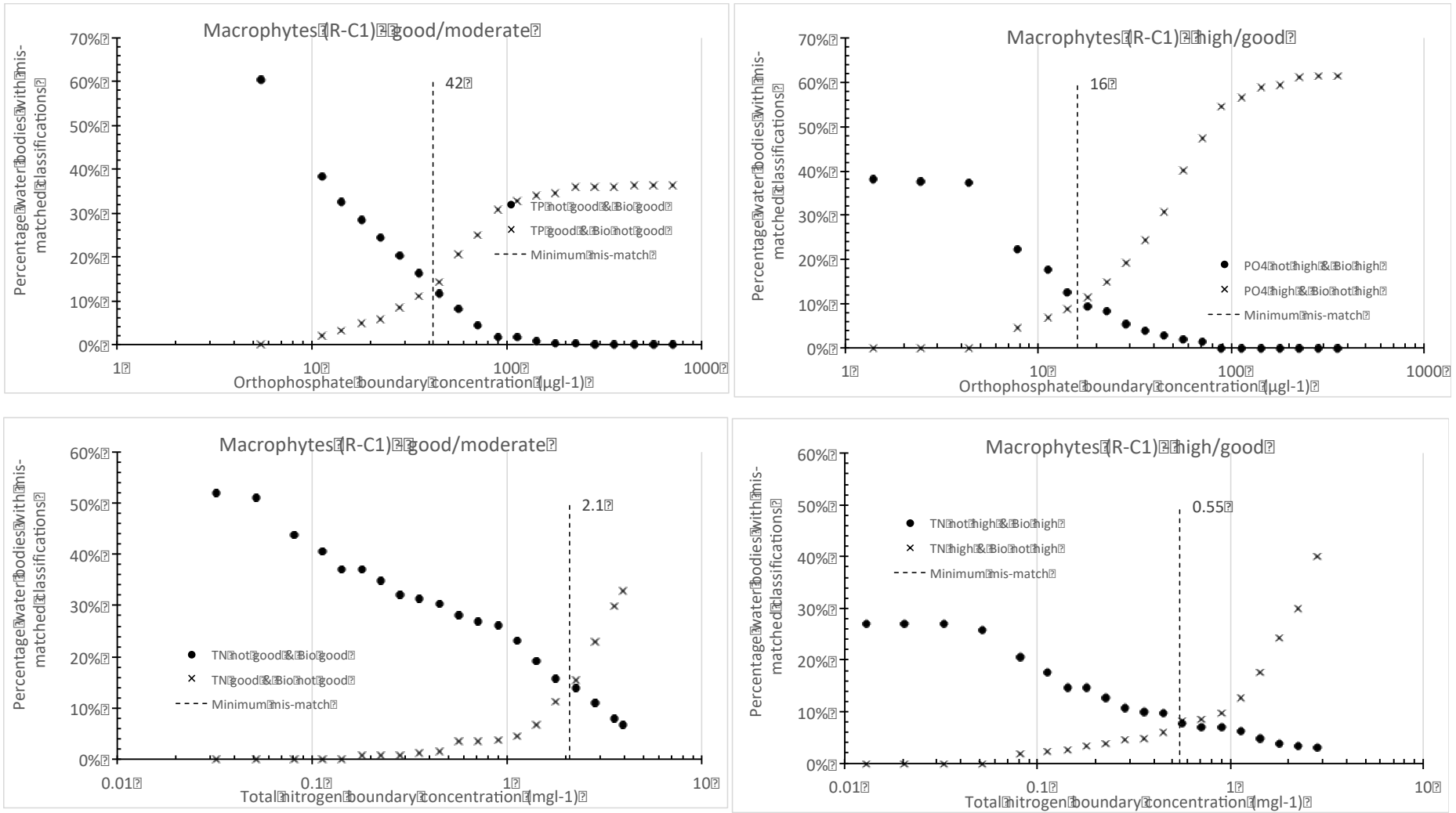


Figure 6-68 Percentage of water bodies where biology or ortho-phosphorus / total nitrogen classifications for good ecological status differ in comparison to the level used to set a) the good/moderate and b) the high/good boundaries. Biological status assessed using national macrophyte metrics in low alkalinity lowland rivers (Type R-C1). Vertical lines mark intersection of curves where mis-match is minimised and equal.

Table 6-42 Regression parameters for R-C1 macrophytes v ortho-phosphorus and total nitrogen

```
Call:
lm(formula = nMP.EQR ~ log10(PO4.P) + log10(total.N), data = data.cc.ex,
    subset = PO4.P > P.minUsed & PO4.P <= P.maxUsed & total.N >
        N.minUsed & total.N <= N.maxUsed)

Residuals:
     Min       1Q   Median       3Q      Max
-0.43757 -0.09031  0.01935  0.09388  0.36620

Coefficients:
              Estimate Std. Error t value Pr(>|t|)
(Intercept)    1.04350    0.06927  15.064  < 2e-16 ***
log10(PO4.P)  -0.24557    0.05126  -4.791 4.49e-06 ***
log10(total.N) -0.07124    0.02880  -2.473   0.0147 *
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 0.1482 on 129 degrees of freedom
Multiple R-squared:  0.4839,     Adjusted R-squared:  0.4759
F-statistic: 60.48 on 2 and 129 DF,  p-value: < 2.2e-16
```

Table 6-43 Regression parameters for R-C1 macrophytes v ortho-phosphorus

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x = "interval",
nperm = 99)

n = 247   r = -0.6379236   r-square = 0.4069466
Parametric P-values:   2-tailed = 1.269438e-29    1-tailed = 6.347192e-30
Angle between the two OLS regression lines = 20.55958 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS  1.134251 -0.3235936       -17.93125              0.01
2     MA  1.205636 -0.3745745       -20.53467              0.01
3    SMA  1.391426 -0.5072607       -26.89689                NA
4    RMA  1.341737 -0.4717743       -25.25674              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS       1.062199        1.206304 -0.3727515  -0.2744356
2     MA       1.127326        1.286933 -0.4326347  -0.3186476
3    SMA       1.325922        1.463586 -0.5587950  -0.4604791
4    RMA       1.246893        1.448907 -0.5483116  -0.4040389

Eigenvalues: 0.2113705 0.02565823

H statistic used for computing C.I. of MA: 0.002490126
```

Table 6-44 Regression parameters for R-C1 macrophytes v total nitrogen

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x = "interval",
nperm = 99)

n = 263   r = -0.6314095   r-square = 0.3986779
Parametric P-values:   2-tailed = 1.158559e-30    1-tailed = 5.792794e-31
Angle between the two OLS regression lines = 17.58203 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS 0.6468926 -0.2405904       -13.52771              0.01
2     MA 0.6491151 -0.2621171       -14.68778              0.01
3    SMA 0.6613925 -0.3810370       -20.85869                NA
4    RMA 0.6576442 -0.3447313       -19.02068              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS      0.6240122       0.6697730 -0.2766040  -0.2045768
2     MA      0.6451002       0.6532080 -0.3017613  -0.2232292
3    SMA      0.6578497       0.6652859 -0.4187488  -0.3467215
4    RMA      0.6525808       0.6632959 -0.3994734  -0.2956866

Eigenvalues: 0.419357 0.0323972

H statistic used for computing C.I. of MA: 0.001347884
```

6.4.2 Phytobenthos: low alkalinity lowland rivers (R-C1)

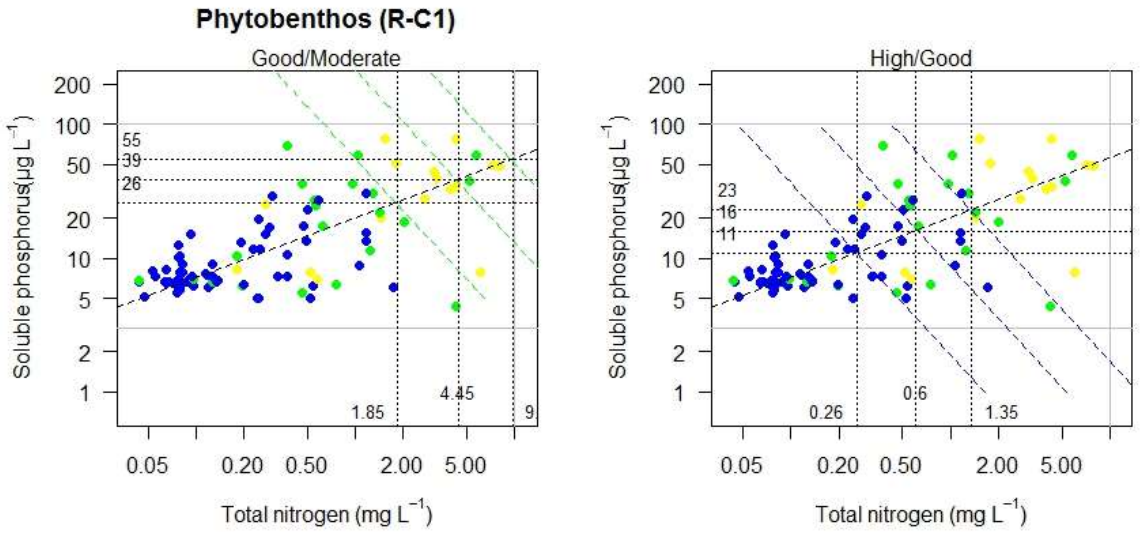


Figure 6-69 Relationship between mean ortho-P and TN, points coloured by WFD class for phytobenthos in low alkalinity lowland rivers (Type R-C1). Dotted lines contours of predicted TN & ortho-P concentration when national phytobenthos metric EQR is at a) good/moderate boundary (green lines) and b) high good boundary, dotted lines show ± 25th & 75th residuals of prediction. Horizontal & vertical lines show intersection with RMA regression of observed ortho-P and TN showing boundary values.

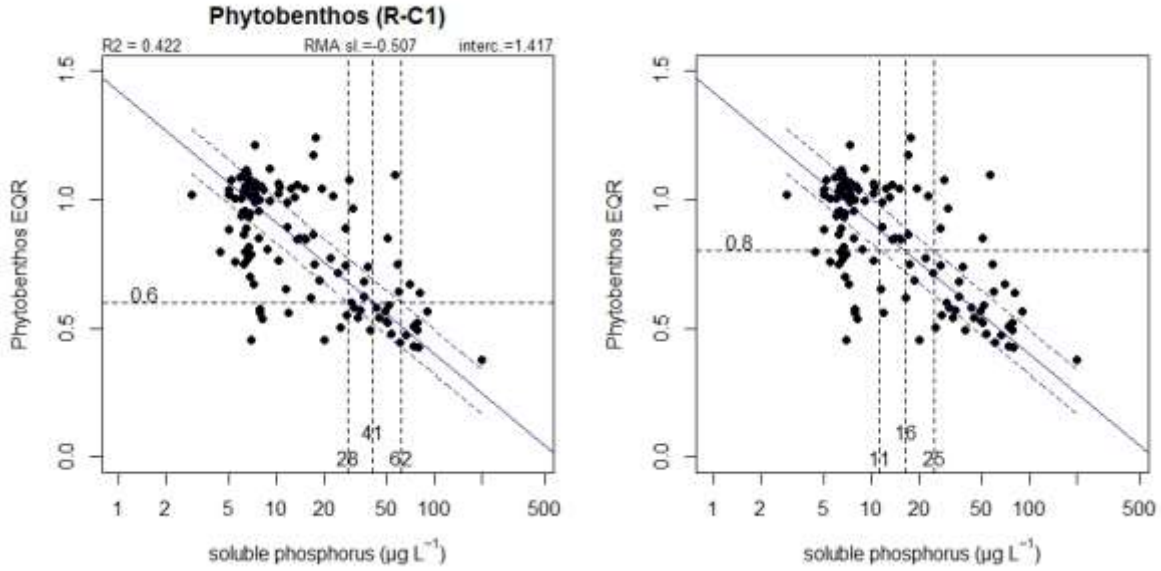


Figure 6-70 Relationship between EQR (as national metric) for phytobenthos and ortho-P for low alkalinity lowland rivers (Type R-C1) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data.

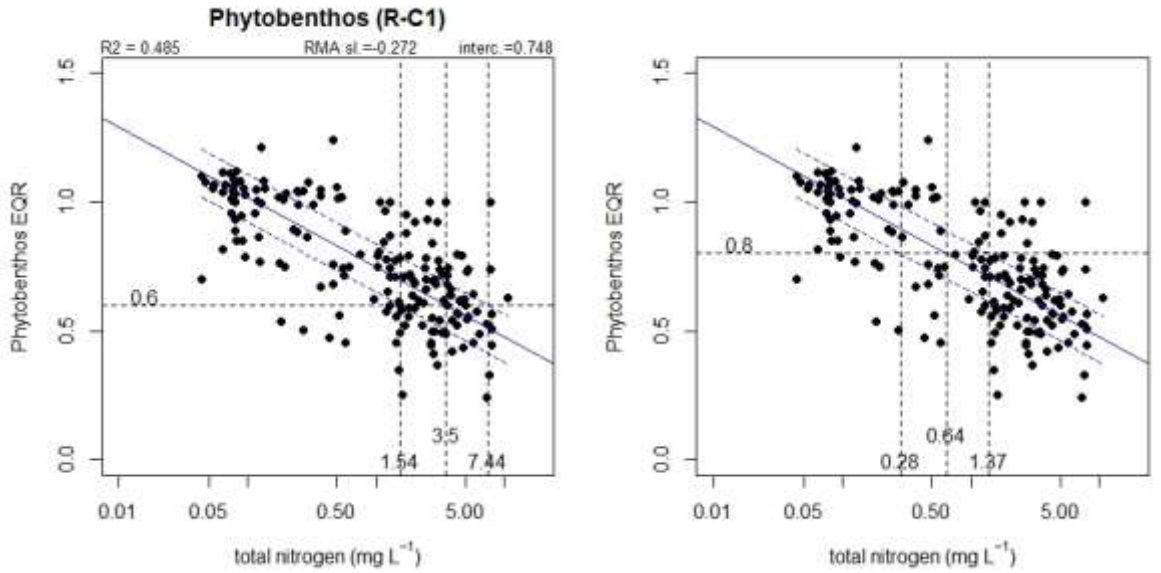


Figure 6-71 Relationship between EQR (as national metric) for phytobenthos and total nitrogen for low alkalinity lowland rivers (Type R-C1) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data.

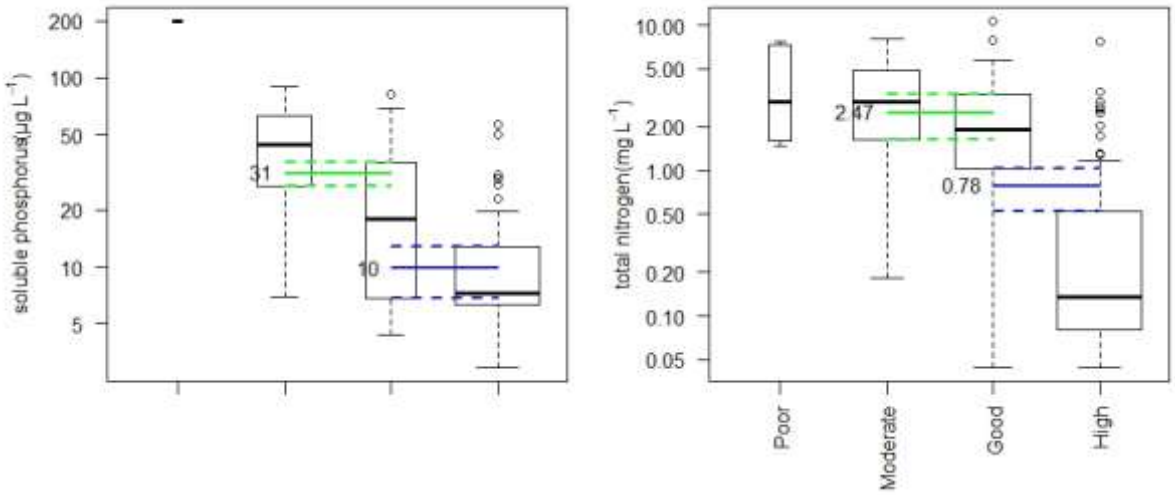


Figure 6-72 Box plots showing range of a) ortho-P and b) TN for low alkalinity lowland rivers (Type R-C1) classified using national phytobenthos metrics showing good/moderate boundary & high/good boundary values determined from the average of the upper and lower quartile values

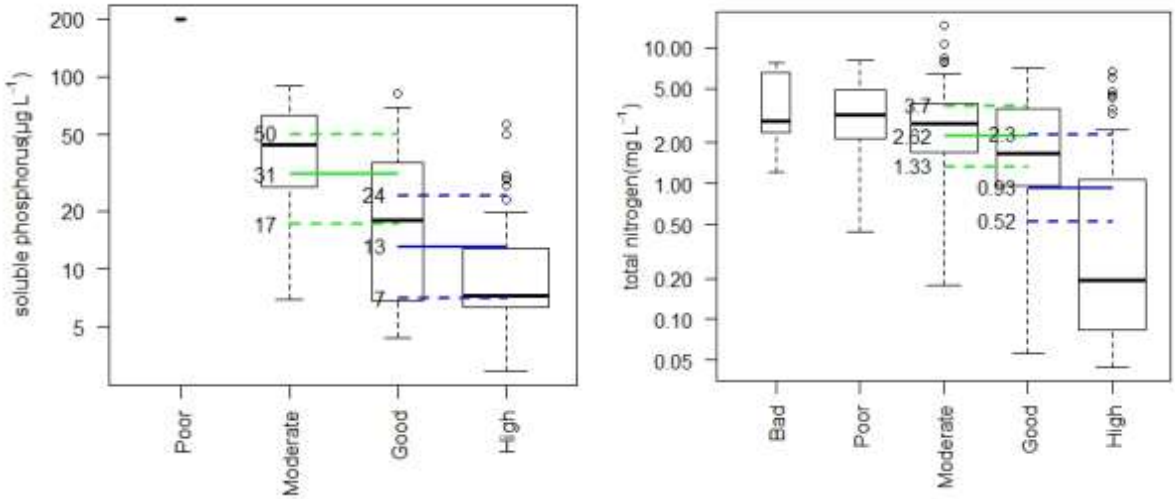


Figure 6-73 Box plots showing range of a) ortho-P and b) TN for low alkalinity lowland rivers (Type R-C1) classified using national phytobenthos metrics showing good/moderate boundary & high/good boundary values determined from the average of adjacent classes.

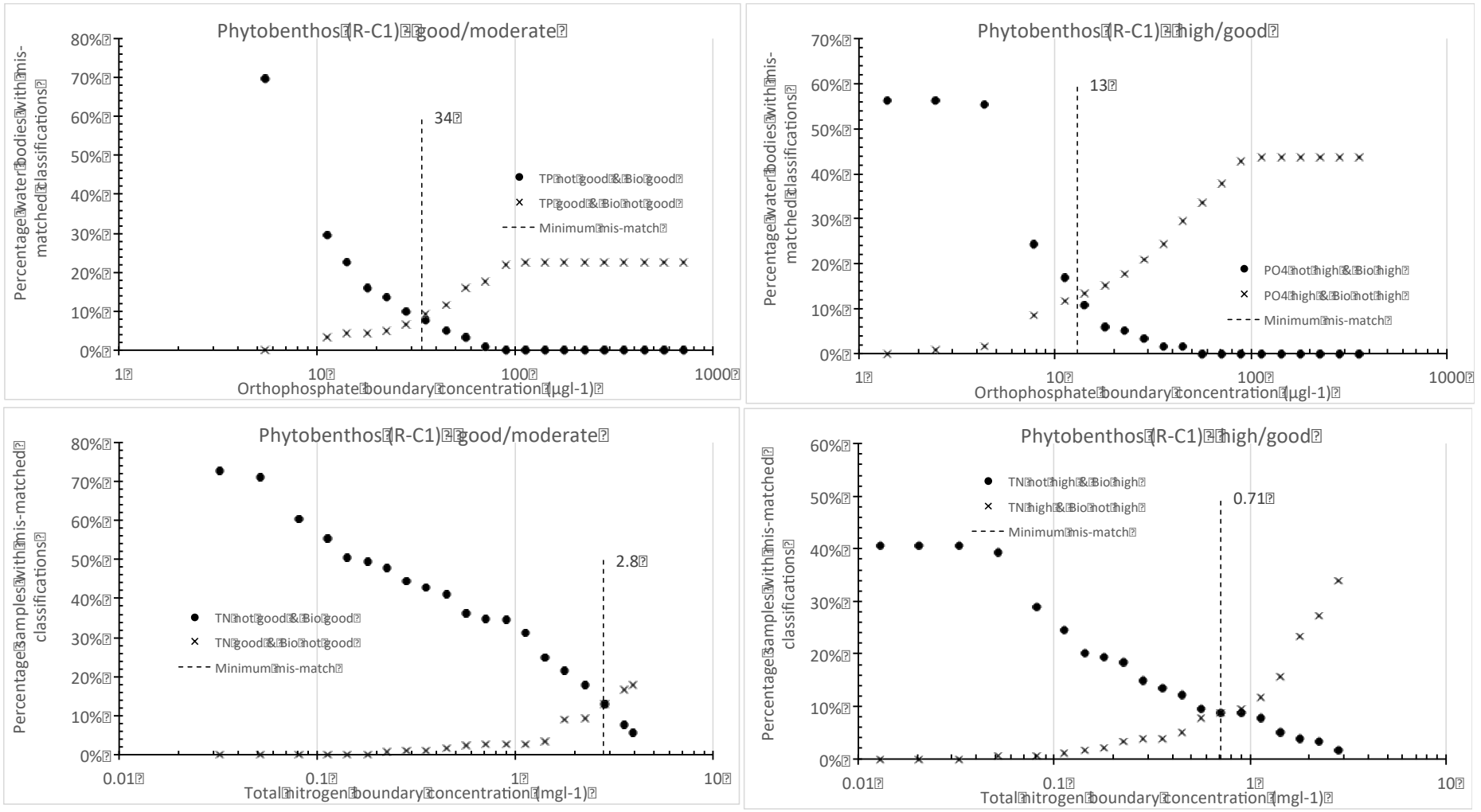


Figure 6-74 Percentage of water bodies where biology or ortho-phosphorus / total nitrogen classifications for good ecological status differ in comparison to the level used to set a) the good/moderate and b) the high/good boundaries. Biological status assessed using national phytobenthos metrics in low alkalinity lowland rivers (Type R-C1). Vertical lines mark intersection of curves where mis-match is minimised and equal.

Table 6-45 Regression parameters for R-C1 phytobenthos v ortho-phosphorus and total nitrogen

```
Call:
lm(formula = nPB.EQR ~ log10(PO4.P) + log10(total.N), data = data.cc.ex,
    subset = PO4.P > P.minUsed & PO4.P <= P.maxUsed & total.N >
        N.minUsed & total.N <= N.maxUsed)

Residuals:
     Min       1Q   Median       3Q      Max
-0.40184 -0.08021  0.02096  0.08981  0.42465

Coefficients:
              Estimate Std. Error t value Pr(>|t|)
(Intercept)    0.92741    0.07783  11.915  < 2e-16 ***
log10(PO4.P)  -0.13622    0.05992  -2.273   0.0253 *
log10(total.N) -0.17553    0.03237  -5.422 4.46e-07 ***
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 0.1454 on 95 degrees of freedom
Multiple R-squared:  0.4988,     Adjusted R-squared:  0.4883
F-statistic: 47.28 on 2 and 95 DF,  p-value: 5.613e-15
```

Table 6-46 Regression parameters for R-C1 phytobenthos v ortho-phosphorus

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x = "interval",
nperm = 99)

n = 120   r = -0.6497954   r-square = 0.422234
Parametric P-values:   2-tailed = 9.791439e-16    1-tailed = 4.895719e-16
Angle between the two OLS regression lines = 20.75786 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS  1.251422 -0.3638300       -19.99290              0.01
2     MA  1.329644 -0.4313649       -23.33367              0.01
3    SMA  1.478537 -0.5599147       -29.24511                NA
4    RMA  1.417377 -0.5071110       -26.89007              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS       1.156463        1.346382 -0.4414157  -0.2862443
2     MA       1.226248        1.440221 -0.5268337  -0.3420952
3    SMA       1.394870        1.574597 -0.6428503  -0.4876788
4    RMA       1.300491        1.553831 -0.6249219  -0.4061946

Eigenvalues: 0.1825412 0.02470201

H statistic used for computing C.I. of MA: 0.00601494
```

Table 6-47 Regression parameters for R-C1 phytobenthos v total nitrogen

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x = "interval",
nperm = 99)

n = 179   r = -0.6965703   r-square = 0.4852102
Parametric P-values:   2-tailed = 2.576818e-27    1-tailed = 1.288409e-27
Angle between the two OLS regression lines = 12.27876 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
```

```
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS 0.7510915 -0.2269095       -12.78448              0.01
2     MA 0.7501652 -0.2393089       -13.45829              0.01
3    SMA 0.7437078 -0.3257525       -18.04315                NA
4    RMA 0.7477527 -0.2716042       -15.19521              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS      0.7276021       0.7745808 -0.2615787  -0.1922403
2     MA      0.7474094       0.7528757 -0.2761999  -0.2030238
3    SMA      0.7409806       0.7461602 -0.3622614  -0.2929230
4    RMA      0.7445413       0.7507601 -0.3145951  -0.2313457

Eigenvalues: 0.4807731 0.0236274

H statistic used for computing C.I. of MA: 0.001195994
```

6.4.3 Combined macrophyte and phytobenthos (CMP) low alkalinity lowland rivers (R-C1)

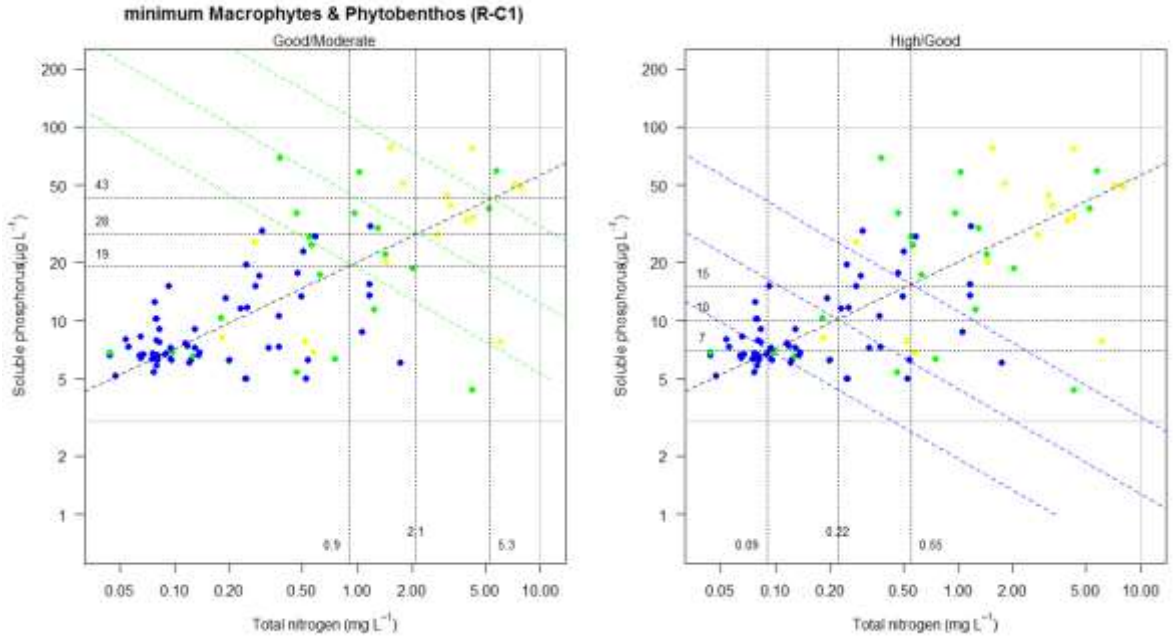


Figure 6-75 Relationship between mean ortho-P and TN, points coloured by WFD class for CMP in low alkalinity lowland rivers (Type R-C1). Dotted lines contours of predicted TN & ortho-P concentration when CMP is at a) good/moderate boundary (green lines) and b) high good boundary, dotted lines show ± 25th & 75th residuals of prediction. Horizontal & vertical lines show intersection with RMA regression of observed ortho-P and TN showing boundary values.

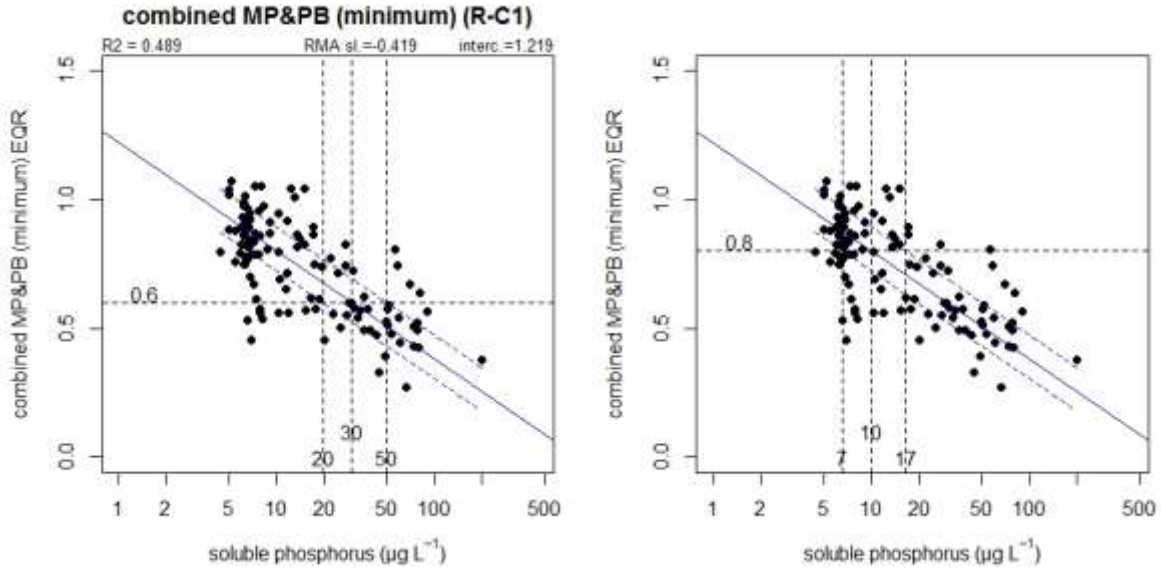


Figure 6-76 Relationship between EQR (as national metrics) for CMP and ortho-P for low alkalinity lowland rivers (Type R-C1) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data.

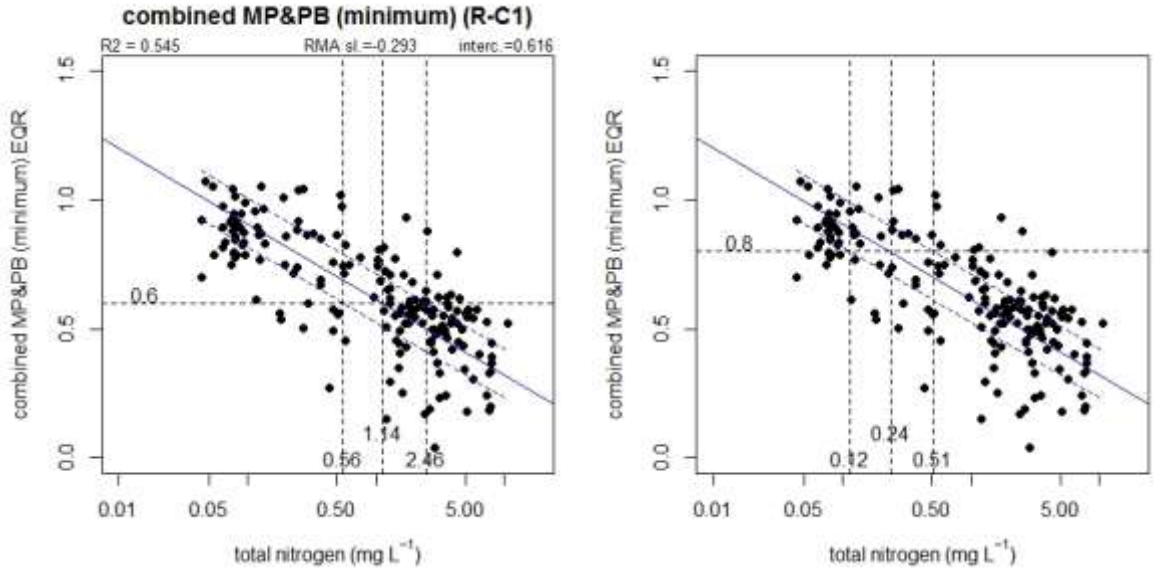


Figure 6-77 Relationship between EQR (as national metrics) for CMP and total nitrogen for low alkalinity lowland rivers (Type R-C1) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data.

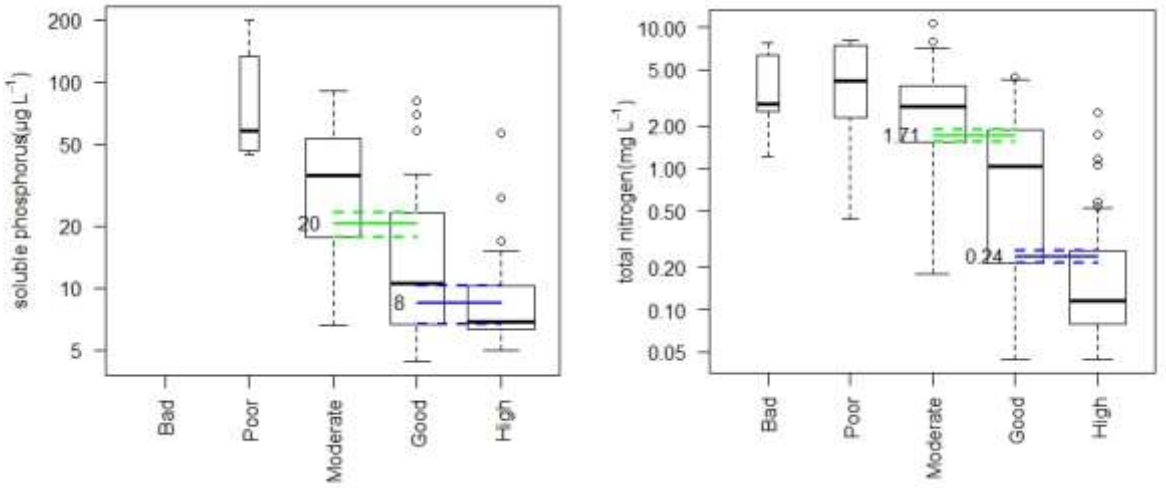


Figure 6-78 Box plots showing range of a) ortho-P and b) TN for low alkalinity lowland rivers (Type R-C1) classified using CMP showing good/moderate boundary & high/good boundary values determined from the average of the upper and lower quartile values.

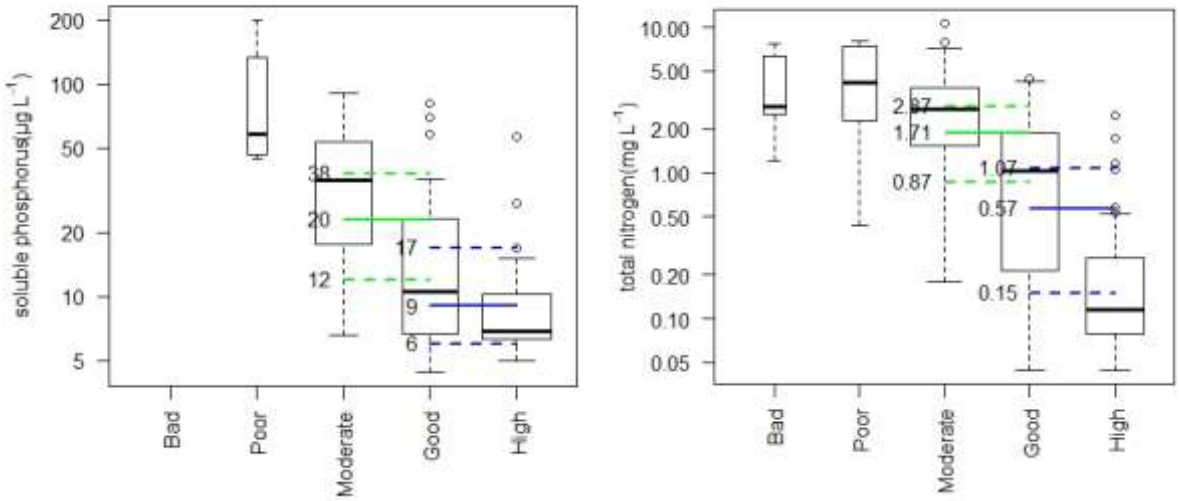


Figure 6-79 Box plots showing range of a) ortho-P and b) TN for low alkalinity lowland rivers (Type R-C1) classified using CMP showing good/moderate boundary & high/good boundary values determined from the average of adjacent classes.

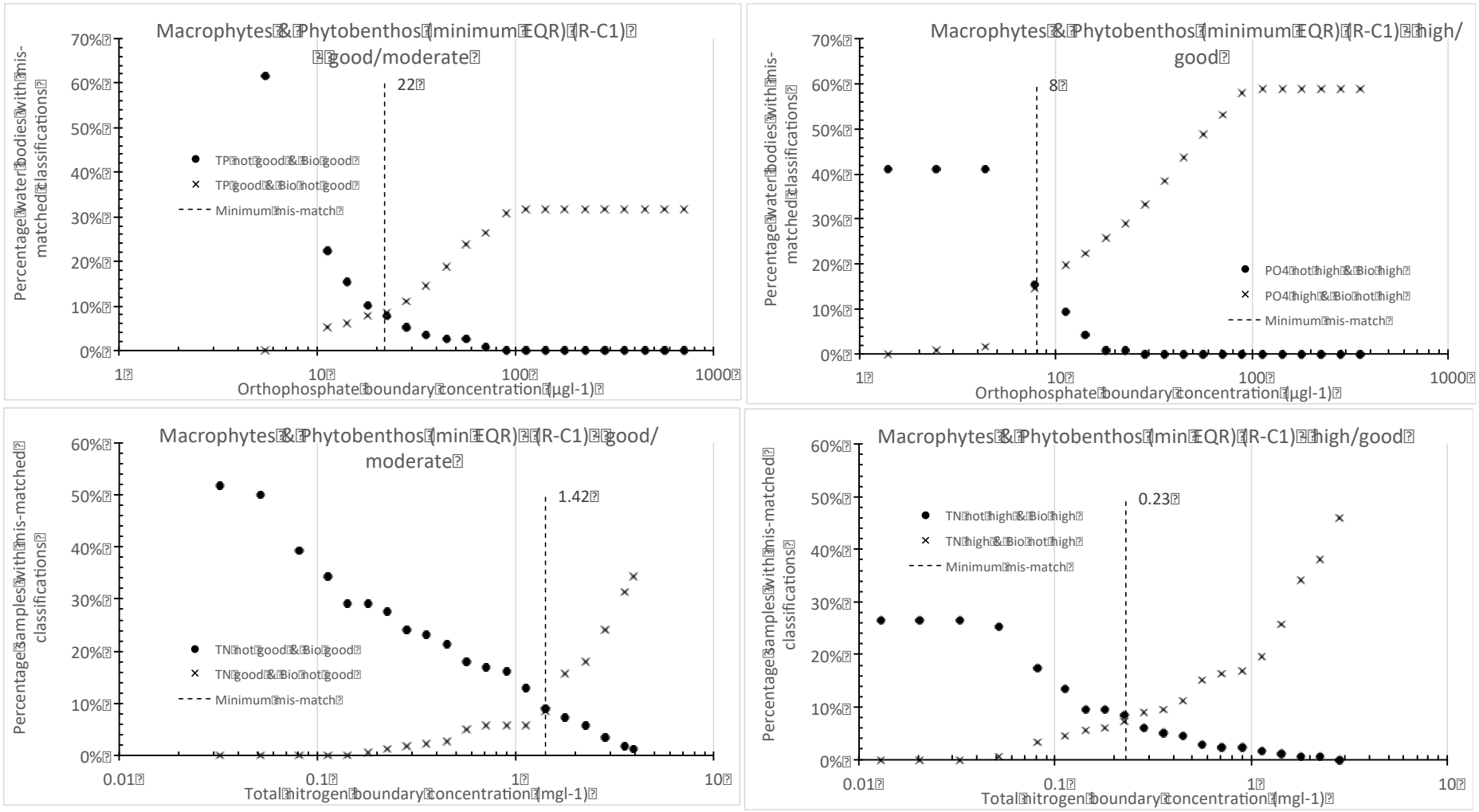


Figure 6-80 Percentage of water bodies where biology or ortho-phosphorus / total nitrogen classifications for good ecological status differ in comparison to the level used to set a) the good/moderate and b) the high/good boundaries. Biological status assessed using CMP metrics in low alkalinity lowland rivers (Type R-C1). Vertical lines mark intersection of curves where mis-match is minimised and equal.

Table 6-48 Regression parameters for R-C1 CMP v ortho-phosphorus and total nitrogen

```
Call:
lm(formula = nEQR.min ~ log10(PO4.P) + log10(total.N), data = data.cc.ex,
    subset = PO4.P > P.minUsed & PO4.P <= P.maxUsed & total.N >
        N.minUsed & total.N <= N.maxUsed)

Residuals:
     Min       1Q   Median       3Q      Max
-0.33432 -0.08101  0.00750  0.07333  0.28733

Coefficients:
              Estimate Std. Error t value Pr(>|t|)
(Intercept)    0.93164    0.06579   14.16  < 2e-16 ***
log10(PO4.P)  -0.20311    0.05065   -4.01 0.000121 ***
log10(total.N) -0.11028    0.02736   -4.03 0.000112 ***
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 0.1229 on 95 degrees of freedom
Multiple R-squared:  0.5128,     Adjusted R-squared:  0.5025
F-statistic: 49.99 on 2 and 95 DF,  p-value: 1.47e-15
```

Table 6-49 Regression parameters for R-C1 CMP v ortho-phosphorus

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x = "interval",
nperm = 99)

n = 119   r = -0.6990799   r-square = 0.4887127
Parametric P-values:   2-tailed = 9.452707e-19    1-tailed = 4.726353e-19
Angle between the two OLS regression lines = 15.81553 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS  1.118022 -0.3317216       -18.35180              0.01
2     MA  1.162146 -0.3696262       -20.28563              0.01
3    SMA  1.284241 -0.4745118       -25.38489                NA
4    RMA  1.219211 -0.4186482       -22.71654              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS       1.041715        1.194328 -0.3938443  -0.2695990
2     MA       1.083198        1.244737 -0.4405762  -0.3018058
3    SMA       1.216639        1.361270 -0.5406837  -0.4164384
4    RMA       1.132447        1.316221 -0.5019850  -0.3441139

Eigenvalues: 0.1740088 0.01589535

H statistic used for computing C.I. of MA: 0.003708895
```

Table 6-50 Regression parameters for R-C1 CMP v total nitrogen

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x = "interval",
nperm = 99)

n = 177   r = -0.7381141   r-square = 0.5448124
Parametric P-values:   2-tailed = 1.003025e-31    1-tailed = 5.015124e-32
Angle between the two OLS regression lines = 10.31116 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS 0.6202835 -0.2409602       -13.54774              0.01
2     MA 0.6194420 -0.2525069       -14.17135              0.01
3    SMA 0.6140528 -0.3264539       -18.07947                NA
4    RMA 0.6164686 -0.2933067       -16.34677              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS      0.5979837       0.6425834 -0.2738197  -0.2081008
2     MA      0.6169105       0.6219325 -0.2872430  -0.2183343
3    SMA      0.6115378       0.6163274 -0.3609629  -0.2952441
4    RMA      0.6134232       0.6192744 -0.3350932  -0.2548065

Eigenvalues: 0.4856879 0.02093574

H statistic used for computing C.I. of MA: 0.001047824
```

6.5 Low alkalinity upland rivers (R-C3)

6.5.1 Macrophytes: low alkalinity upland rivers (R-C3)

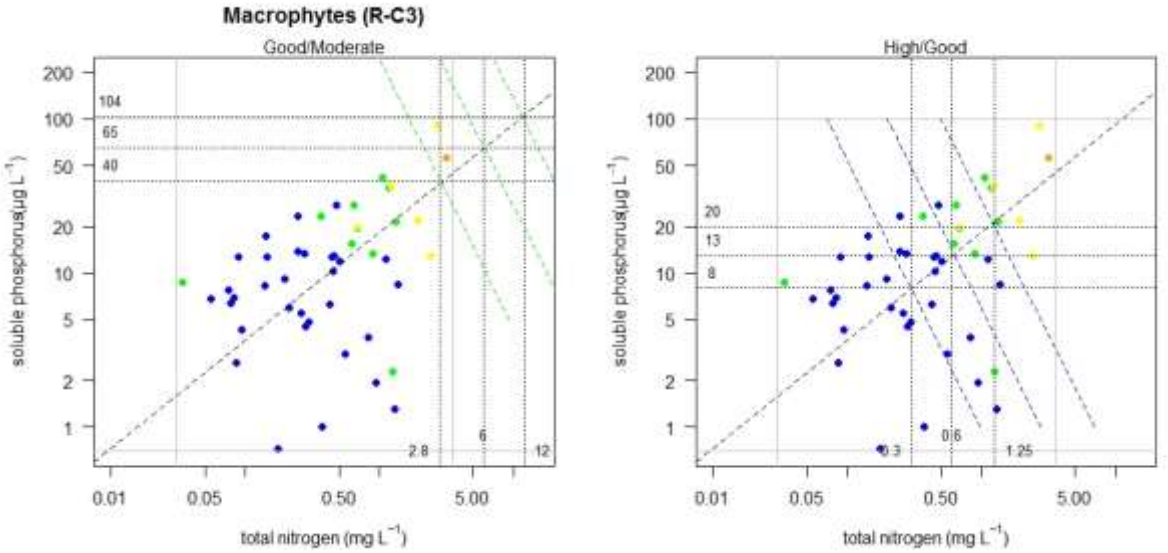


Figure 6-81 Relationship between mean ortho-P and TN, points coloured by WFD class for macrophytes in low alkalinity upland rivers (Type R-C3). Dotted lines contours of predicted TN & ortho-P concentration when national macrophyte metric EQR is at a) good/moderate boundary (green lines) and b) high good boundary, dotted lines show ± 25th & 75th residuals of prediction. Horizontal & vertical lines show intersection with RMA regression of observed ortho-P and TN showing boundary values.

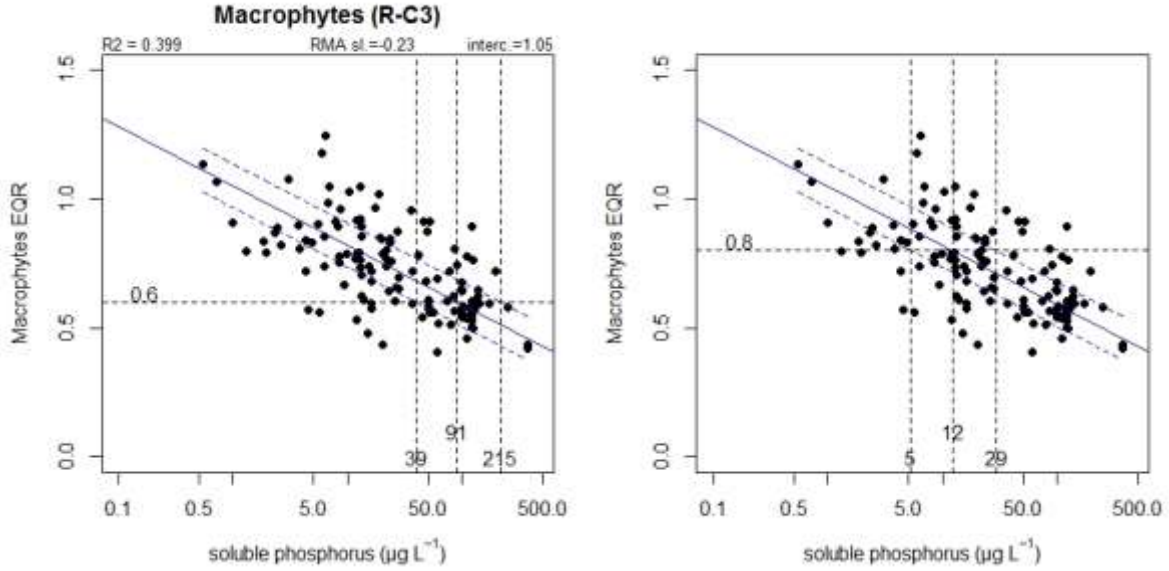


Figure 6-82 Relationship between common metric for macrophytes and ortho-P for low alkalinity upland rivers (Type R-C3) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data.

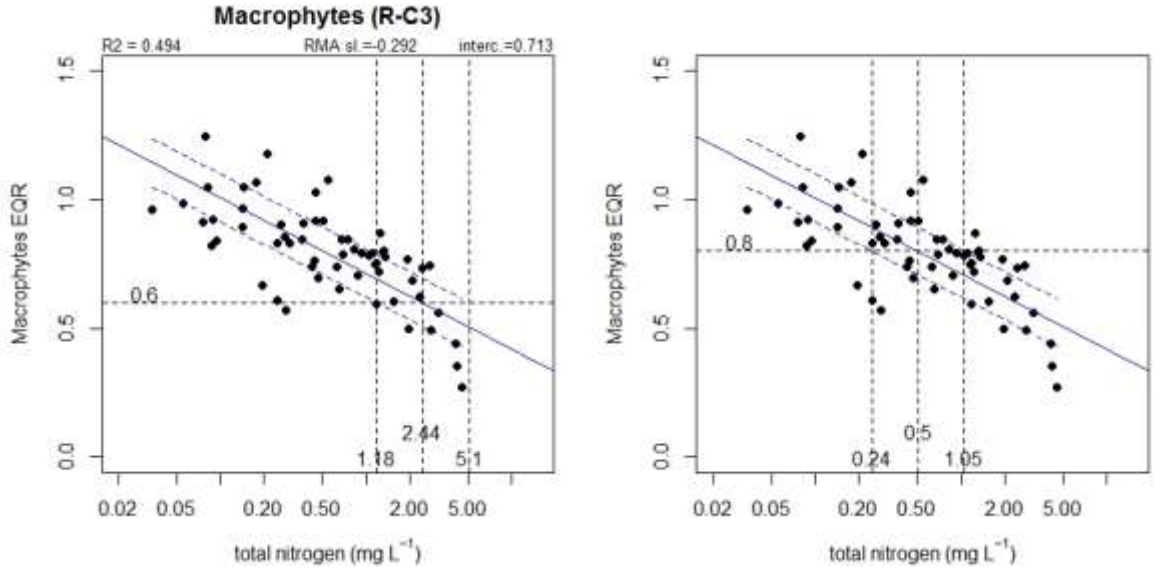


Figure 6-83 Relationship between common metric for macrophytes and total nitrogen for low alkalinity upland rivers (Type R-C3) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data.

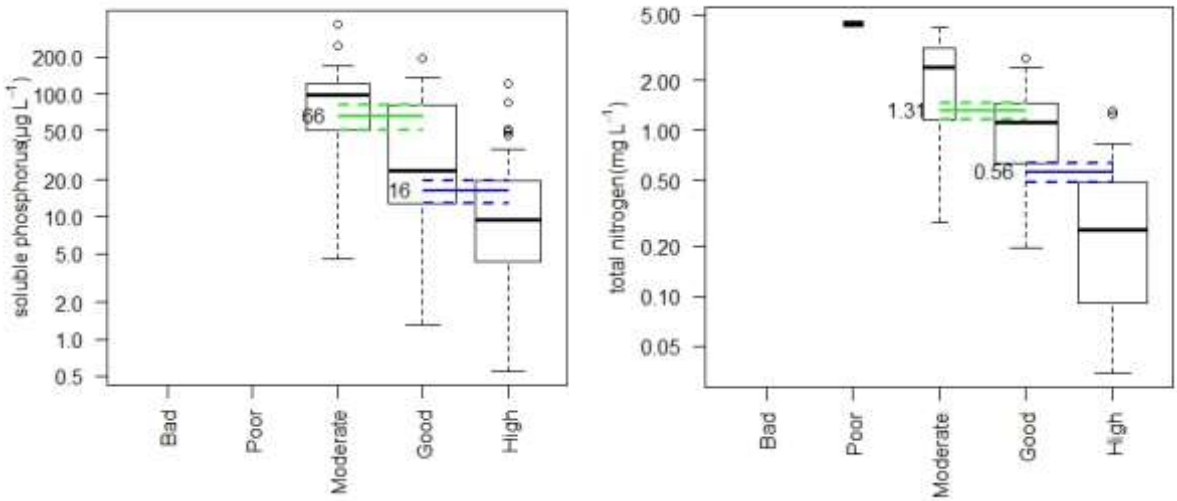


Figure 6-84 Box plots showing range of a) ortho-P and b) TN for low alkalinity upland rivers (Type R-C3) classified using national macrophyte metrics showing good/moderate boundary & high/good boundary values determined from the average of the upper and lower quartile values.

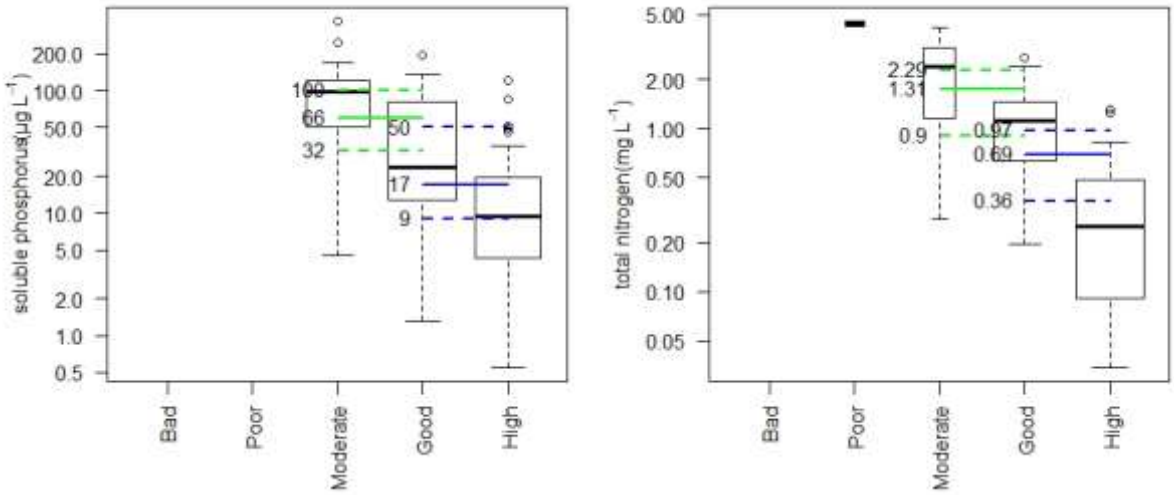


Figure 6-85 Box plots showing range of a) ortho-P and b) TN for low alkalinity upland rivers (Type R-C3) classified using national macrophyte metrics showing good/moderate boundary & high/good boundary values determined from the average of adjacent classes.

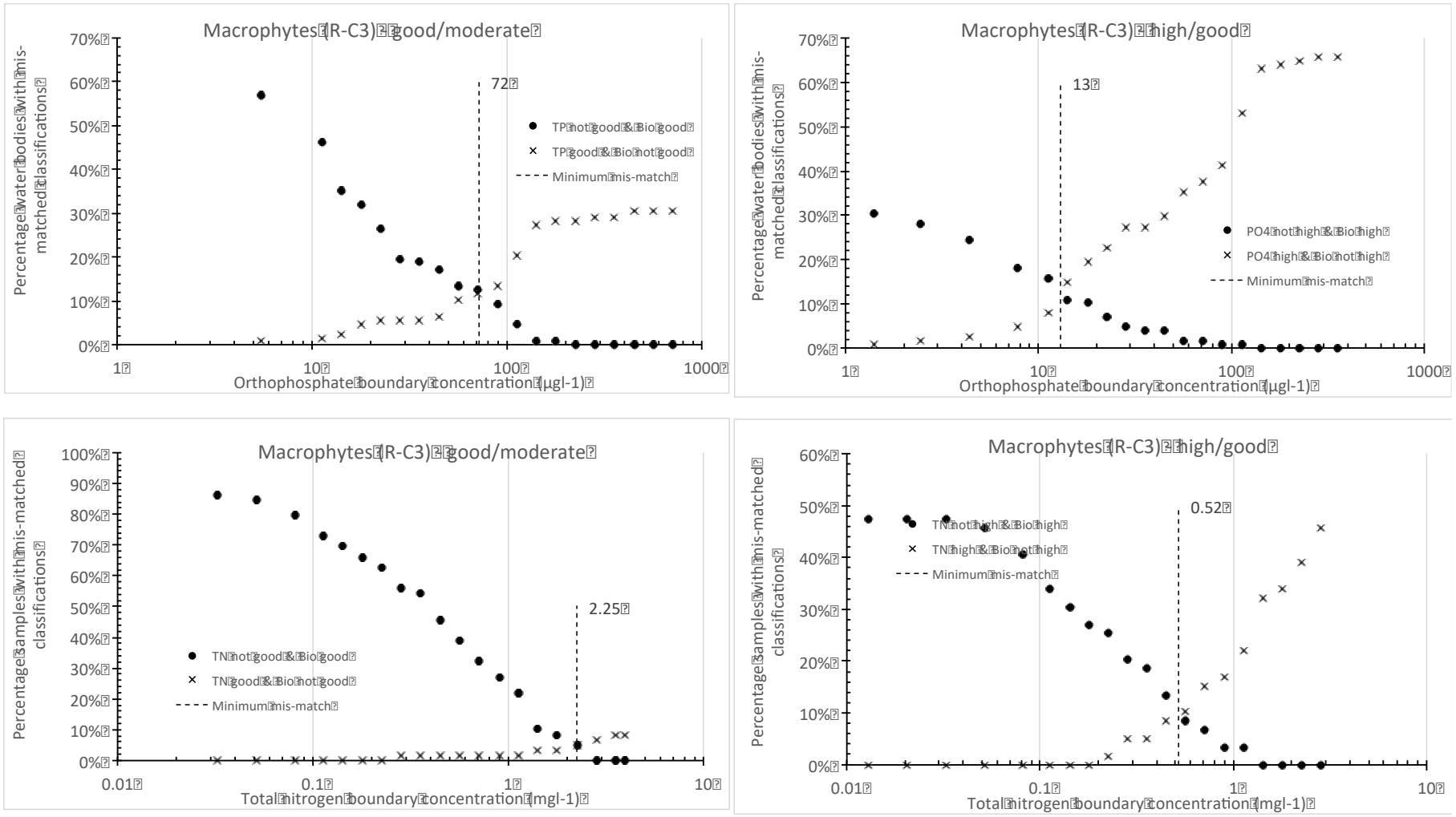


Figure 6-86 Percentage of water bodies where biology or ortho-phosphorus / total nitrogen classifications for good ecological status differ in comparison to the level used to set a) the good/moderate and b) the high/good boundaries. Biological status assessed using national macrophyte metrics in low alkalinity upland rivers (Type R-C3). Vertical lines mark intersection of curves where mis-match is minimised and equal.

Table 6-51 Regression parameters for R-C3 macrophytes v ortho-phosphorus and total nitrogen

```
Call:
lm(formula = nMP.EQR ~ log10(PO4.P) + log10(total.N), data = data.cc.ex,
    subset = PO4.P > P.minUsed & PO4.P <= P.maxUsed & total.N >
        N.minUsed & total.N <= N.maxUsed)

Residuals:
     Min       1Q   Median       3Q      Max
-0.31933 -0.05806  0.00018  0.06535  0.28619

Coefficients:
              Estimate Std. Error t value Pr(>|t|)
(Intercept)    0.86385    0.05294  16.318  < 2e-16 ***
log10(PO4.P)  -0.08296    0.04290  -1.934 0.059448 .
log10(total.N) -0.14511    0.03960  -3.664 0.000652 ***
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 0.1224 on 45 degrees of freedom
Multiple R-squared:  0.3555,     Adjusted R-squared:  0.3269
F-statistic: 12.41 on 2 and 45 DF,  p-value: 5.096e-05
```

Table 6-52 Regression parameters for R-C3 macrophytes v ortho-phosphorus

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x = "interval",
nperm = 99)

n = 128   r = -0.6318899   r-square = 0.3992848
Parametric P-values:   2-tailed = 1.263012e-15    1-tailed = 6.315062e-16
Angle between the two OLS regression lines = 14.14503 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS 0.9818552 -0.1813024       -10.27624              0.01
2     MA 0.9945952 -0.1904043       -10.78032              0.01
3    SMA 1.1296909 -0.2869209       -16.00929                NA
4    RMA 1.0497803 -0.2298302       -12.94352              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS      0.9221908        1.041520 -0.2205082  -0.1420966
2     MA      0.9373512        1.052713 -0.2319252  -0.1495073
3    SMA      1.0785459        1.188300 -0.3287929  -0.2503813
4    RMA      0.9830569        1.123046 -0.2821738  -0.1821610

Eigenvalues: 0.3719764 0.01718818

H statistic used for computing C.I. of MA: 0.001578756
```

Table 6-53 Regression parameters for R-C3 macrophytes v total nitrogen

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x = "interval",
nperm = 99)

n = 58   r = -0.7041294   r-square = 0.4957982
Parametric P-values:   2-tailed = 6.980084e-10    1-tailed = 3.490042e-10
Angle between the two OLS regression lines = 13.0449 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS 0.7245784 -0.2585506       -14.49640              0.01
2     MA 0.7203335 -0.2760678       -15.43311              0.01
3    SMA 0.6982510 -0.3671919       -20.16283                NA
4    RMA 0.7146806 -0.2993948       -16.66742              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS      0.6856890       0.7634679 -0.3283473  -0.1887540
2     MA      0.7018763       0.7380965 -0.3522321  -0.2027677
3    SMA      0.6797437       0.7135718 -0.4435633  -0.3039700
4    RMA      0.6940053       0.7334752 -0.3847123  -0.2218379

Eigenvalues: 0.2744263 0.01625292

H statistic used for computing C.I. of MA: 0.004795263
```

6.5.2 Phytobenthos: Low alkalinity rivers (R-C3)

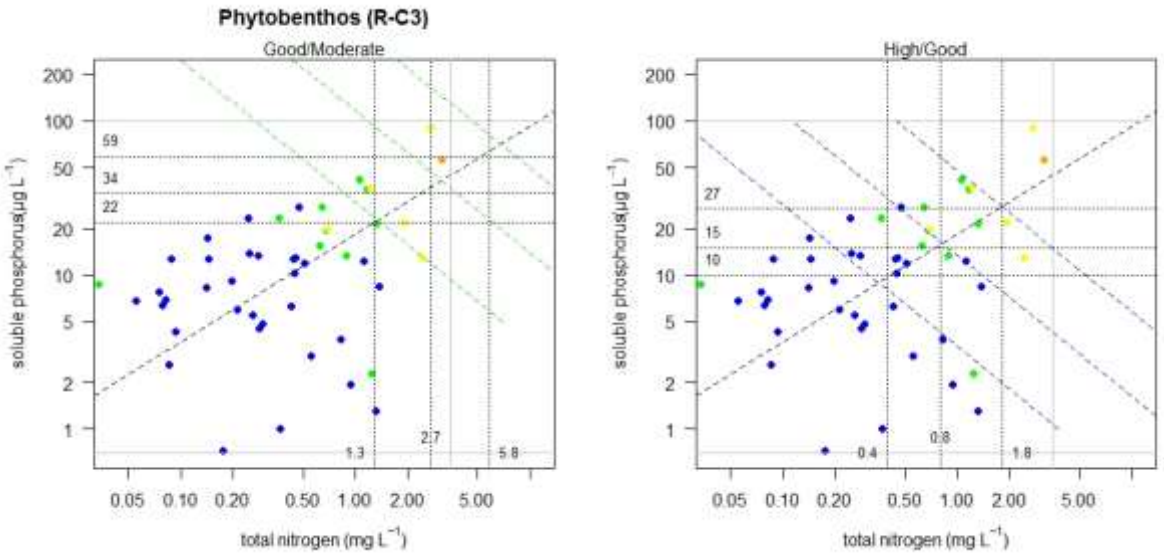


Figure 6-87 Relationship between mean ortho-P and TN, points coloured by WFD class for phytobenthos in low alkalinity upland rivers (Type R-C3). Dotted lines contours of predicted TN & ortho-P concentration when national phytobenthos metric EQR is at a) good/moderate boundary (green lines) and b) high good boundary, dotted lines show ± 25th & 75th residuals of prediction. Horizontal & vertical lines show intersection with RMA regression of observed ortho-P and TN showing boundary values.

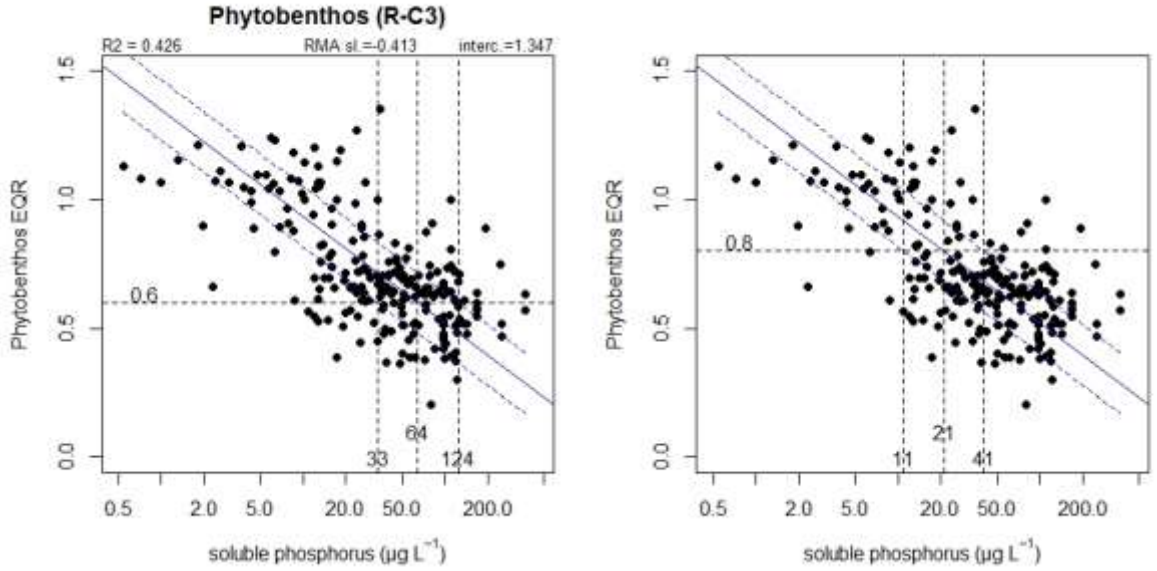


Figure 6-88 Relationship between EQR (as national metric) for phytobenthos and ortho-P for low alkalinity upland rivers (Type R-C3) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data.

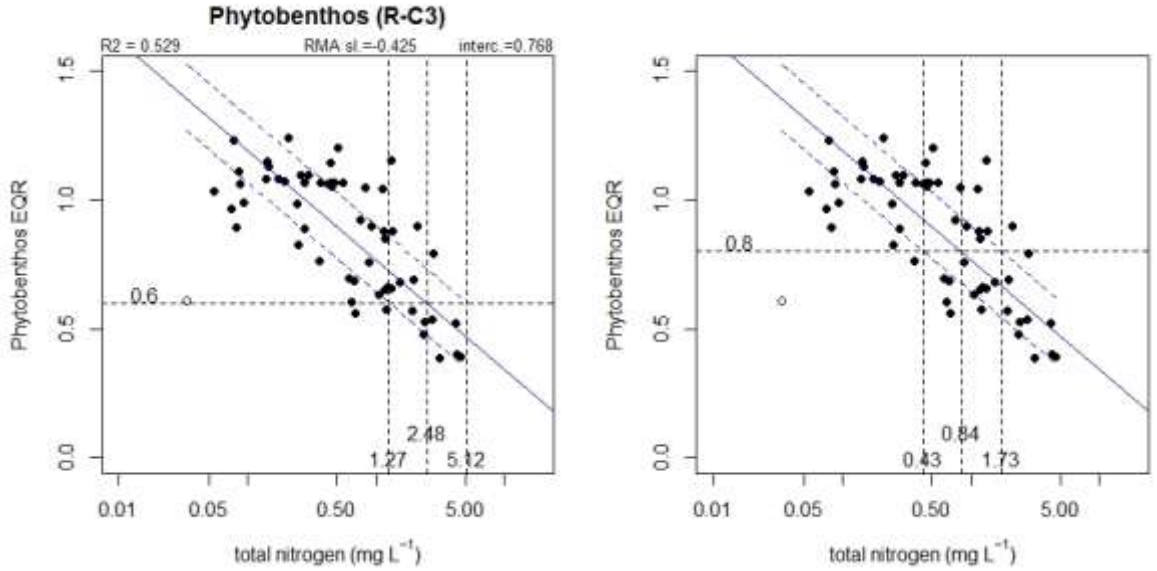


Figure 6-89 Relationship between EQR (as national metric) for phytobenthos and total nitrogen for low alkalinity upland rivers (Type R-C3) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data, open circle: data point excluded from regression.

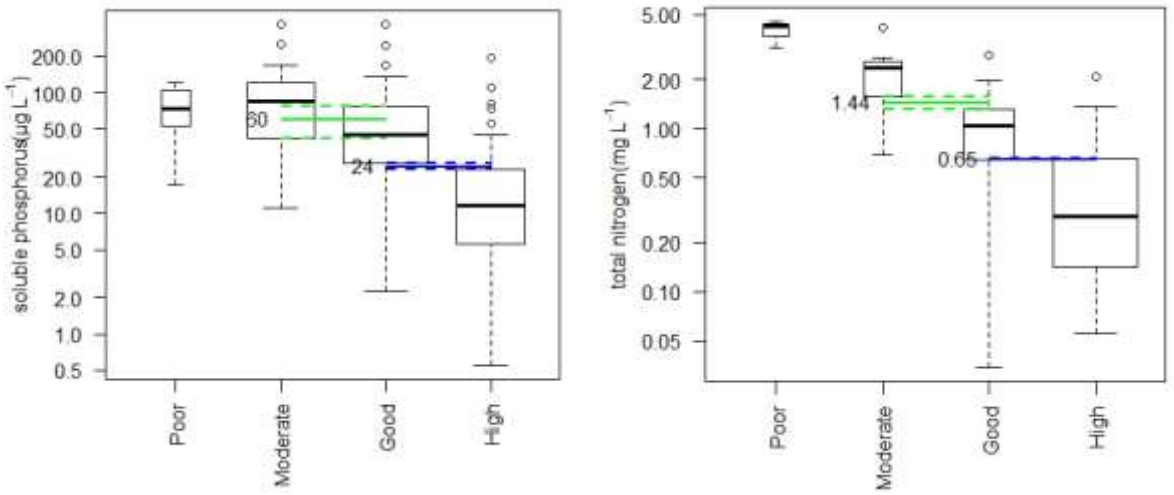


Figure 6-90 Box plots showing range of a) ortho-P and b) TN for low alkalinity upland rivers (Type R-C3) classified using national phytobenthos metrics showing good/moderate boundary & high/good boundary values determined from the average of the upper and lower quartile values.

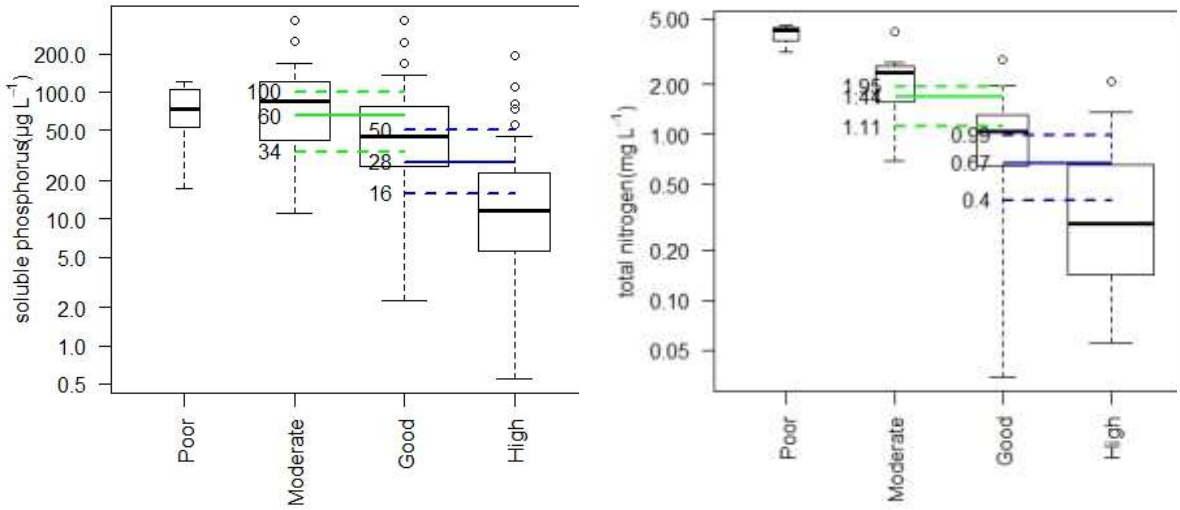


Figure 6-91 Box plots showing range of a) ortho-P and b) TN for low alkalinity upland rivers (Type R-C3) classified using national phytobenthos metrics showing good/moderate boundary & high/good boundary values determined from the average of adjacent classes.

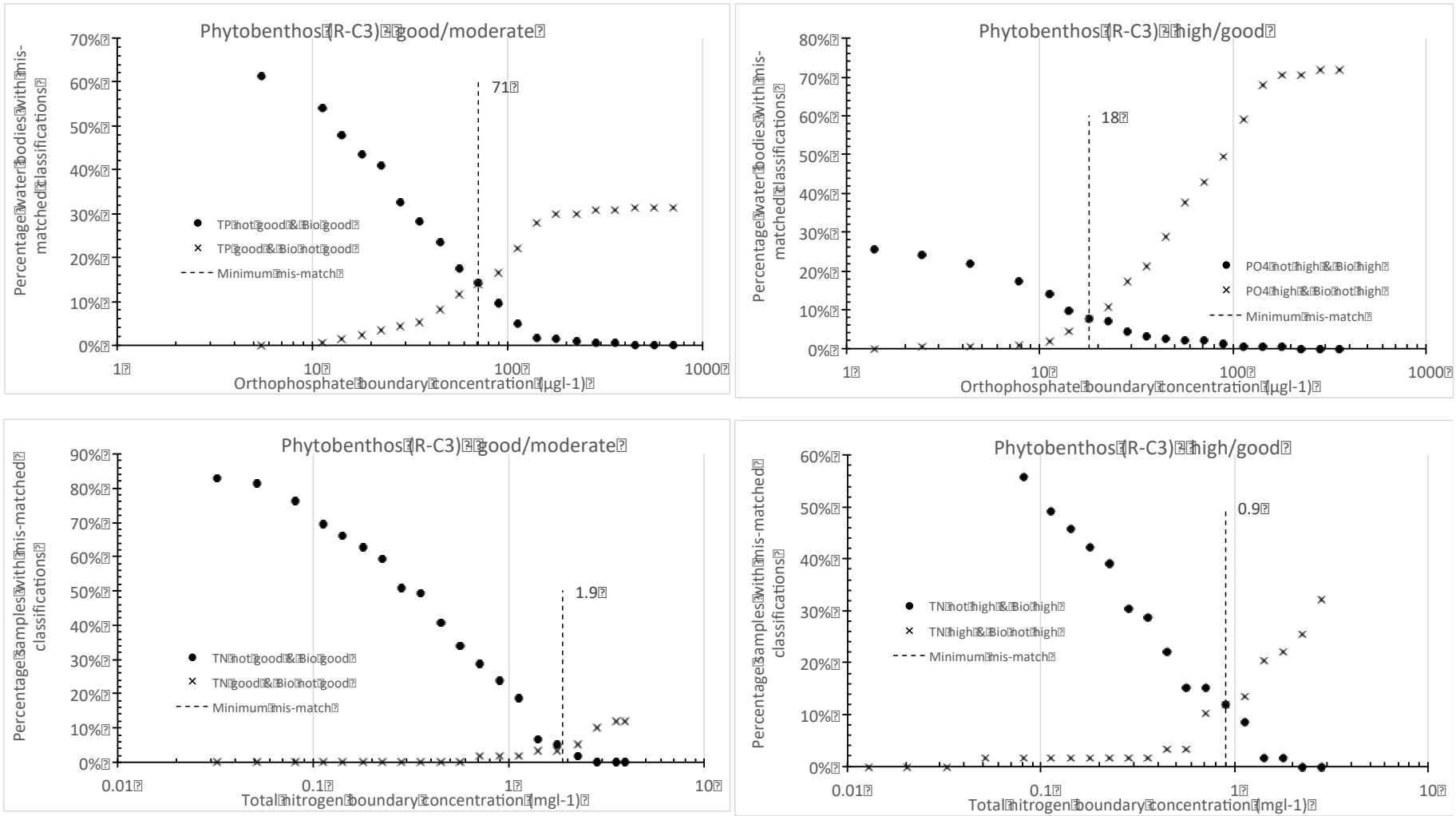


Figure 6-92 Percentage of water bodies where biology or ortho-phosphorus / total nitrogen classifications for good ecological status differ in comparison to the level used to set a) the good/moderate and b) the high/good boundaries. Biological status assessed using national phytobenthos metrics in low alkalinity upland rivers (Type R-C3). Vertical lines mark intersection of curves where mis-match is minimised and equal.

Table 6-54 Regression parameters for R-C3 phytobenthos v ortho-phosphorus and total nitrogen

```
Call:
lm(formula = nPB.EQR ~ log10(PO4.P) + log10(total.N), data = data.cc.ex,
    subset = PO4.P > P.minUsed & PO4.P <= P.maxUsed & total.N >
        N.minUsed & total.N <= N.maxUsed)

Residuals:
     Min       1Q   Median       3Q      Max
-0.52123 -0.11705 -0.04896  0.12647  0.33447

Coefficients:
              Estimate Std. Error t value Pr(>|t|)
(Intercept)    1.04041    0.07524  13.827  < 2e-16 ***
log10(PO4.P)  -0.21297    0.06097  -3.493  0.00108 **
log10(total.N) -0.19632    0.05629  -3.488  0.00110 **
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 0.1739 on 45 degrees of freedom
Multiple R-squared:  0.4503,     Adjusted R-squared:  0.4259
F-statistic: 18.43 on 2 and 45 DF,  p-value: 1.422e-06
```

Table 6-55 Regression parameters for R-C3 phytobenthos v ortho-phosphorus

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x = "interval",
nperm = 99)

n = 230   r = -0.6529178   r-square = 0.4263016
Parametric P-values:   2-tailed = 2.483503e-29    1-tailed = 1.241751e-29
Angle between the two OLS regression lines = 17.78548 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS  1.148578 -0.2832194       -15.81315              0.01
2     MA  1.196293 -0.3143815       -17.45218              0.01
3    SMA  1.379107 -0.4337750       -23.45000                NA
4    RMA  1.347146 -0.4129022       -22.43584              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS       1.079360        1.217797 -0.3260939  -0.2403450
2     MA       1.124334        1.270241 -0.3626762  -0.2673855
3    SMA       1.316694        1.447992 -0.4787632  -0.3930143
4    RMA       1.257039        1.449814 -0.4799530  -0.3540540

Eigenvalues: 0.2864809 0.02607484

H statistic used for computing C.I. of MA: 0.001875855
```

Table 6-56 Regression parameters for R-C3 phytobenthos v total nitrogen

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x = "interval",
nperm = 99)

n = 58   r = -0.7276164   r-square = 0.5294256
Parametric P-values:   2-tailed = 9.798736e-11    1-tailed = 4.899368e-11
Angle between the two OLS regression lines = 14.05469 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS 0.7869596 -0.3449540       -19.03208              0.01
2     MA 0.7783381 -0.3805312       -20.83338              0.01
3    SMA 0.7556662 -0.4740878       -25.36506                NA
4    RMA 0.7676805 -0.4245103       -23.00172              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS      0.7384520       0.8354672 -0.4320127  -0.2578954
2     MA      0.7542097       0.8009685 -0.4800982  -0.2871456
3    SMA      0.7326479       0.7748424 -0.5690736  -0.3949563
4    RMA      0.7393750       0.7919604 -0.5413143  -0.3243178

Eigenvalues: 0.2897663 0.02394773

H statistic used for computing C.I. of MA: 0.007037523
```

6.5.3 Combined macrophyte and phytobenthos (CMP) low alkalinity upland rivers (R-C3)

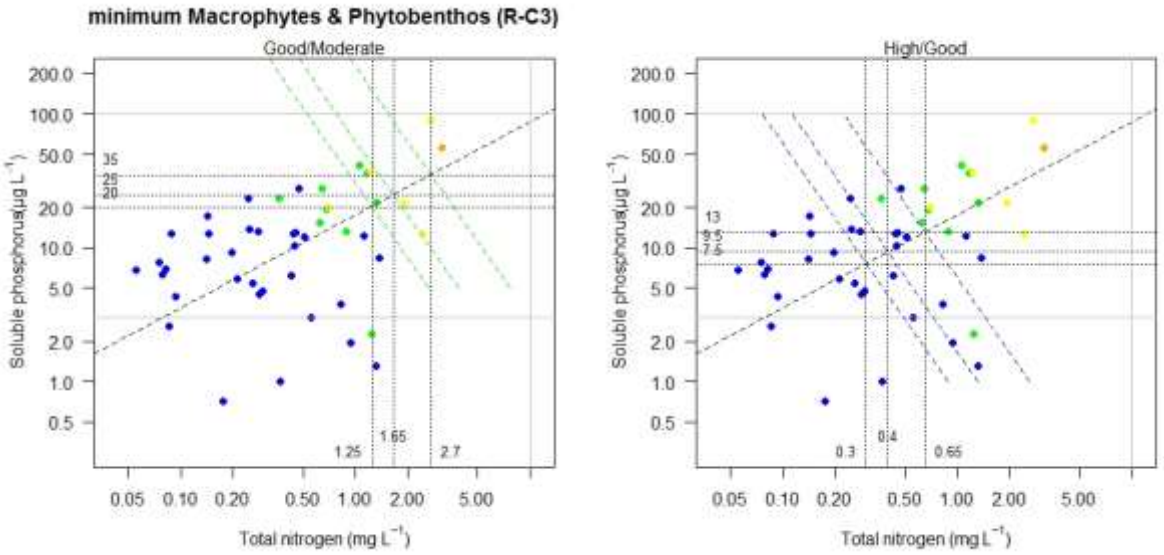


Figure 6-93 Relationship between mean ortho-P and TN, points coloured by WFD class for combined macrophyte and phytobenthos (CMP) in low alkalinity upland rivers (Type R-C3). Dotted lines contours of predicted TN & ortho-P concentration when national phytobenthos metric EQR is at a) good/moderate boundary (green lines) and b) high good boundary, dotted lines show ± 25th & 75th residuals of prediction. Horizontal & vertical lines show intersection with RMA regression of observed ortho-P and TN showing boundary values.

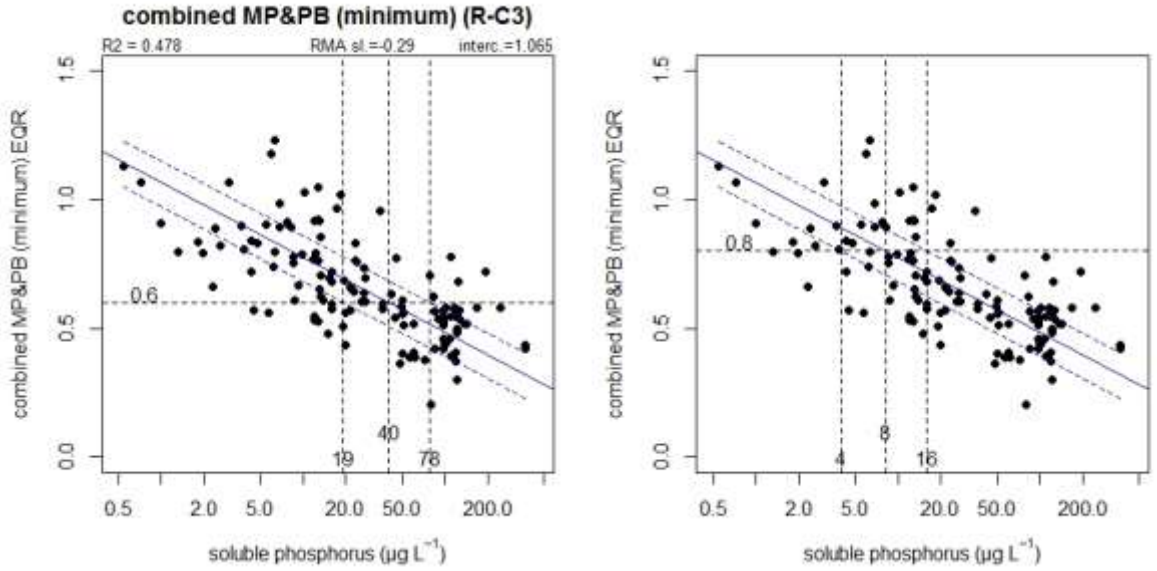


Figure 6-94 Relationship between EQR (as national metric) for combined macrophyte and phytobenthos (CMP) and ortho-P for low alkalinity upland rivers (Type R-C3) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data.

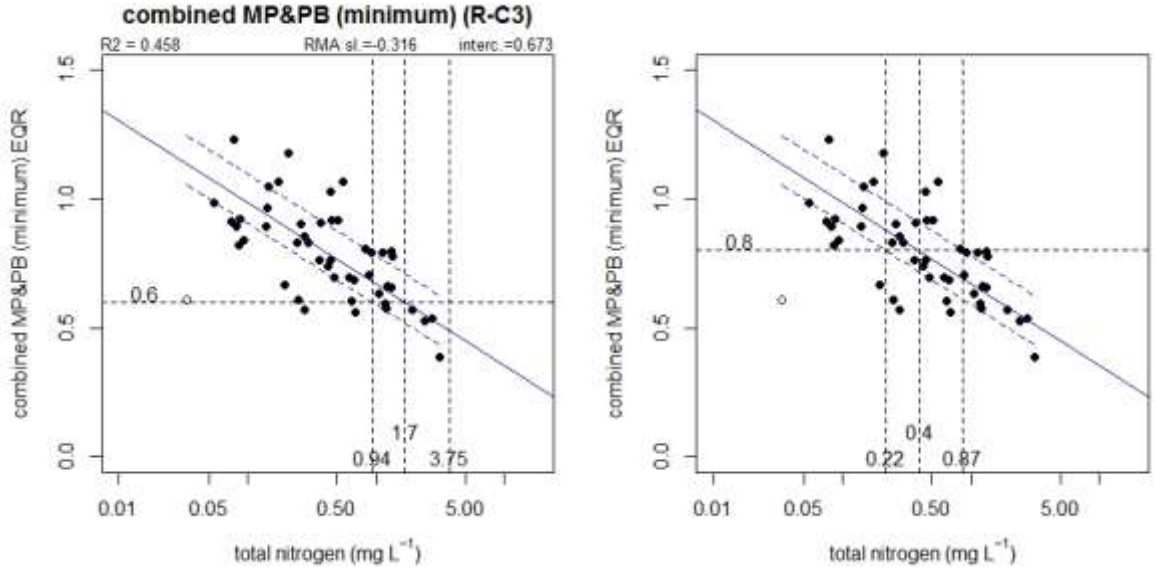


Figure 6-95 Relationship between EQR (as national metric) for combined macrophyte and phytobenthos (CMP) and total nitrate for low alkalinity upland rivers (Type R-C3) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data, open circles data point excluded from regression.

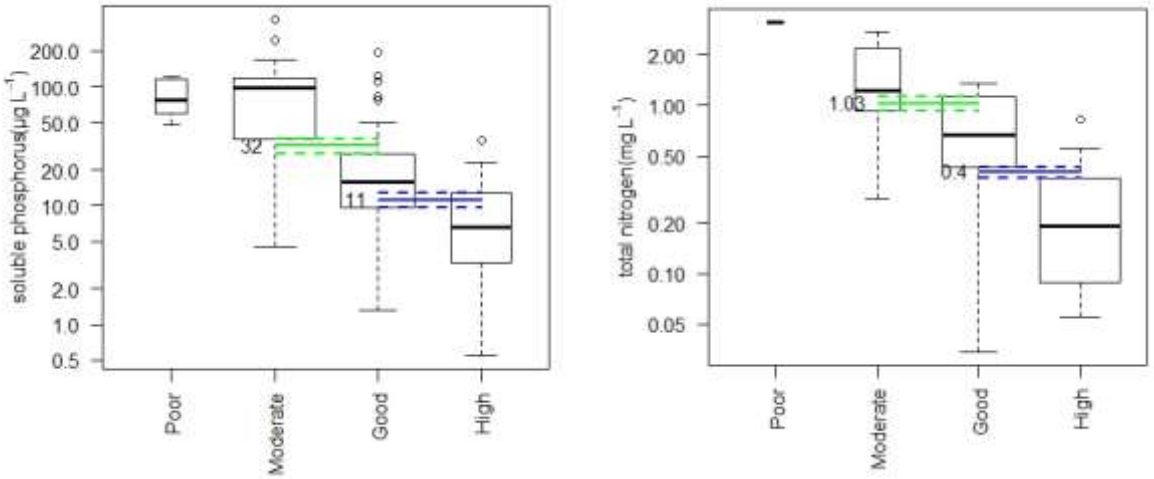


Figure 6-96 Box plots showing range of a) ortho-P and b) TN for low alkalinity upland rivers (Type R-C3) classified using national combined macrophyte and phytobenthose (CMP) metrics showing good/moderate boundary & high/good boundary values determined from the average of the upper and lower quartile values.

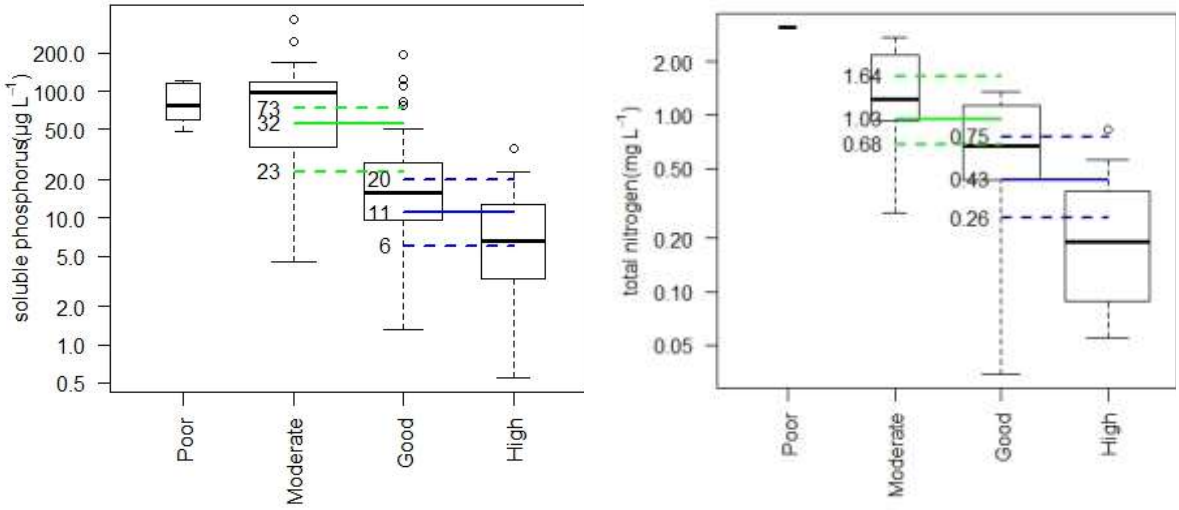


Figure 6-97 Box plots showing range of a) ortho-P and b) TN for low alkalinity upland rivers (Type R-C3) classified using national combined macrophyte and phytobenthos (CMP) metrics showing good/moderate boundary & high/good boundary values determined from the average of adjacent classes.

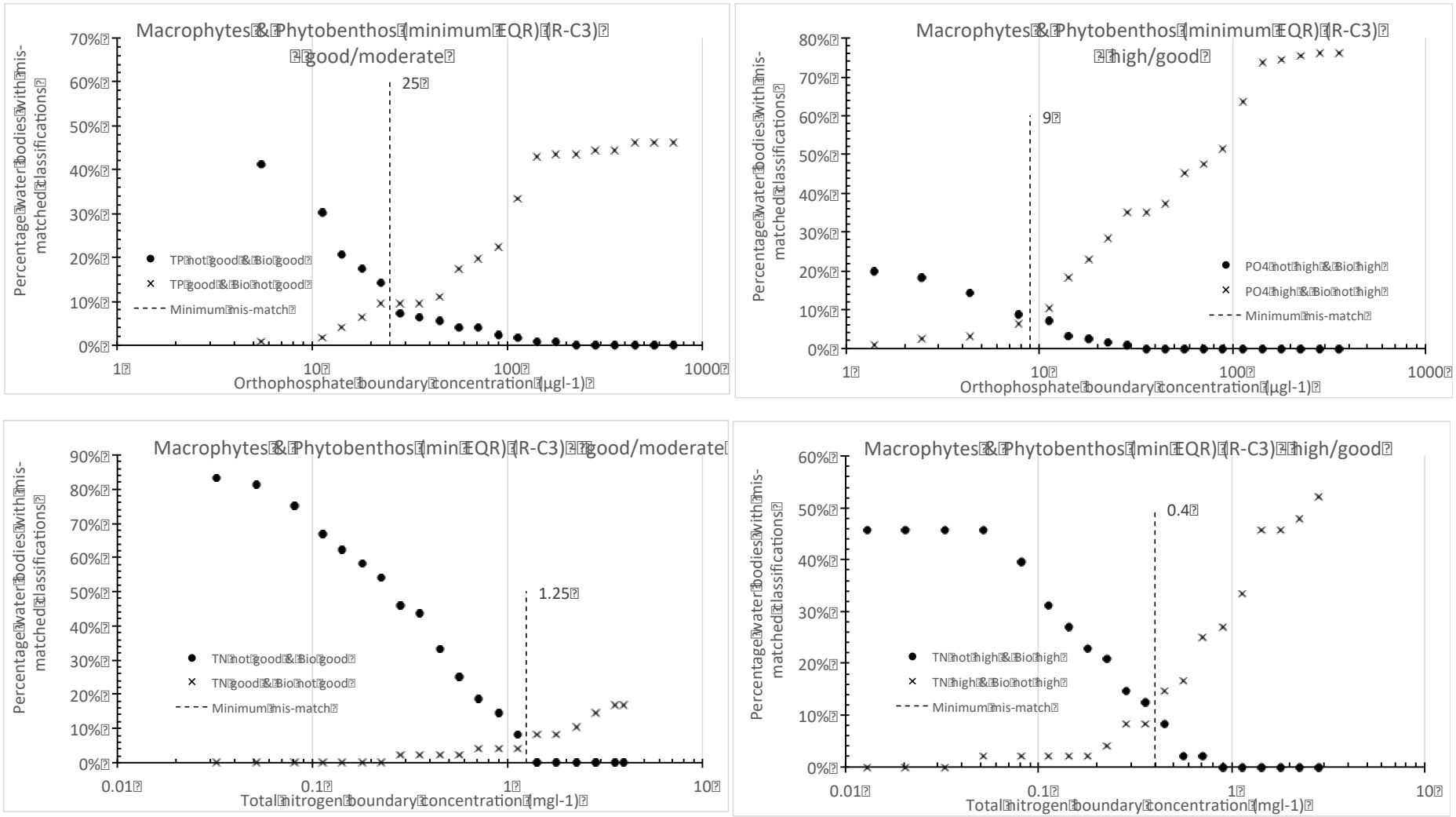


Figure 6-98 Percentage of water bodies where biology or ortho-phosphorus / total nitrogen classifications for good ecological status differ in comparison to the level used to set a) the good/moderate and b) the high/good boundaries. Biological status assessed using national combined macrophyte and phytobenthos (CMP) metrics in low alkalinity upland rivers (Type R-C3). Vertical lines mark intersection of curves where mis-match is minimised and equal.

Table 6-54 Regression parameters for R-C3 combined macrophyte and phytobenthos (CMP) v ortho-phosphorus and total nitrogen

```
Call:
lm(formula = nEQR.min ~ log10(PO4.P) + log10(total.N), data = data.cc.ex,
    subset = PO4.P > P.minUsed & PO4.P <= P.maxUsed & total.N >
        N.minUsed & total.N <= N.maxUsed)

Residuals:
     Min       1Q   Median       3Q      Max
-0.30593 -0.06981 -0.01596  0.04025  0.28452

Coefficients:
              Estimate Std. Error t value Pr(>|t|)
(Intercept)    0.82806    0.10463   7.914 1.78e-09 ***
log10(PO4.P)  -0.12645    0.07910  -1.599 0.118395
log10(total.N) -0.23729    0.05449  -4.355 0.000101 ***
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 0.1257 on 37 degrees of freedom
Multiple R-squared:  0.5608,     Adjusted R-squared:  0.5371
F-statistic: 23.63 on 2 and 37 DF,  p-value: 2.446e-07
```

Table 6-55 Regression parameters for R-C3 combined macrophyte and phytobenthos (CMP) v ortho-phosphorus

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x = "interval",
nperm = 99)

n = 128   r = -0.6912045   r-square = 0.4777637
Parametric P-values:   2-tailed = 1.71111e-19    1-tailed = 8.555549e-20
Angle between the two OLS regression lines = 12.58378 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS 0.9759434 -0.2260643       -12.73841              0.01
2     MA 0.9936532 -0.2387168       -13.42619              0.01
3    SMA 1.1173064 -0.3270585       -18.11077                NA
4    RMA 1.0650143 -0.2896994       -16.15627              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS      0.9125304        1.039356 -0.2677333  -0.1843953
2     MA      0.9326178        1.055915 -0.2831985  -0.1951112
3    SMA      1.0626822        1.179332 -0.3713712  -0.2880332
4    RMA      0.9940399        1.144557 -0.3465270  -0.2389931

Eigenvalues: 0.378968 0.01905758

H statistic used for computing C.I. of MA: 0.001732965
```

Table 6-56 Regression parameters for R-C3 combined macrophyte and phytobenthos (CMP) v total nitrogen

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "relative", range.x = "interval",
nperm = 99)

n = 47   r = -0.6766235   r-square = 0.4578193
Parametric P-values:   2-tailed = 1.780177e-07    1-tailed = 8.900887e-08
Angle between the two OLS regression lines = 15.38792 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS 0.6902938 -0.2691755       -15.06553              0.01
2     MA 0.6815833 -0.2924375       -16.30090              0.01
3    SMA 0.6421220 -0.3978217       -21.69374                NA
4    RMA 0.6727779 -0.3159529       -17.53408              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS      0.6386506       0.7419369 -0.3571255  -0.1812255
2     MA      0.6447120       0.7165970 -0.3909049  -0.1989309
3    SMA      0.6055920       0.6714581 -0.4953777  -0.3194776
4    RMA      0.6315506       0.7096850 -0.4260534  -0.2173898

Eigenvalues: 0.2254758 0.01662669

H statistic used for computing C.I. of MA: 0.007748035
```

6.6 Large rivers

6.6.1 Phytobenthos: medium and high alkalinity very large rivers (R-L2)

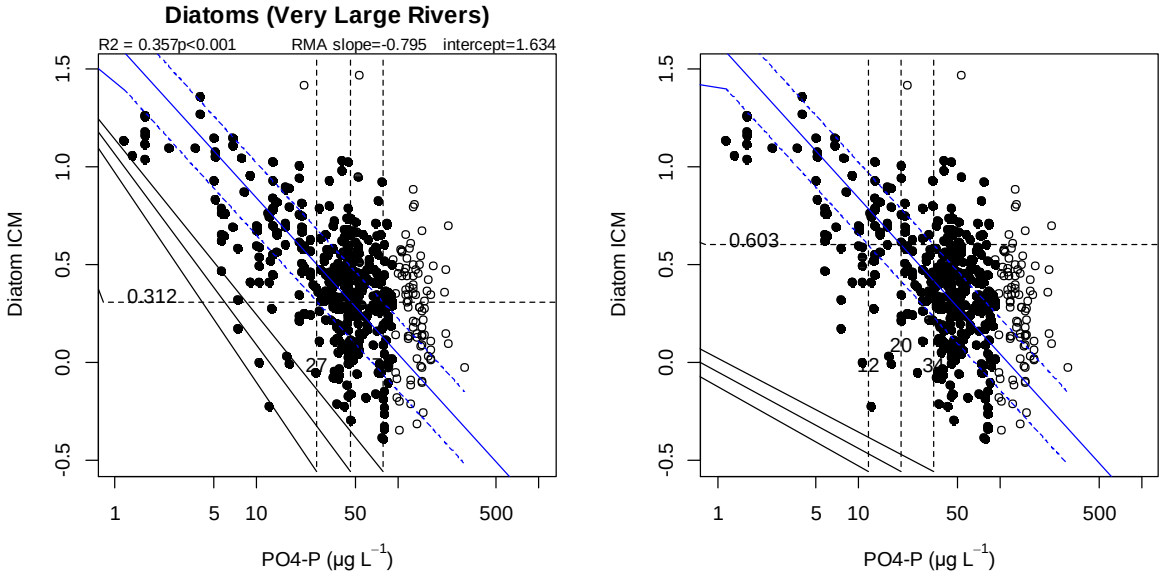


Figure 6-99 Relationship between EQR (common metric) for phytobenthos and ortho-P for high and medium alkalinity large rivers (Type R-L2) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data, open circles data points excluded from regression.

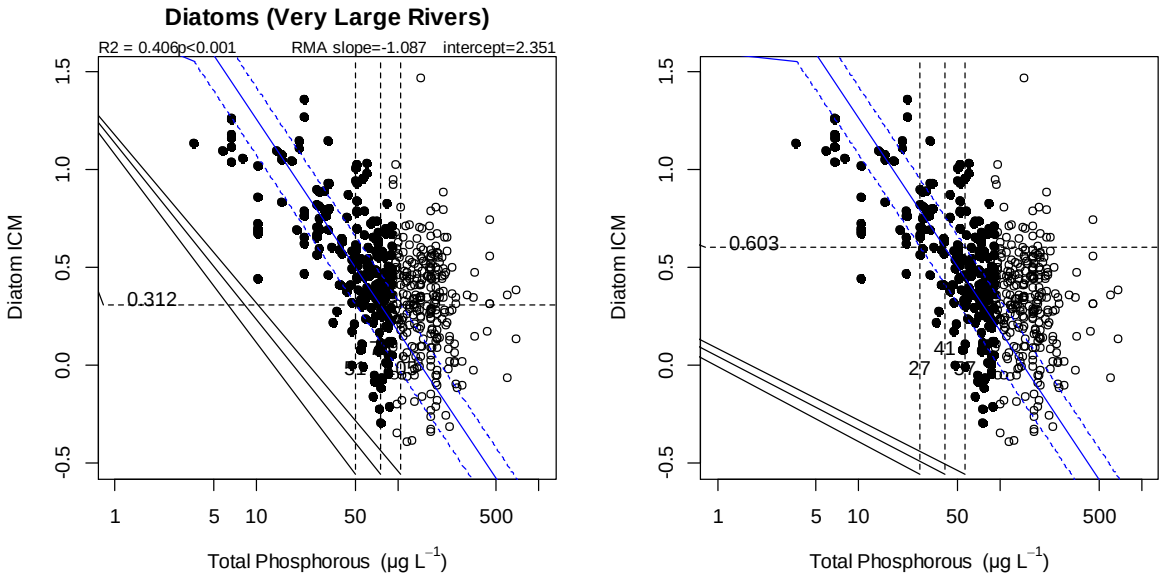


Figure 6-100 Relationship between EQR (common metric) for phytobenthos and total-P for high and medium alkalinity, large rivers (Type R-L2) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data, open circles data points excluded from regression.

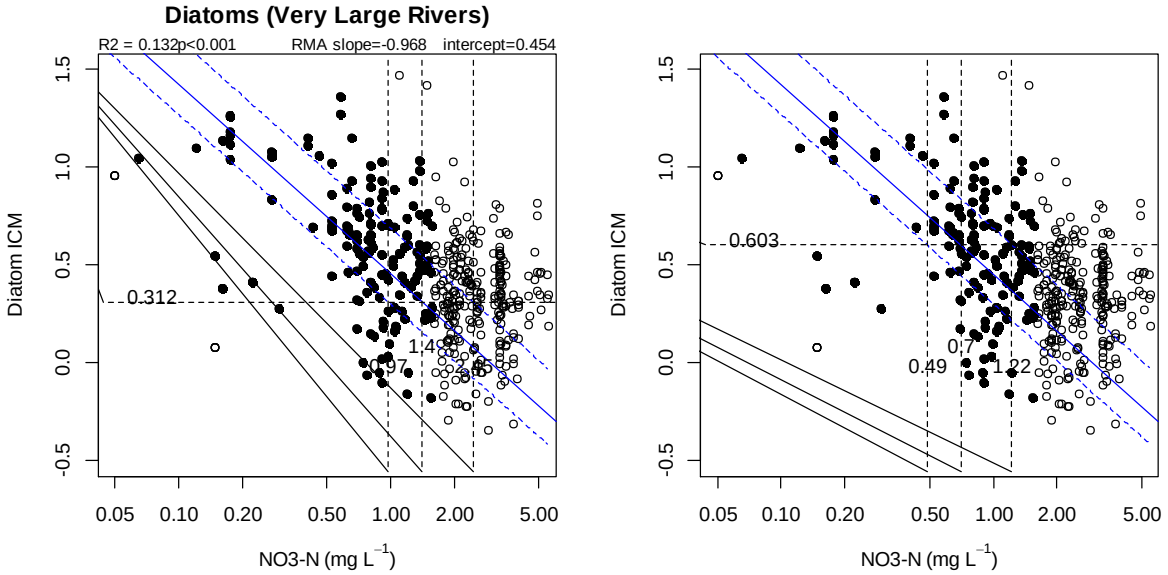


Figure 6-101 Relationship between EQR (as national metrics) for phytobenthos and Nitrate-N for high and medium alkalinity, large rivers (Type R-L) showing a) good/moderate boundary and b) high/good boundary values. Line shows type II RMA regression, dotted lines show area containing 50% of the data, open circles data points excluded from regression.

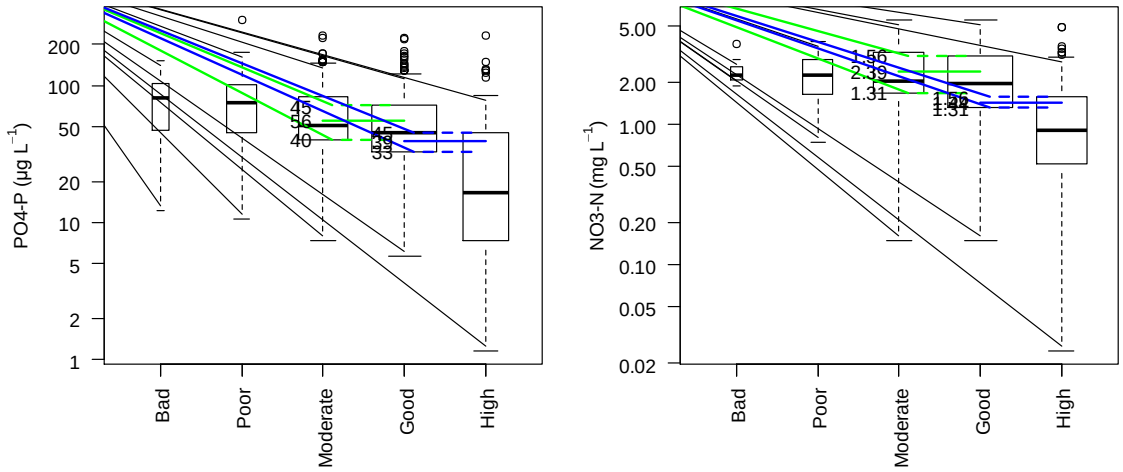


Figure 6-102 Box plots showing range of a) ortho-P and b)Nitrate-N for high and medium alkalinity large rivers (Type R-L) classified using phytobenthos showing good/moderate boundary & high/good boundary values determined from the average of the upper and lower quartile values

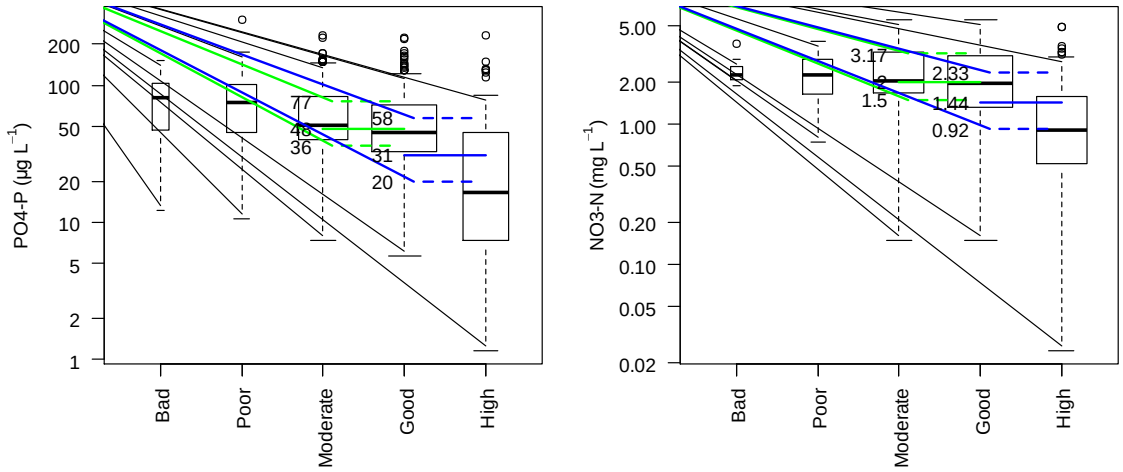
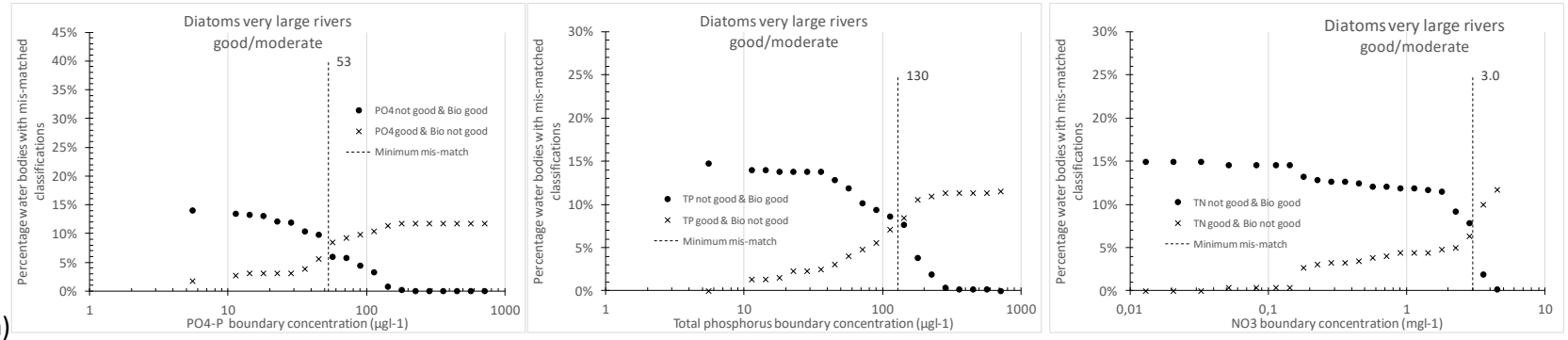


Figure 6-103 Box plots showing range of a) ortho-P and b)Nitrate-N for high and medium alkalinity large rivers (Type R-L) classified using phytobenthos showing good/moderate boundary & high/good boundary values determined from the average of adjacent classes

a)



b)

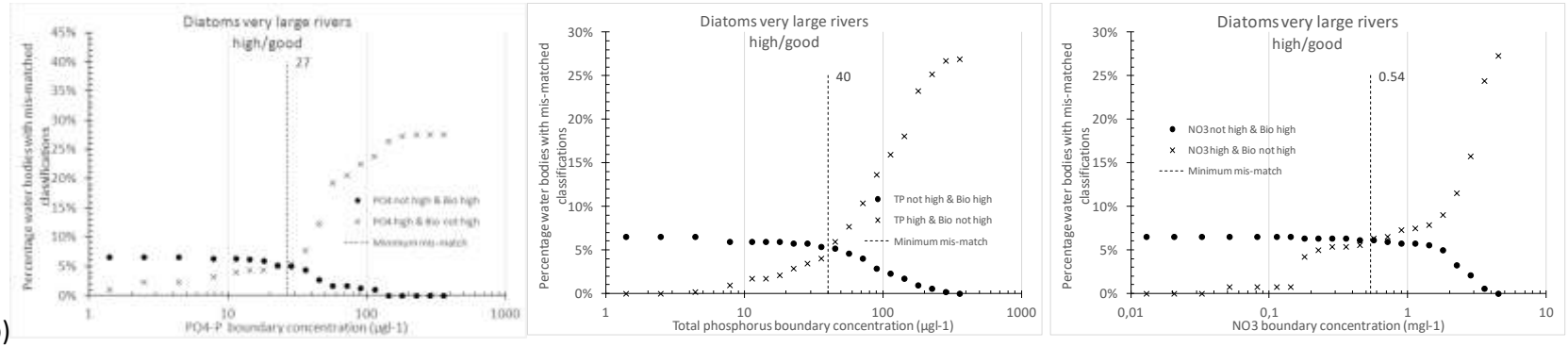


Figure 6-104 Percentage of water bodies where biology or ortho-phosphorus / total nitrogen classifications for good ecological status differ in comparison to the level used to set the a) good/moderate and b) the high/good boundaries. Biological status assessed using phytobenthos metrics in high and medium alkalinity large rivers (Type R-L) Vertical lines mark intersection of curves where mismatch is minimised and equal.

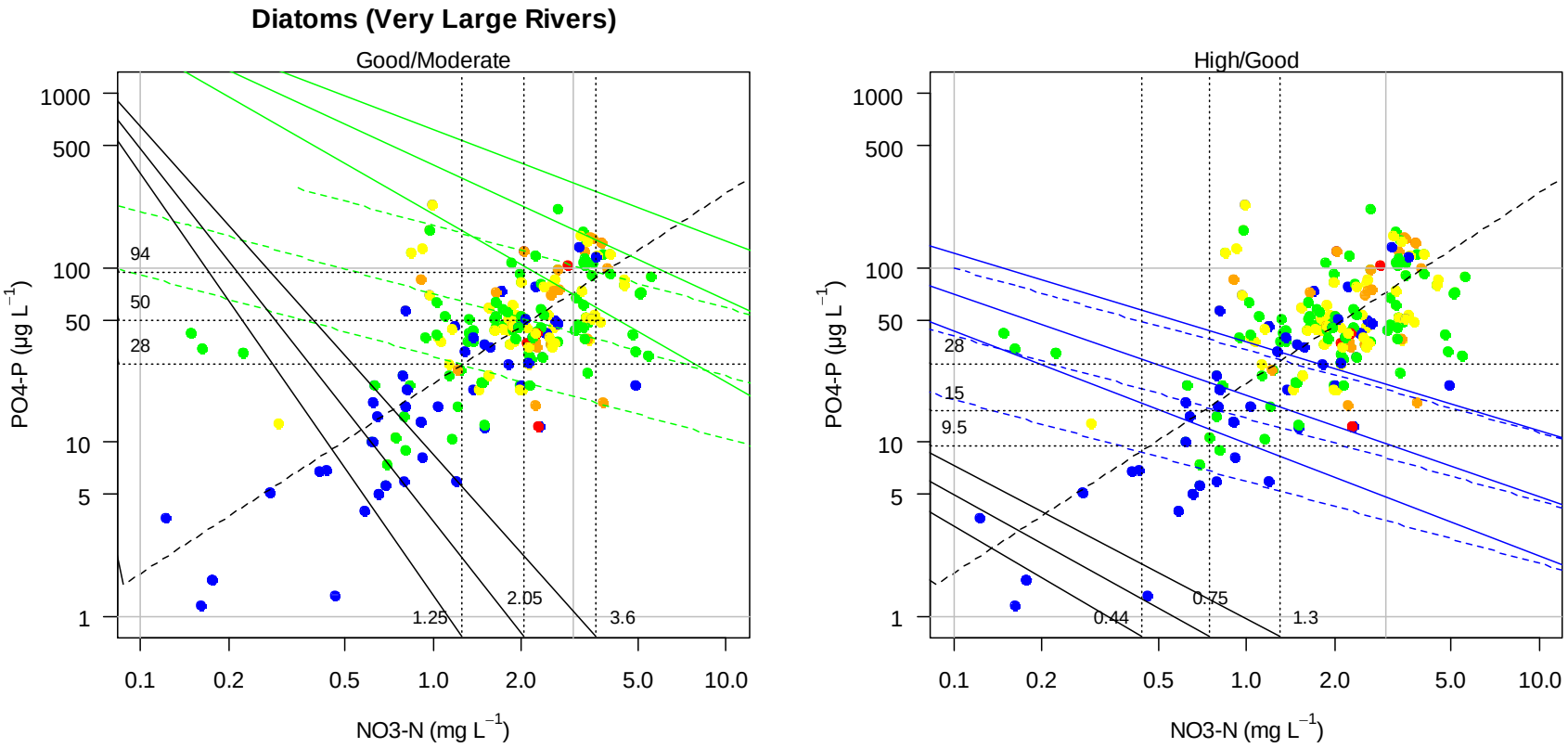


Figure 6-105 Relationship between mean ortho-P and nitrate N, points coloured by WFD class for phytobenthos in very large rivers (Type R-L). Dotted lines contours of predicted TN & ortho-P concentration when national phytobenthos ICM is at a) good/moderate boundary (green lines) and b) high good boundary, dotted lines show ± 25th & 75th residuals of prediction. Horizontal & vertical lines show intersection with RMA regression of observed ortho-P and nitrate N showing boundary values.

Table 6-57 Regression parameters for R- L phytobenthos v ortho- phosphorus and total nitrogen

```
Call:
lm(formula = CM.EQR ~ log10(total.P) + log10(total.N), data = data.cc.ex,
    subset = total.P > P.minUsed & total.P <= P.maxUsed & total.N >
        N.minUsed & total.N <= N.maxUsed)

Residuals:
     Min       1Q   Median       3Q      Max
-0.77710 -0.15856 -0.00752  0.14707  0.69325

Coefficients:
               Estimate Std. Error t value Pr(>|t|)
(Intercept)     1.06484    0.06669  15.967  < 2e-16 ***
log10(total.P) -0.40677    0.04779  -8.511  8.1e-16 ***
log10(total.N) -0.19143    0.05760  -3.324 0.000998 ***
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 0.2425 on 303 degrees of freedom
Multiple R-squared:  0.3989,    Adjusted R-squared:  0.3949
F-statistic: 100.5 on 2 and 303 DF,  p-value: < 2.2e-16
```

Table 6-58 Regression parameters for R- L phytobenthos v ortho- phosphorus

```
Model II regression

Call: lmodel2(formula = y.u ~ x.u, range.y = "interval", range.x = "interval",
nperm =99)

n = 435   r = -0.5975717   r-square = 0.3570919
Parametric P-values:   2-tailed = 1.860003e-43    1-tailed = 9.300014e-44
Angle between the two OLS regression lines = 27.93247 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS  1.193953 -0.5038423       -26.74090              0.01
2     MA  1.572031 -0.7534463       -36.99606              0.01
3    SMA  1.707905 -0.8431495       -40.13590                NA
4    RMA  1.634378 -0.7946076       -38.47092              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS       1.094510        1.293395 -0.5676978  -0.4399867
2     MA       1.433492        1.723923 -0.8537243  -0.6619842
3    SMA       1.614840        1.808285 -0.9094197  -0.7817085
4    RMA       1.489391        1.795974 -0.9012918  -0.6988882

Eigenvalues: 0.180936 0.04344757

H statistic used for computing C.I. of MA: 0.003710201
```

Table 6-59 Regression parameters for R- L phytobenthos v total nitrogen

```
Model II regression
Call: lmodel2(formula = y.u ~ x.u, range.y = "interval", range.x = "interval",
nperm =99)

n = 170   r = -0.3637685   r-square = 0.1323275
Parametric P-values:   2-tailed = 1.082201e-06    1-tailed = 5.411006e-07
Angle between the two OLS regression lines = 49.97237 degrees

Permutation tests of OLS, MA, RMA slopes: 1-tailed, tail corresponding to sign
A permutation test of r is equivalent to a permutation test of the OLS slope
P-perm for SMA = NA because the SMA slope cannot be tested

Regression results
  Method Intercept      Slope Angle (degrees) P-perm (1-tailed)
1    OLS 0.5361937 -0.3856349       -21.08838              0.01
2     MA 0.4252643 -1.1733670       -49.56076              0.01
3    SMA 0.4412132 -1.0601106       -46.67132                NA
4    RMA 0.4541404 -0.9683123       -44.07768              0.01

Confidence intervals
  Method 2.5%-Intercept 97.5%-Intercept 2.5%-Slope 97.5%-Slope
1    OLS      0.4869549       0.5854325 -0.5360399  -0.2352299
2     MA      0.3399481       0.4786475 -1.7792148  -0.7942824
3    SMA      0.4185380       0.4608984 -1.2211320  -0.9203219
4    RMA      0.3887896       0.5004433 -1.4323815  -0.6395055

Eigenvalues: 0.1276166 0.05898456

H statistic used for computing C.I. of MA: 0.03707304
```

Table 6-42- Predicted phosphorus and nitrogen boundary values for high and medium large rivers using regression models and categorical methods

IC Type	Phytobenthos Models	R2	nutrient range PO4-P µgl^{-1}	GM PO4			HG PO4		
				Pred	25th	75th	Pred	25th	75th
R-L	EQR v PO4 (OLS)	0.357	1 - 92	56	31	117	14.89	8	31
	PO4 v EQR (OLS)			40	28	63	25	18	39
	EQR v PO4 (RMA)			46	27	79	20	12	34
R-L	Average adjacent quartiles			56			39		
	Average adjacent classes			48			31		
	Minimise class difference			53			27		

IC Type	Phytobenthos Models	R2	nutrient range TP µgl^{-1}	GM TP			HG TP		
				Pred	25th	75th	Pred	25th	75th
R-L	EQR v TP (OLS)	0.406	4 - 91	95	59	151	37	23	59
	TP v EQR (OLS)			63	48	90	43	33	61
	EQR v TP (RMA)			75	51	105	41	27	57
R-L	Average adjacent quartiles			125			77		
	Average adjacent classes			108			74		
	Minimise class difference			130			40		

IC Type	Phytobenthos Models	R2	nutrient range NO3-N µgl^{-1}	GM NO3			HG NO3		
				Pred	25th	75th	Pred	25th	75th
R-L	EQR v NO3 (OLS)	0.132	0.06 - 1.56	3.8	1.04	13.4	0.67	0.18	2.35
	NO3 v EQR (OLS)			0.9	0.74	1.4	0.72	0.59	1.11
	EQR v NO3 (RMA)			1.4	0.97	2.45	0.7	0.49	1.22
R-L	Average adjacent quartiles			56			39		
	Average adjacent classes			48			31		
	Minimise class difference			53			27		

6.6.2 Invertebrates: Very large rivers (R-L)

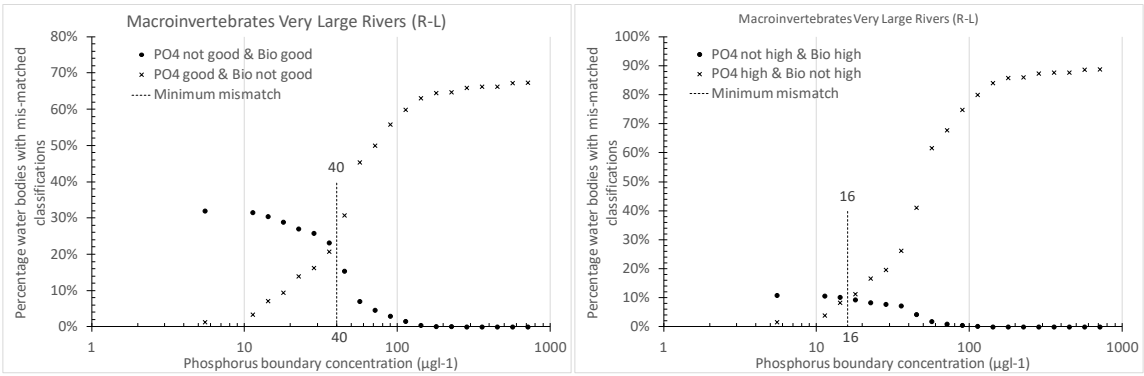


Figure 6-106 Percentage of water bodies where biology or ortho-phosphorus / total nitrogen classifications for good ecological status differ in comparison to the level used to set the a) good/moderate and b) the high/good boundaries. Biological status assessed using macroinvertebrate metrics in very large rivers (Type R-L) Vertical lines mark intersection of curves where mismatch is minimised and equal.

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