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1 **Macro-invertebrate response to phosphorus levels in inland waters**

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13

14

1 **Abstract**

2 The magnitude of ecological damage caused by elevated phosphorus concentrations in Dutch inland waters was  
3 evaluated and expressed as the fraction of disappeared genera. We used national abundance data stored in the  
4 *Limnodata Neerlandica* from 1980 until 2005. We derived the presence or absence in the concentration range  
5 between 0.001 and 40 mg/l total phosphorus for 867 macro-invertebrate genera. At concentrations above 0.1 mg/l,  
6 which is considered a signal of nutrient enrichment of freshwater, the fraction of disappeared macroinvertebrate  
7 genera (DF) can be written as a logistic function of the phosphorus concentration ( $C_p$ ), i.e.  $DF = 1/(1 + 4.07 \cdot C_p^{-1.11})$ .  
8 This implies that half of the macro-invertebrate genera that potentially occur in freshwater in the Netherlands would  
9 disappear at a phosphorus concentration of 3.5 mg/l. This field-based impact expression resembles the cumulative  
10 sensitivity distribution function for a toxic substance, based on the Species Sensitivity Distribution (SSD) concept  
11 and artificial exposure test data. While the SSD for a compound relies on laboratory sensitivity data for a small  
12 number of species, the fraction of disappeared macro-invertebrate genera is here derived from field observations of  
13 many macroinvertebrate genera at numerous phosphorus concentration intervals. By applying the new damage  
14 function to measured phosphorus concentrations over the period 1975-2005 in the rivers Rhine, Meuse and Scheldt,  
15 we found that the observed change in phosphorus concentrations would imply a loss of macro-invertebrates of 20-30  
16 % initially to 5-10 % in 2005. The cumulative sensitivity distribution function for phosphorus from national  
17 freshwater monitoring data can be applied in various environmental screening systems such as multi-stress impact  
18 assessment of surface waters and in life cycle impact assessment of products.

19  
20 Keywords: freshwater eutrophication, total phosphorus concentration, macro-invertebrate  
21 sensitivity distribution, *Limnodata Neerlandica*

## 1 **1 Introduction**

2

3 The natural phosphorus cycle is the predominant source of phosphorus for organisms. For inland  
4 waters in temperate zones, phosphorus is most often the limiting nutrient (Crouzet et al., 1999)  
5 and its natural availability regulates algae and macrophyte growth. In large regions, however,  
6 industrial and agricultural sources exceed natural inputs by far. As a result, undesirable growth of  
7 phytoplankton and floating plants may locally occur, with a chain of subsequent adverse  
8 ecological effects such as oxygen depletion, production of biotoxins and decrease of habitat  
9 diversity (Kristensen and Hansen, 1994). In densely populated industrial areas, eutrophication is  
10 one of the major factors causing ecological impairments in inland waters: nutrient enrichment  
11 accounts for about half of the ecologically impaired lake areas and 60% of the impaired river  
12 reaches in the United States (Carpenter et al., 1998). When evaluated on a national scale,  
13 phosphorus enrichment in the Dutch situation ranks higher as cause of ecological impairments in  
14 surface waters than exposure to toxic substances (RIVM, 2000).

15 The unbalanced growth of phytoplankton and macrophytes causes freshwater to become  
16 obfuscated and depleted from oxygen to an extent that life of macro-invertebrates (and fishes) has  
17 become virtually impossible. Quantitative description of ecological impacts and analysis of their  
18 probable causes in extensive water bodies is rather difficult , because the magnitude of the  
19 impairments not only depends on nutrient enrichments, but also on local factors like topography  
20 and the physical and chemical nature of water bodies, as well as on earlier community responses  
21 to all these factors. Moreover, when conditions such as nutrient loading approach a certain  
22 critical level, the response pattern may not be a continuous increase of impact magnitudes but is  
23 rather an unexpectedly strong change (Scheffer et al. 2001). As an example, phosphorus  
24 enrichment may push the aquatic ecosystem over an apparent edge and may cause a shift to

1 another metastable state, with a completely different species composition and associated physico-  
2 chemical consequences.

3 Several attempts have been made to assess ecological impairment quantitatively. As examples,  
4 Smith et al. (2007) established a nutrient biotic index in order to score thresholds and  
5 corresponding nutrient concentrations, above which invertebrate communities show impairment  
6 due to increased stream water nutrient concentrations. Solheim and Gulati (2008) made attempts  
7 to describe the relationships between ecological and chemical status, two key concepts in the EU-  
8 Water Framework Directive. According to these authors, knowledge of quantitative associations  
9 is needed for a successful implementation of the Framework Directive.

10 Many studies of this kind however, have a short reach of application within the wide diversity of  
11 aquatic ecosystems, due to focus on a limited range of water bodies, for example shallow or  
12 oligotrophic lakes with specific characteristics (Leuven et al. 1987; Leuven, 1988), or water  
13 systems that are under heavy anthropogenic pressure like ditches and streams in agricultural areas  
14 (Chambers, et al. 2008) and urban drainage systems (Vermonden et al., 2009). This was also the  
15 conclusion of Nijboer and Verdonschot (2004) who stated that the sensitivity of a stream to  
16 eutrophication depends on local stream characteristics and that this aspect should be included in a  
17 generally applicable model.

18 Protective and curative practical management decisions are currently served by, tools, such as life  
19 cycle assessment of products and multi-stress ecological risk assessment of economic activities  
20 (Kapo et al, 2009; De Zwart et al., 2006; De Zwart et al. 2009). There is a need for a quantitative  
21 approach to assess the damage due to anthropogenic emissions on aquatic ecosystems in a way  
22 that is scientifically justifiable, sufficiently robust and applicable to generic problems (like in  
23 LCA) and for local assessments.

1 Multi-stress analysis of freshwater ecosystems implies the need to evaluate not only the  
2 impairment due to nutrient enrichment, but also due to toxic stress, habitat destruction and a suite  
3 of other stress factors. This was shown by, e.g., De Zwart et al. (2006), who conducted a  
4 diagnostic analysis using freshwater (bio)monitoring data of natural waters in the State of Ohio,  
5 USA. They derived, amongst others, multivariate empirical relationships between stressor  
6 variables and fish species occurrences and abundances. As shown by De Zwart et al. (2009), such  
7 explicit stressor-specific field-based concentration-response functions can be helpful in the  
8 context of multi-stress analyses and the derivation of river basin management plans.

9 Concentration-response relationships, also known as Species Sensitivity Distributions (SSDs)  
10 have been practically used for the risk assessment of toxic chemicals in the freshwater  
11 environment since the 80-ties of the last century (Posthuma et al., 2002; De Vries et al., 2008),  
12 but also for assessing the impacts of physical factors like suspended clays, sediment burial, and  
13 grain size change in the marine environment (Smit et al., 2008), and for analysing the probable  
14 impacts of acid deposition on European forests (Van Zelm et al., 2006). A quantitative  
15 description of the concentration-response relationship between ambient phosphorus  
16 concentrations and the occurrence of species for freshwater systems is however, still lacking.

17 For fill this gap, we propose a method to disclose the relationships between stressor variables and  
18 species occurrences that are concealed in (bio)monitoring data, by analysing macro-invertebrate  
19 occurrence and total phosphorus concentrations for freshwater systems (e.g., rivers, lakes and  
20 ditches) in the Netherlands. Evidently, the same method can be applied to other stressors, and to  
21 subsets of the data, or to larger geographies. In this paper we relate the disappearance of macro-  
22 invertebrate genera from natural surface waters to the total phosphorus concentration in Dutch  
23 inland waters.

24 The aims of this study were to:

- 1 1. Develop and describe a model to quantify ecological impacts as a function of the level of an  
2 ambient stressor variable, based on (bio)monitoring data (in this case concerning aquatic  
3 macro-invertebrate diversity affected by enhanced phosphorus exposure in temperate zones,  
4 using data from Dutch inland waters collected between 1980 and 2005);
- 5 2. Illustrate its usefulness in model development, adapted to aquatic macro-invertebrate  
6 diversity affected by enhanced phosphorus exposure in temperate zones.
- 7 3. Indicate the possibility to apply this model generically in decision support systems like  
8 environmental risk assessment and Life Cycle Impact Assessment.

9 We note that the method can equally be used for selected water bodies.

10

## 11 **2 Methodology**

12

### 13 *2.1 General concept*

14 We used field data stored in the *Limnodata Neerlandica* (STOWA, 2006) to analyze the  
15 abundance of macro-invertebrate genera in relation to the variability of phosphorus (P)  
16 concentrations. The diversity of macro-invertebrates is often considered a suitable and sensitive  
17 parameter to characterize the ecological condition of water, because of their relatively high  
18 abundance, high diversity and an often high but species-dependent sensitivity to changing  
19 environmental conditions (e.g. Metcalfe, 1989). Moreover, they are included in standard  
20 monitoring programs of water boards in the Netherlands and elsewhere.

21 A sensitivity distribution at the species level could have been used to analyze trends in ecological  
22 impacts. For practical reasons however (e.g. taxonomic resolution of the available database and  
23 impossibility to identify all sampled invertebrates to species level) and considering arguments of

1 Lenat & Resh (2001) regarding the effect of the taxonomic level on biological assessment, we  
2 used genera diversity as a starting point for modeling.

3  
4 An empirical relationship between the number of “occurring genera” and the total phosphorus  
5 concentration ( $P_{tot}$ ) was derived from field data. Abundance data of macro-invertebrate genera are  
6 useful provided these data were collected on locations where the concentration of  $P_{tot}$  is known.

7 Therefore we converted a large data set containing the concentration of  $P_{tot}$  and the abundance of  
8 macro-invertebrate genera into a matrix of a binary variable, representing the presence or absence  
9 of a specific genus in a  $P_{tot}$  concentration interval. The predictor or independent variable is the  
10  $P_{tot}$  concentration and the dependent variable is the dichotomous value for the macroinvertebrate  
11 genera. This approach allows for an evaluation of the number of potentially occurring  
12 macroinvertebrate genera, distributed over the  $P_{tot}$  concentration. Thus, whether a certain genus is  
13 “occurring” (1) or “not occurring” (0) at a  $P_{tot}$  concentration depends on the presence of an  
14 abundance number.

15 Key questions are whether the number of occurring genera (N) displays a trend with  $P_{tot}$   
16 concentration and whether N has a single optimum ( $N_{max}$ ). Conceivably, the latter maximum  
17 would represent a genera richness that occurs under optimal conditions (minimal disturbance) for  
18 all stressors: phosphorus is – at these sites – not the factor reducing genera diversity. In general,  
19 that situation indicates the apparent lowest pressure of a suite of stressors on the ecosystems due  
20 to human activities.

21  
22 Apart from eutrophication, lower P levels may indicate oligotrophication, which may imply  
23 nutrient deficiency, reflected by a diminished diversity of macro-invertebrate genera at lower P  
24 concentrations. For assessing these data however, it should be taken into consideration that a low



1 diversity of macro-invertebrate genera at low P levels may also be due to anthropogenic stress  
2 factors such as acidification. Nevertheless, a phosphorus concentration higher than the optimal  
3 level is associated with nutrient enrichment, which may cause a diminished number of occurring  
4 genera. Phosphorus concentrations higher than 0.1 mg/l are usually of anthropogenic origin  
5 (Smith et al., 2003).

6

## 7 *2.2 Data pre-treatment*

8

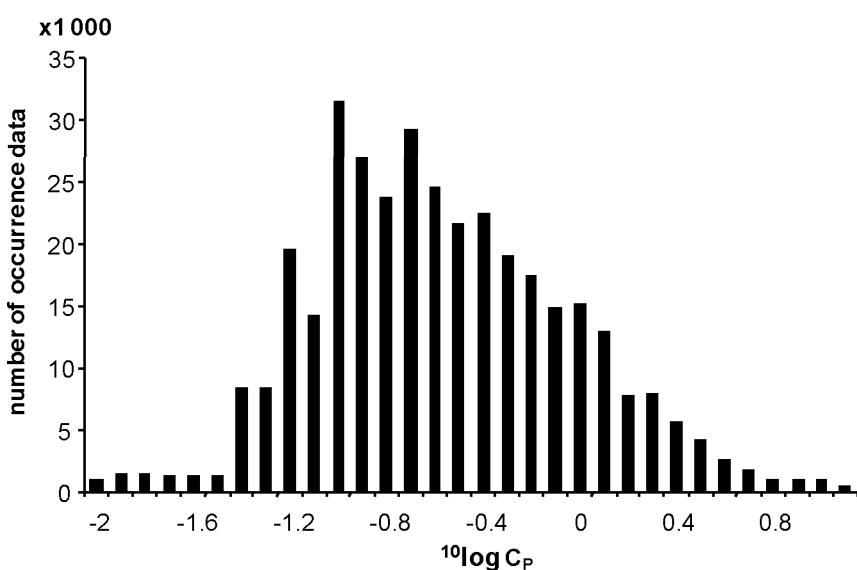
9 **Raw data.** Monitoring data on (pelagic) aquatic species and chemical and physical parameters of  
10 Dutch surface waters between June 1980 and March 2005 are stored in the *Limnodata*  
11 *Neerlandica* (STOWA, 2006). From this database abundance data of macroinvertebrate and  
12 concentrations of  $P_{\text{tot}}$  were selected. Each record, identified by a unique combination of location  
13 and time, contains the observed abundance of a single macro-invertebrate taxon, or a single  
14 abiotic parameter value ( $P_{\text{tot}}$ ).

15 A set of 1 056 194 records with 2 790 different macrofauna taxa was combined with a set of 265  
16 959 records of total phosphorus concentrations. The measured phosphorus concentrations were  
17 log-normally distributed in the range of 0.0015 – 40 mg  $P_{\text{tot}}$ /l (see Figure 1). Only one  
18 concentration (244 mg/l) falls outside this range and because it is probably due to an artefact  
19 (incidental spill or erroneous data input) it was discarded.

20

21 **Aggregation of total phosphorus concentrations.** If the monitoring frequency at a location was  
22 higher than once per month, we calculated the monthly average of  $P_{\text{tot}}$ . This operation provided a  
23 dataset that is suitable for comparison with the abundance data of macrofauna taxa as these data  
24 were also aggregated on a monthly basis.

1 The concentration of  $P_{\text{tot}}$  was subdivided into intervals of one tenth of log units. The intervals  
 2 (denoted as  $\log C_P$  with  $C_P$  in mg/l) used in the analysis range from -2.8 to 1.6 (1.6  $\mu\text{g/l}$  to 39.8  
 3 mg/l). Because on the extreme sides some  $\log C_P$  intervals were missing (at the low concentration  
 4 range -2.4, -2.2 and -2.1, and at the high concentration range 1.4 and 1.5) there were 40 different  
 5  $\log C_P$  intervals. After this step, 10 024 records identified by location, year and month and linked  
 6 to a  $\log C_P$  interval remained. These records are distributed according to Figure 1.  
 7



8 *Figure 1 Distribution of 348 658 data on occurrence of macroinvertebrate genera that all share a phosphorus concentration*

9 **Treatment of abundance data.** Multiple abundance data can be present for a single site-year-  
 10 month data point. In order to obtain a single value, we applied two procedures. If for one  
 11 monitoring location in one month multiple abundance data points for a particular taxon existed,  
 12 we used the maximum number. Else, we used the sole value available. Secondly, we considered  
 13 exclusively abundance records that share location, year and month with at least one record of  $\log$   
 14  $C_P$ . As a result, the number of taxa was reduced from 2 790 to 1 746. More than 1 000 taxa with  
 15 abundance numbers are in the database, however without a  $\log C_P$  value.

1  
2 Different taxa (1 746) were aggregated to 867 different genera by taking the sum of abundances  
3 of different taxa within one genus. These steps resulted in 348 903 genera abundance records,  
4 distributed over log  $C_p$  intervals according to Figure S1 (Supporting Information). The set of  
5 abundance records with attributes location, year, month and  $C_p$  interval (0.1 log units) occupies 4  
6 % of the elements in a matrix of 867 genera records and 10 024 phosphorus records. Every record  
7 out of 10 024 records of total phosphorus concentration was linked to at least one abundance  
8 number with similar location, year and month.

9 Table S1 (Supporting Information) gives background information on the number of genera that  
10 were counted over 8 different abundance ranges (for example observed abundance numbers  
11 between 100 and 300) per  $C_p$  interval.

12  
13 **Combining abundances that share a  $C_p$  interval.** For each  $C_p$  interval the abundance data were  
14 combined. Here “combined” has the meaning of counting genera with an abundance number in a  
15 log  $C_p$  interval. A considerable number of genera (303 of the 867) appeared rare as these genera  
16 occurred less than 10 times in the whole database (see Figure S2 in Supporting Information for  
17 the number of genera per abundance range). This implies that in at least 30 out of 40  $C_p$  intervals  
18 a genus with this rate of occurrence is absent. In contrast, for example, 147 genera were found  
19 more than 500 times.

20 For each genus and for each  $C_p$  interval an abundance number (one or higher) was converted into  
21 one (presence), while zero was assigned if no abundance number was recorded (absence). The  
22 matrix of 867 genera and 40  $C_p$  intervals contains 12 954 ones and 21 726 zeros from which the  
23 “number of occurring genera” (N) was evaluated for every  $C_p$  interval simply by summing up.

1 The result is the number of occurring genera for each  $C_p$  interval. These data were analysed using  
2 the quantitative model.

3

### 4 2.3 *Quantitative model for P in freshwater*

5 To analyse the association between a (logarithmic) concentration interval of  $P_{tot}$  (hereafter  
6 denoted as  $C_p$ ) and the number of genera, we applied the concept of Species Sensitivity  
7 Distributions (SSD), developed in ecotoxicology (see Posthuma et al., 2002). This concept is  
8 commonly used to describe a sigmoidal association between a toxicant concentration in a  
9 laboratory toxicity test, and the so-called Potentially Affected Fraction (PAF) of species (or  
10 genera), based on a suite of test data for various taxa. This concept has been adapted to account  
11 for essentiality of various (essential) metals, by taking into account the natural background  
12 concentration of these metals, associated with a naturally affected fraction of species (Struijs et  
13 al., 1996). Analogously, we apply this concept to the available (bio)monitoring data by equating  
14 the maximum number of genera ( $N_{max}$ ) with 100 % genera richness and the ratio  $N/N_{max}$  as a  
15 measure of Disappeared Fraction (DF) which is a function of  $C_p$ :

16

$$17 \quad DF = 1 - \frac{N(C_p)}{N_{max}} \quad \text{Equation 1}$$

18

19 Obviously,  $N_{max}$  is lower than the total number of macroinvertebrate genera ( $N_{tot}$ ) in the data set,  
20 because genera neither do all co-occur in some  $P_{tot}$  interval nor in a natural water body. Some  
21 genera require extreme oligotrophic conditions and others are opportunistic and are able to  
22 survive severe levels of nutrient enrichment. We do not consider  $N_{tot}$  but rather  $N_{max}$  zero stress,

1 comparable to the procedure proposed in deriving risk limits for toxic but naturally occurring  
2 essential metals.

3  
4 We postulate that the function DF has a sigmoid shape on the eutrophication part on the log  $C_p$   
5 axis, analogously to the cumulative log-normal (or log-logistic) representation of the distribution  
6 of sensitivity of species for toxic substances (Posthuma et al., 2002). Here we verify if a logistic  
7 model is appropriate to predict DF by testing the linearity of the logit function of DF to the data  
8 using linear regression. This would yield the parameters  $a$  and  $b$  that describe this logistic  
9 relationship:

$$\text{logit } DF = \log \left[ \frac{DF}{1-DF} \right] = a + b \cdot \log C_p \quad \text{Equation 2}$$

12  
13

## 14 **3 Results**

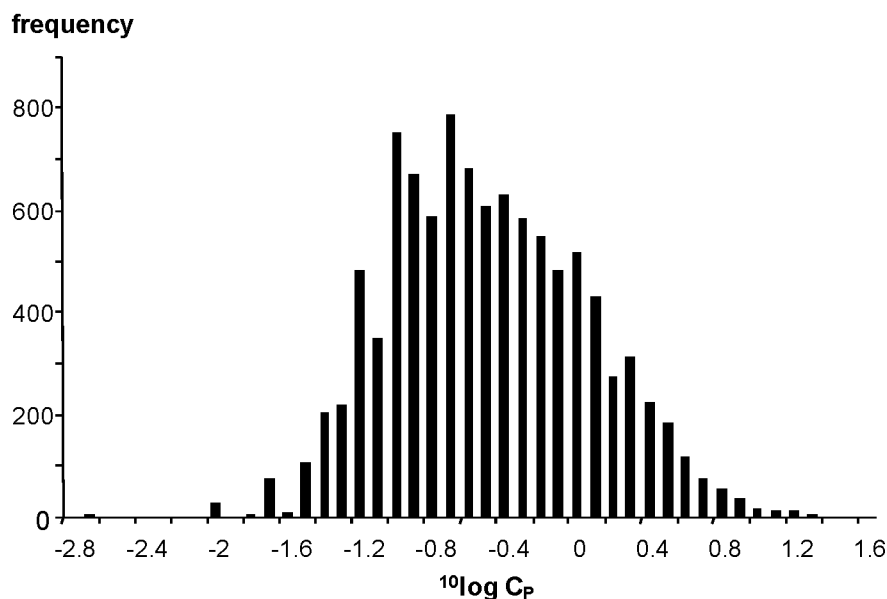
15

### 16 *3.1. Model derivation*

17 Figure 2 displays the number of genera related to each of the 10 024 records but grouped in 40  
18 log  $C_p$  intervals. Across  $C_p$ -intervals, there appears to be a dominant pattern in the number of  
19 genera, which is peaking near 0.1 mg/l with reduced highest diversity numbers towards the tails.  
20 Furthermore, the number of genera is often highly variable within specific  $C_p$ -intervals. For  
21 instance, the frame in Figure 2 encloses as many as 754 dots that all share  $C_p = 0.1$  mg/l. Each  
22 dot represents a number of genera varying between 1 and 90. Apparently, low genera richness  
23 can be found at any trophic level, a high richness, however, only at  $C_p$  values around 0.1 mg/l.

1 Between 0.03 and 0.3 mg  $P_{\text{tot}}$ /l freshwater may accommodate any number of macroinvertebra  
2 genera between 1 and 100. The latter reflects optimal environmental conditions and the absence  
3 of environmental stressors, whereas the occurrence of only one genus indicates a great influence  
4 of all other possible stressors and natural limiting environmental factors, such as low  
5 temperature. Outside the 0.03 – 0.3 mg/l ‘window’, genera diversity declines, both at de- and  
6 increasing phosphorus concentrations. At the lowest phosphorus concentrations, the declining  
7 number of invertebrate genera may be a consequence of phosphorus limitation or a response to  
8 other stress factors.

9

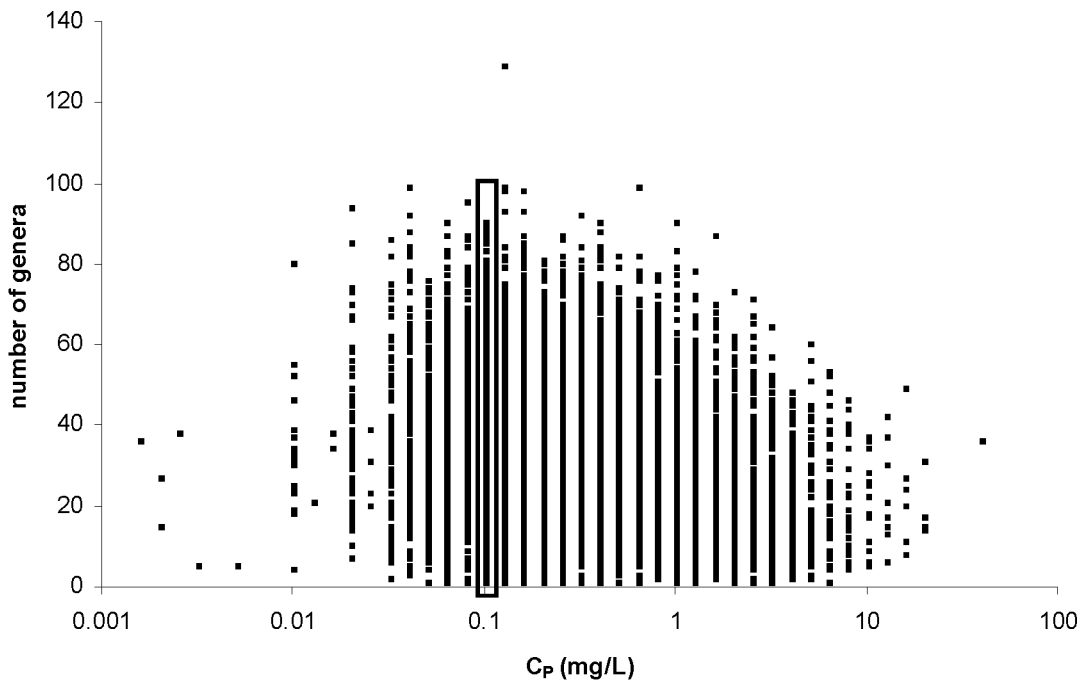


10 *Figure 2 Distribution of 10 024 records of total phosphorus concentration (mg/L) subdivided into intervals of 0.1  $10\log$  units*

11 At the very high phosphorus concentrations (when  $C_p$  exceeds 3 mg/l) macroinvertebrate  
12 diversity is considerably reduced, even if all other conditions would be optimal.  
13 The analysis of 868 genera showed that there is no phosphorus concentration interval where they  
14 all occur. There is however, an optimal concentration range between approximately 0.1 and 0.3

1 mg/l where approximately 600 different genera were counted (Figure 3). We consider this  
 2 number the reference diversity ( $N_{\max}$ ), with the absolute maximum in the bin occurring at a  
 3 phosphorus concentration of 0.1 mg/l. The decrease of the number of genera on the right side of  
 4 the curve reflects ecological damage associated to phosphorus enrichment, either directly and  
 5 solely, or as combination of phosphorus enrichments with other stressors (amongst which other  
 6 nutrients).

7



8 *Figure 3 Number of genera plotted as 10 024 data points versus the logarithm of the phosphorus concentration (mg/L); each dot represents the number of genera counted at a specific concentration (one of the 10 024) within an interval of  $^{10}\log C_p$  equal to 0.1*

9 The disappeared fraction of genera (DF) is derived from data points of Figure 3 according to  
 10 equation 1. The reference diversity  $N_{\max}$  should be interpreted as the maximum number of pelagic  
 11 macro-invertebrate genera that could be encountered in a temperate climate at a phosphorus  
 12 concentration of 0.1 mg/l, summed up over all seasons and all water types. The result is shown in  
 13 Figure 4 where for  $C_p \geq 0.3$  mg/l small circles represent DF due to nutrient enrichment.

1 The logit-model was fitted to the data in the eutrophication part of Figure 3, yielding:

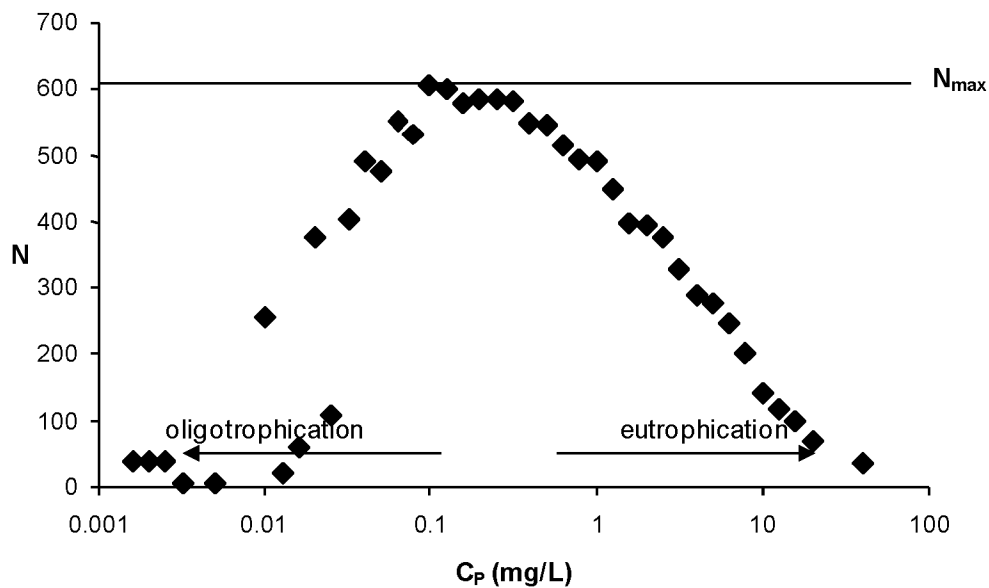
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$$\text{logit } DF = \log \left[ \frac{DF}{1-DF} \right] = 1.11 \cdot \log C_p - 0.61 \quad \text{Equation 3}$$

4

5 This is represented by Figure 4.

6



7 *Figure 4 Number of genera versus the phosphorus concentration*

8 The slope and intercept were thus derived from linear regression of 20 data points between  $C_p =$

9 0.3 and 40 mg/l. This range covers exclusively eutrophic circumstances. Figure 4 confirms the

10 linearity of Equation 3 and the parameters  $a = -0.61$  and  $b = 1.11$  characterize the logistical

11 distribution of DF (Equation 3). The explained variance ( $R^2$ ) is 99% and  $p < 0.001$  indicates an

12 almost zero chance of lacking a logistic relationship between DF and  $\log C_p$ . Equation 3 can be

13 rearranged according to:

14



1 
$$DF = \frac{1}{1 + 4.07 \cdot C_P^{-1.11}}$$

**Equation 4**

2

3 Re-plotting the data on a concentration-effect plot (both on a log  $C_P$ -scale, or on a  $C_P$ -scale) the  
4 logistic nature of DF is apparent for  $C_P$  higher than 0.1 mg/l (Figure 5). Above 0.3 mg/l  
5 freshwater is genuinely eutrophic and genera richness is increasingly affected if  $C_P$  increases.  
6 Therefore DF can be interpreted as ecological damage due to exposure of macro-invertebrate  
7 genera to phosphorus.

8

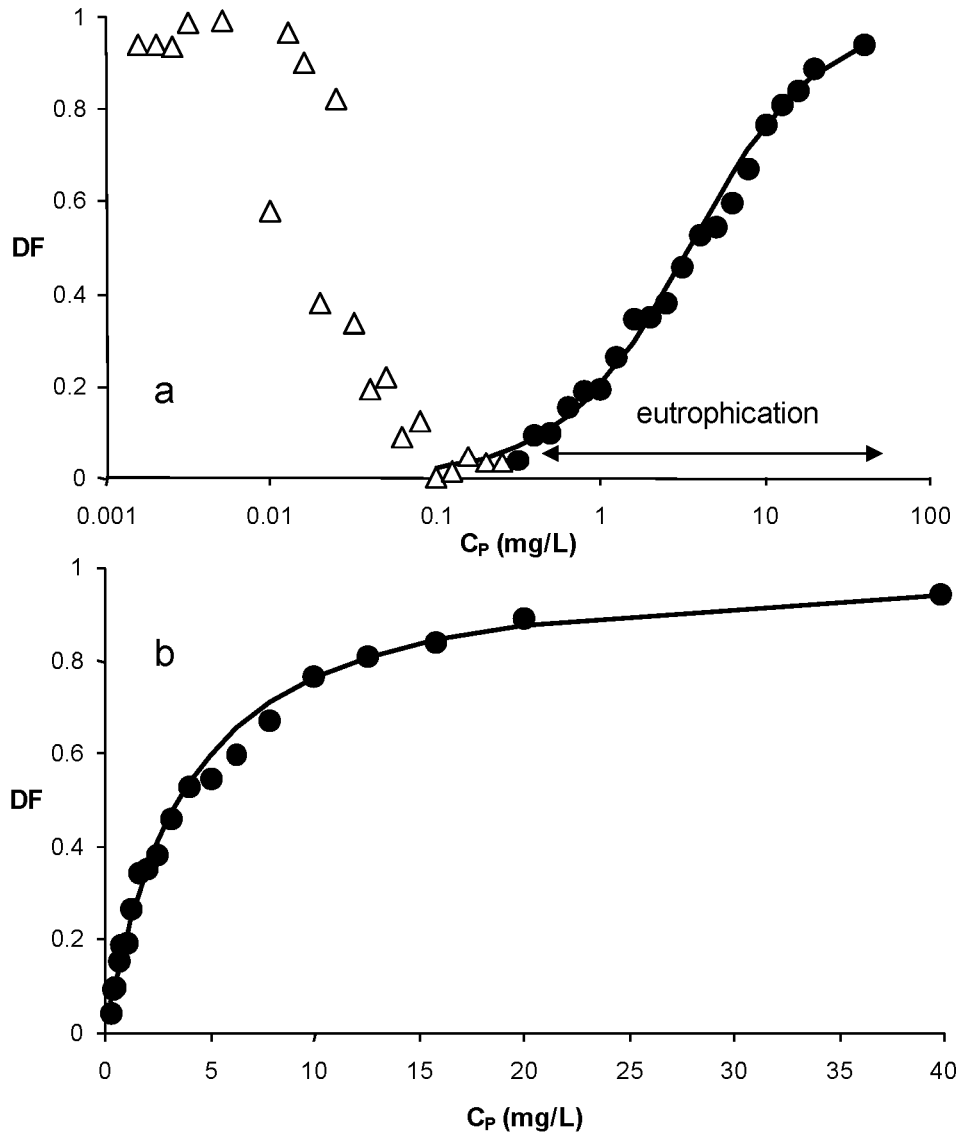


Figure 5 Empirical (circles) and fitted (drawn line) disappeared fraction (DF) of macro-invertebrate genera versus enhanced phosphorus concentration ( $\geq 0.3$  mg/L) on a logarithmic (a) and a linear horizontal axis (b). The triangles (a) are DF data not included in the fitting procedure (Figure b)

1

2

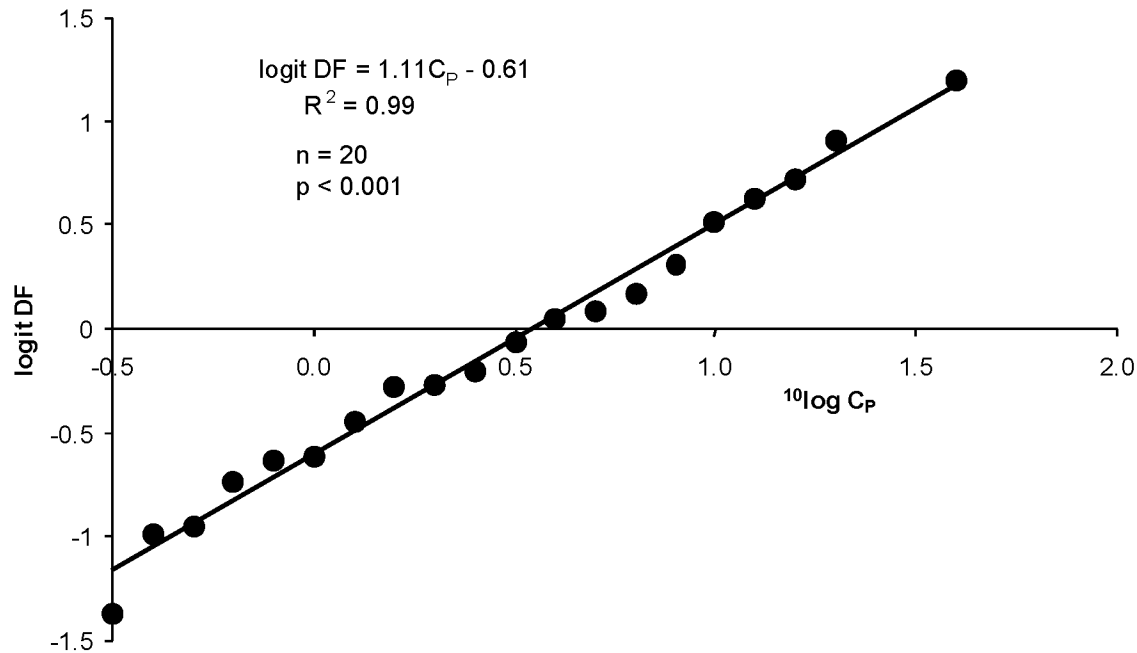


Figure 6 Logit function of DF versus  $^{10}\log C_P$  for  $C_P \geq 0.3 \text{ mg/L}$

1

2

### 3 3.2. Model application example

4 Equation 4 was applied to evaluate the potentially disappeared fraction of macroinvertebrate  
5 genera in the rivers Rhine, Meuse and Scheldt on the Dutch borders over the last decades, using

6 P-concentration measurements. The decline of phosphorus concentrations (shown in Table S2

7 Supporting Information) is associated with a decreasing trend in the potentially disappeared

8 fraction of macroinvertebrate genera. Figure 6 shows that the potential DF due to nutrient

9 enrichment in the rivers Meuse, Rhine and Scheldt was considerable in the early 70's (up to

10 30%), but gradually decreased to a level between 5 and 10 %. Since 1997 the phosphorus

11 concentrations reached a lower impact plateau in the river Rhine at an estimated DF = 0.05.

12

## 13 4 Discussion

1 The Disappeared Fraction of genera (DF) is an empirical function that describes the relationship  
2 between a stressor variable in the field and its impact on taxa loss, and has similarities with the  
3 Potentially Affected Fraction (PAF) of species based on Species Sensitivity Distribution (SSD),  
4 according to Posthuma et al. (2002). While SSD and PAF for a toxic chemical rely on differences  
5 in sensitivity of species as observed in laboratory tests, our approach has produced an estimate of  
6 a similar kind as PAF which we called DF, the Disappeared Fraction of species. We derived and  
7 illustrated the approach using data on total phosphorus in freshwater, based on (bio)monitoring  
8 data with respect to macro-invertebrate genera and the concentration of phosphorus in many  
9 freshwater samples. We considered the DF-concept useful for various purposes (see below).  
10 Within the general DF-method, some key aspects require specific attention. Although on one  
11 location at one moment the number of genera has never exceeded the value of 80, consolidation  
12 of all abundance data that share  $C_p = 0.1$  mg/l yields a number of genera (approximately 600) that  
13 could be interpreted as the maximum number of macroinvertebrate genera that could occur.  
14 Obviously, this number of genera will never be found simultaneously at one location, but rather  
15 represents the potential of these genera showing up at a phosphorus concentration of  
16 approximately 0.1 mg/l. The ratio  $N/N_{\max}$  is therefore not a location-specific yardstick that can be  
17 applied to assess the ecological health of individual aquatic ecosystems. It is rather a generic  
18 indicator for nutrient enrichment. An individual freshwater ecosystem may be subject to  
19 hysteresis (Scheffer et al., 2001) which implies that over a wide concentration range both high  
20 and very low species richness can be found, depending on the stage of nutrient loading and  
21 subsequent remediation.  
22  
23 We found a potential maximum of more than 600 macroinvertebrate genera in the concentration  
24 interval around 0.1 mg/l. This value corresponds with the findings of Smith et al. (2003), who

1 reported that natural background concentrations of phosphorus in freshwater in North America  
2 are below 0.1 mg/l. Higher concentrations are attributed to human activities. From field data we  
3 have shown that above 0.1 mg/l increasing phosphorus concentrations more and more impair the  
4 occurrence of these genera also in the Netherlands. There appears to be a surprising association  
5 between the regulatory water quality objective with respect to  $P_{\text{tot}}$  for lakes in the European  
6 Community, which is 0.15 mg/l (European Commission, 2000), and our modelling approach. Our  
7 assessment appeared to result in a similar critical value. For this concentration our model can be  
8 used to quantify a potentially disappeared fraction of macroinvertebrate genera, which is equal to  
9 4.2 %. Despite this similarity, there are still various limitations and assumptions associated to the  
10 approach we applied. Below, these issues are discussed.

11  
12 First of all, a field-based technique will conceptually never result in a real causal relationship  
13 between phosphorus concentration (or any other stressor variable) and macroinvertebrate  
14 diversity. This constitutes a fundamental source of uncertainty in the analysis and also in the  
15 application of the model to potential effects in the three rivers (Figure 6). Penning et al. (2008)  
16 used the REBECCA database to analyze species richness of macrophytes in European lakes.  
17 They concluded that the absolute value of an observed species richness as an indicator for  
18 eutrophication is of limited value, because low species richness was found across the whole  
19 trophic gradient. These findings are comparable to our analysis. However, we introduced the  
20 concept of the potentially occurring number of macroinvertebrate genera with respect to a  
21 maximum number, which revealed a generalized association between potential impacts of  
22 exposure to phosphorus and changes of taxa diversity (biodiversity).

23

1 Second, we focused on macro-invertebrates only, neglecting the potential impact of phosphorus  
2 concentrations on the occurrence of other taxonomic groups in fresh waters. However, numerous  
3 genera of macro-invertebrates occur in many types of freshwater systems, each having its own  
4 specific environmental requirements and sensitivity to stressors (Van de Brink & Ter Braak,  
5 1998; Dyer & Belanger, 1999). Macroinvertebrate diversity is therefore technically suitable to  
6 indicate deterioration of the aquatic ecosystem.

7  
8 Third, the construction of a table, containing a “binary chance of occurring” by assigning one or  
9 zero to each genus for each  $P_{\text{tot}}$  concentration interval, includes several critical steps in handling  
10 the data. For example, all abundance data within a concentration range of 0.1 log unit were  
11 aggregated, regardless location or time. Records referring to the wintertime, when only few  
12 macrofauna species are observable in temperate zones, were also included in the analysis. In the  
13 current study, we deliberately took as many circumstances as available in the database into  
14 account, anticipating that the derived damage function (DF) would then be based on as much  
15 data as possible, while being applicable to analyze “national” environmental problems, like an  
16 assessment of the potential biological relevance of water quality management for the large rivers  
17 over time in terms of fauna diversity. It is acknowledged that, given the same approach,  
18 environmental problem settings could focus on certain water types (e.g., lakes). This might result  
19 in another DF-function, and hence refined analysis results (e.g. another critical value for  
20 naturally oligotrophic lakes). In other words: choosing the “training set” should relate to the  
21 problems that need be solved. Our analyses currently focus on a large-scale dataset, covering a  
22 wide range of conditions and water bodies, so as to derive a damage function, to be applied e.g.  
23 in decision support tools, that would stretch beyond Dutch borders. Struijs et al. (2010) have  
24 applied this concept in Life Cycle Impact Assessment of industrial products, with regard to

1 assessing the relative biological relevance of emission of substances that could lead to  
2 enrichment of inland waters in temperate zones (European Union). This problem definition is  
3 highly relevant to help designing industrial products with low environmental impacts, but does  
4 not “allow” for (water-body) specific details.

5  
6 Fourth, although  $N < N_{\max}$  may be interpreted as loss of genera, at  $C_P < 0.1$  mg/l this does not  
7 necessarily reflect natural nutrient deficiency only. It could be the result of anthropogenic  
8 interventions, for example it may represent a pristine oligotrophic ecosystem affected by  
9 acidification (Leuven et al., 1987; Leuven 1988). Furthermore, a combination of low  $C_P$  and  
10 relatively high species diversity may also occur in spring when surface water rich in species may  
11 almost entirely lose its phosphorus due to a fast uptake by aquatic macrophytes or algae.

12

## 13 **5. Conclusion**

14

15 The following targets were reached:

- 16 1. Conceptually associated to the Species Sensitivity Distribution model as applied in the risk  
17 assessment of toxic chemicals, it appeared possible to derive a general model to describe a  
18 Disappeared Fraction (DF) of taxa as a function of a stressor variable, based on  
19 (bio)monitoring data.
- 20 2. The general model derivation was applied to assess ecological damage due to nutrient  
21 enrichment (especially phosphorus) in temperate inland waters of the Netherlands. Macro-  
22 invertebrates constitute an important fauna group in aquatic ecosystems. A major stressor of  
23 freshwater ecosystems in temperate regions is the phosphorus concentration, especially in  
24 densely populated and industrialized areas and regions with high agricultural production.

1 From field data we have shown how increasing phosphorus concentrations above 0.1 mg/l  
2 impairs the diversity of macroinvertebrate genera. In this case, the DF-model was:  $DF = 1/(1$   
3  $+ 4.07 \cdot C_P^{-1.11})$ . The analyses for phosphorus yielded a critical phosphorus concentration very  
4 close to the current regulatory water quality objective for phosphorus (0.15 mg  $P_{tot}/l$ ) in the  
5 Water Framework Directive of the EU. According to the derived function, this value  
6 corresponds to a potential biodiversity loss of 4.2 %. Moreover, application of the function to  
7 the water quality of the rivers Rhine, Meuse and Scheldt suggested that the reduction of  
8 phosphorus loads in these rivers during the last decades would be associated to lower values  
9 of DF, towards a possibly realistic plateau of low impacts.

- 10 3. Application of the model derivation approach is useful in various contexts, amongst which  
11 decision support systems such as life cycle assessment and multistress analysis of freshwaters  
12 in temperate regions.

13  
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15 for our study.

16



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