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**French Expertise Report (2015-12-21) for the 'VLRs'  
GIG: IBD 2007: Context, abiotic variables included,  
pressure-impact relationships**

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## “Very Large Rivers” GIG

### *French Expertise Report:*

#### **IBD 2007: context, abiotic variables included, pressure-impact relationships**

Also see :

-The Master 2 report: Boutry, S., 2006. « Application de méthodes statistiques pour la mise en œuvre d'une nouvelle version de l'Indice Biologique Diatomées ». Université Paul Sabatier Toulouse 3 – Cemagref, 30 pages + annexes

- The scientific paper : Coste, M., Boutry, S., Tison-Rosebery, J., Delmas, F. (2009). Improvements of the Biological Diatom Index (BDI): Description and efficiency of the new version (BDI-2006). Ecological Indicators, vol. 9, n° 4, p. 621-650.

#### **1) Context, general elements :**

A first version of the IBD or Indice Biologique Diatomées (Lenoir et Coste, 1995; Prygiel et Coste, 2000) has been set-up in 1995, then French AFNOR-standardized in 2000. In France, this first version was commonly named IBD 2000.

IBD 2000 has then been routinely applied in the French monitoring networks for the routine monitoring from 2000 to 2006. It included 209 taxa profiles, 173 of them being true taxa and 36 profiles having been built on a regroupment of easily-confounded taxa.

Between 2006 and 2007, with the consequent increase of available biological and abiotic data in the French national databases, a renewed version of IBD (IBD 2006) has been worked and then produced, based on an increased number of constitutive taxa (836) and also including profiles for observed species exhibiting teratogenic deformities. This second version has been French AFNOR-standardized in 2007. From then, it is commonly named IBD 2007.

The national diatom assessment system of rivers using IBD 2007 has been published at the French Official Journal, then intercalibrated at the EU level. Depending on their location and their typology, most of the French rivers were part of the Central-Baltic GIG; the rivers located in high mountains (Alpes, Pyrénées) were inter-calibrated within the Alpine GIG; the rivers located in the Mediterranean zone with the Mediterranean GIG.

Following an evolution of the French national assessment system, the new French evaluation system has been re-intercalibrated during the 2<sup>nd</sup> Intercalibration Round.

#### **2) Methodological aspects of the IBD construction :**

Whatever the IBD version, the automated calculation of the index is done by extracting the ecological profiles (following Daget & Godron, 1982) of the contributing taxa along 7 quality classes. The final note is the barycentre of the presence probabilities of the “fictive taxon” through the 7 quality classes.

The calculation formulas, unchanged between the 2 versions, are the following ones:

$$P_{\text{class}_{(i)}} = \frac{N_{(i)} \times A_{(i)}}{N_{\text{sites}} \times S_{\text{om}}}$$

$$S_{\text{om}} = \sum_{i=1}^7 \frac{N_{(i)} \times A_{(i)}}{N_{\text{sites}}}$$

Where:

- P class\_(i) is the species presence probability for water quality class (i);
- N (i) is the species occurrence number for water quality class (i);
- A (i) is the species cumulated abundance in %;
- N sites (i) is the number of sites within the water quality class (i).

The 2 successive versions of IBD have been built on a composite gradient based on 7 man-impacted variables (see the list in Table 1).

In the case of IBD 2000, for memory, the composite gradient was integrated through a co-inertia analysis linking the axis wearing the maximum inertia of the abiotic dataset, with that one of the biological dataset.

For IBD 2007, the composite gradient has been built using a Principal Component Analysis (PCA). The component loadings of the variables on the 3 first axes of the PCA are summarized in Table 1:

<b>Table 1 – Component loadings on axis 1, axis 2 and axis 3.</b>			
Parameter	Axis 1	Axis 2	Axis 3
pH	-0.084	0.388	0.113
Conductivity	0.164	0.208	-0.597
BOD	0.342	0.069	-0.408
O <sub>2</sub> (%)	-0.31	0.196	-0.111
NH <sub>4</sub>	0.412	0.049	0.356
PO <sub>4</sub>	0.411	0.028	-0.121
NO <sub>3</sub>	0.066	0.066	1.768

Axis 1 and Axis 3 are mostly under the structuring influence of man-impacted abiotic descriptors, Axis 2 primarily integrates physico-chemical information under natural influence.

So the composite anthropogenic gradient has been built using the information of:

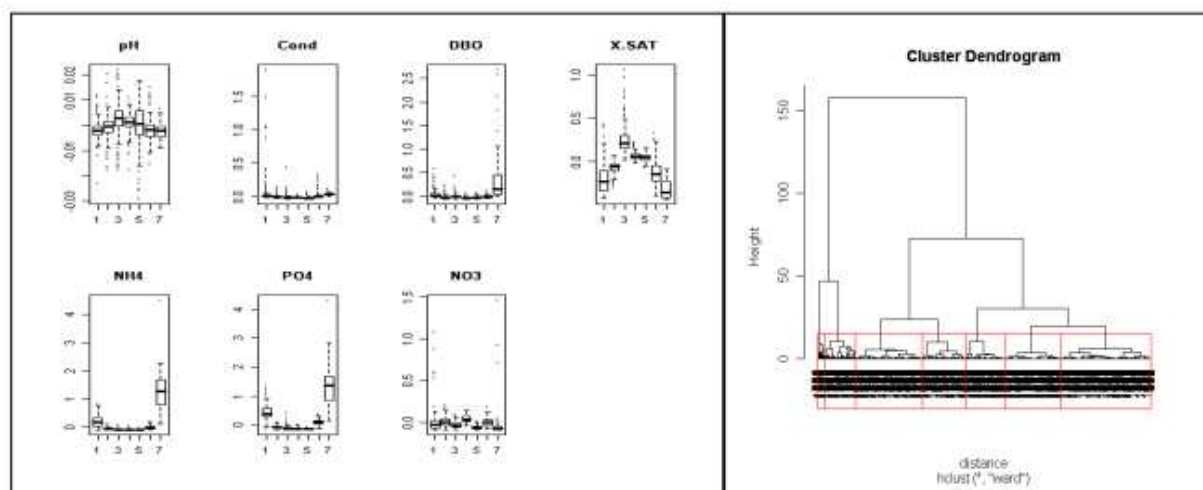
- **Axis 1**, which wears a positive structuring information from NH<sub>4</sub>, PO<sub>4</sub> and BOD<sub>5</sub> and a negative structuring information from O<sub>2</sub> Sat. (%);
- **Axis 3**, mostly influenced by NO<sub>3</sub> (+), Electric Conductivity (-), BOD 5 (-), NH<sub>4</sub> (+).

Hydro-chemical information worn by Axis 2, mostly caused by natural factors due to geochemistry of substrates and to oxygenation linked to river slope, has been removed and didn't serve to build the composite anthropogenic gradient.

The weight assigned to each parameter for the definition of the seven water quality classes was determined as follows (Table 2):

Table 2 – Parameter weights for the definition of the seven water quality classes (BOD: biological oxygen demand).		
Parameter	Weight on axis 1	Weight on axis 3
pH	0.061	0.009
Conductivity	0.122	0.048
BOD	0.253	0.033
O <sub>2</sub> (%)	0.230	0.009
NH <sub>4</sub>	0.305	0.029
PO <sub>4</sub>	0.305	0.009
NO <sub>3</sub>	0.049	0.143

Weight = ( % of variance explained by axis 1 ) \* (correlation coefficient between axis 1 and the parameter) \* (parameter value) + ( % of variance explained by axis 3 ) \* (correlation coefficient between axis 3 and the parameter) \* (parameter value).



**Figure 1: Hierarchical Cluster Analysis (HCA) to set-up the quality classes (right); box-plots per abiotic variable obtained (left)**

The cluster tree obtained is shown in the right part of Figure 1 above. The distribution of each hydrochemical variable per cluster is shown in the left part of Figure 1.

Using a PCA based on the values of the 7 abiotic variables found at each site, correspondence between the clusters obtained through the HCA and the quality classes is the following (Figure 2 on next page):

Taking into account the 7 abiotic variables, the composite quality classes obtained extend along the Axis 1, from Class 7 corresponding to Cluster C on the left (best qualities), to Class 1 corresponding to Cluster G on the right (the worst qualities).

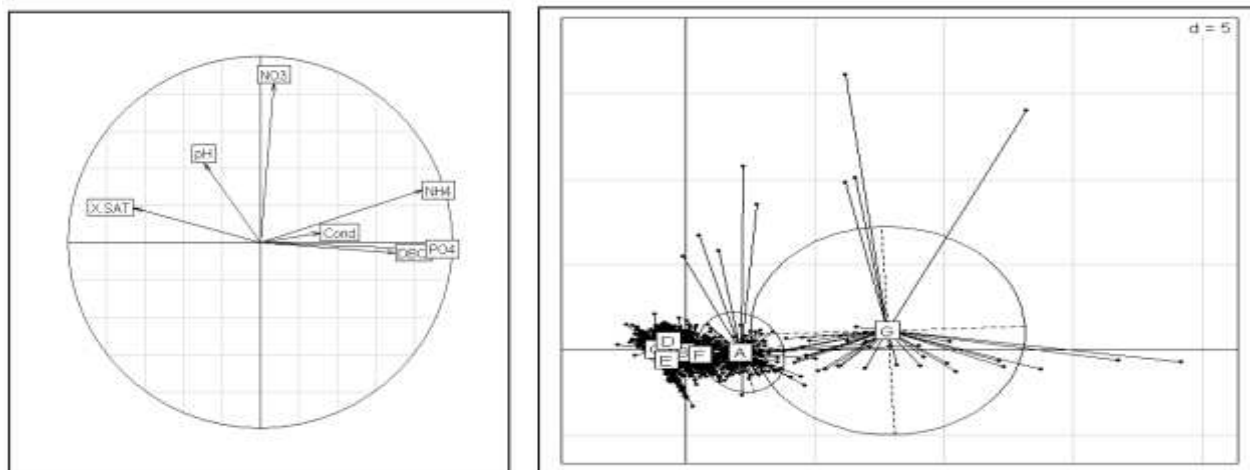


Figure 2: Regroupment of the sites into 7 quality classes, using a PCA based on the abiotic variables

### 3) IBD 2007 performance assessment :

At the general scale of France, every river type regrouped together, we assessed the biological performance of IBD 2007, compared to the previous one (IBD 2000) and to IPS, on a biological dataset of 7722 diatom inventories available in 2007. The result is shown below (Figure 3).

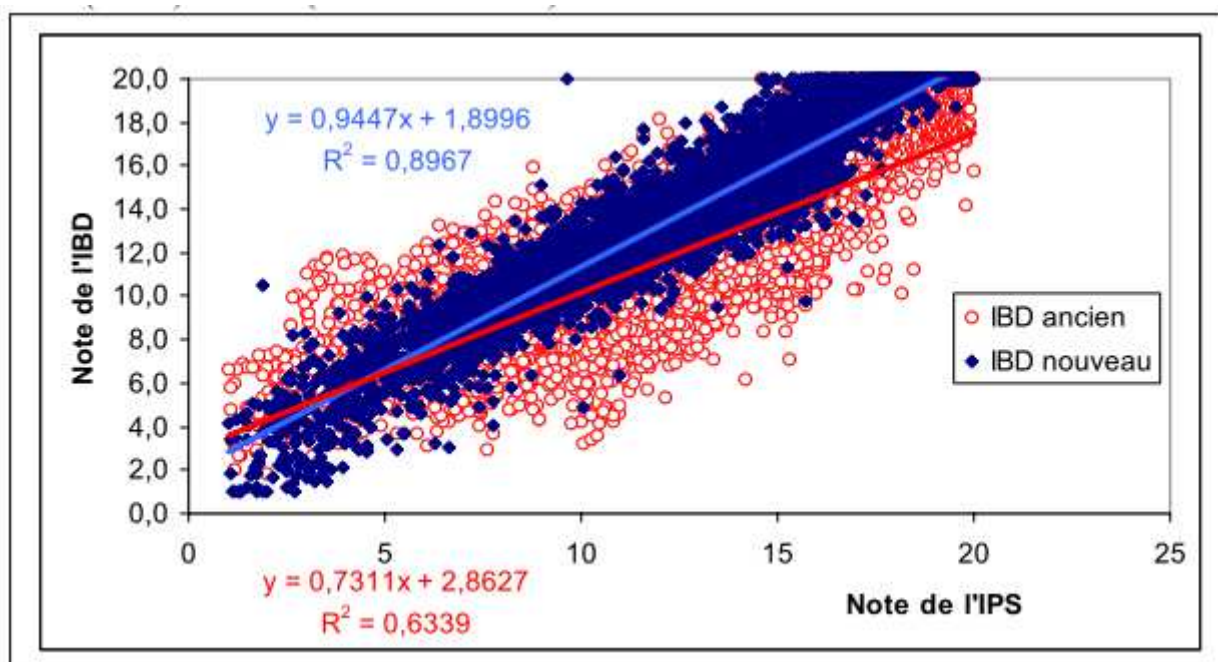


Figure 3: Linear relationships between the 2 successive versions of IBD, and IPS

We can observe that the response of the new IBD (IBD 2007, in blue) is consistent with that of IPS, which is a kind of “reference index” at the EU level, with a  $R^2$  close to 0,9. It performs better than the previous version of IBD (IBD 2000). This correlation has been obtained with the complete raw dataset (without removing any sample which could be considered as outlier...).

As indicated above, IBD 2007 has been built, and its response finely tuned, taking into account a **composite anthropogenic gradient** summarizing man-made disturbance.

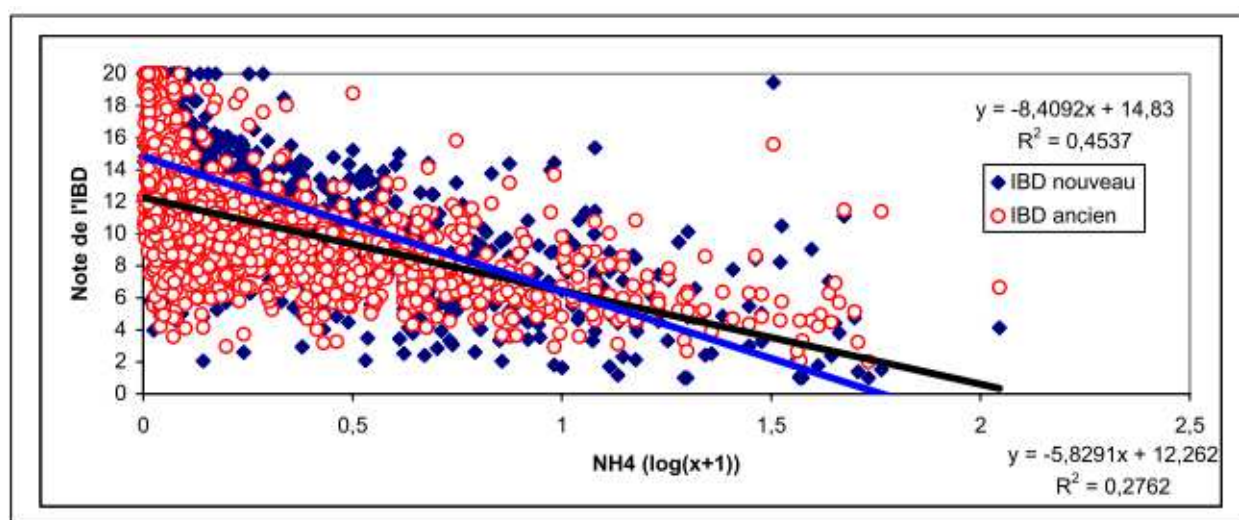
It is of course possible to assess its pressure-impact relationships with unique chemical anthropogenic stressors (e.g. PO4, BOD5...) taken into account the one after the other.

Nevertheless, it could be considered as normal that its biological response, built under the integrated influence of the 7 above-cited chemical descriptors, wouldn't be in perfect accordance with the separate effect of each single pressure.

Anyway, we can present below a few pressure-impact relationships which have been produced in 2007, coming in support to the change of version of our national diatom index (replacement of IBD 2000 by IBD 2007).

As for the revision of IBD, all biological samplings taken into account for these pressure-impact studies were collected during the low flow period of the warm season (between June and September-October) in every type of French rivers, from the smallest ones to the very large ones.

**744 samples** have been used, coupling biological counts and a complete set of abiotic descriptors. No outlier has been previously removed. To ensure a correct consistency of the time-representativeness between the biofilm samples and the abiotic data integration, mean values of each abiotic variable were calculated on 2 months' data: from 45 days before the sampling date to 15 days after.



**Figure 4: Pressure-impact relationships between the 2 successive versions of IBD and log (NH4+1)**

Even if several  $\text{NH}_4^+$  outlier values come altering the quality of the relationship, that is quite logical as explained above ( $\text{NH}_4^+$  rarely acts alone on biological communities but together with other anthropogenic stressors; the result of one or two grab samplings of water is weakly time-integrative ...), BDI 2000 showed a clear relationship with this nutrient. Even without any removal of outliers, IBD 2007 reached at least the same quality of relationship with this nutrient as the IPS ( $R^2 > 0,45$ ). This determination coefficient could quickly and strongly progress with the removal of a given percentage of the strongest outliers.

Concerning  $\text{PO}_4^{2-}$ , even keeping in mind that IBD has been constructed on a more complex set of anthropogenic variables, the relationship with its 2 successive versions can also be shown (see Figure 5) and globally follows the same pattern (linear model with values of  $\text{PO}_4$  log-transformed). Here also, IBD 2007 (in blue) exhibits a stronger slope and a better determination coefficient than IBD 2000 (red dots, black regression curve), justifying the change of the previous version by the new one at the national level.

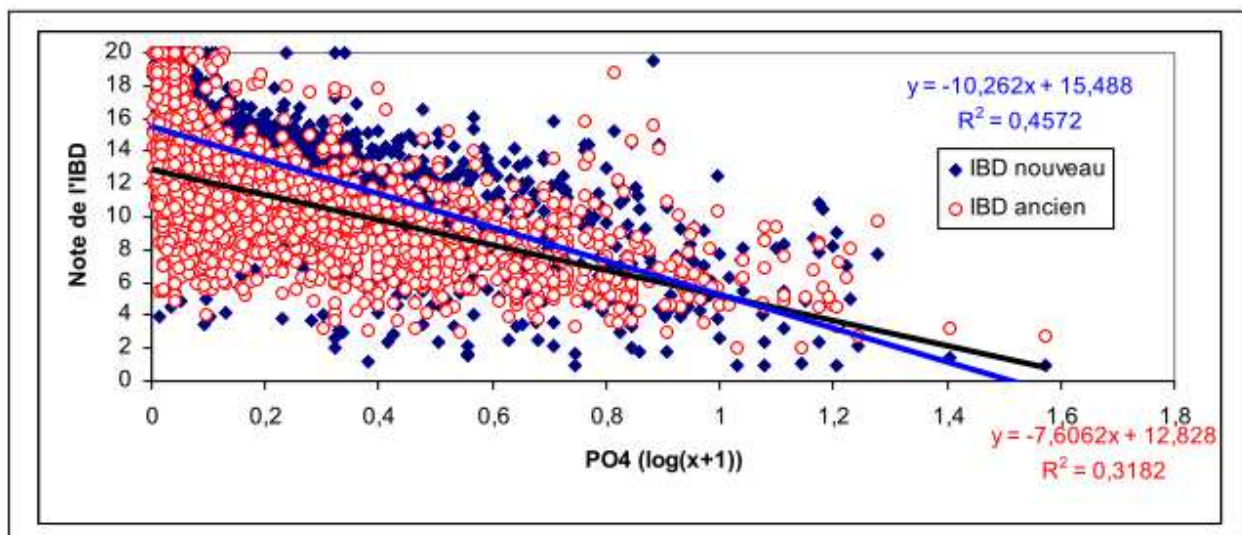


Figure 5: Pressure-impact relationships between the 2 successive versions of IBD and log (PO4+1)

Using a selection of french sites newly extracted from the national dataset in the frame of the current « Very Large Rivers » Intercalibration Exercise, we tried to quickly realize several tests in order to show if pressure-impact relationships between IBD 2007 and nutrients could also be shown in these specific types of hydrosystems.

The biplot presented in Figure 6 below has been constructed on a selection of 523 French samplings done on rivers of more than 8 000 km<sup>2</sup>, which couple availability of IBD 2007 index notes, and that of abiotic descriptors of anthropogenic pressure.

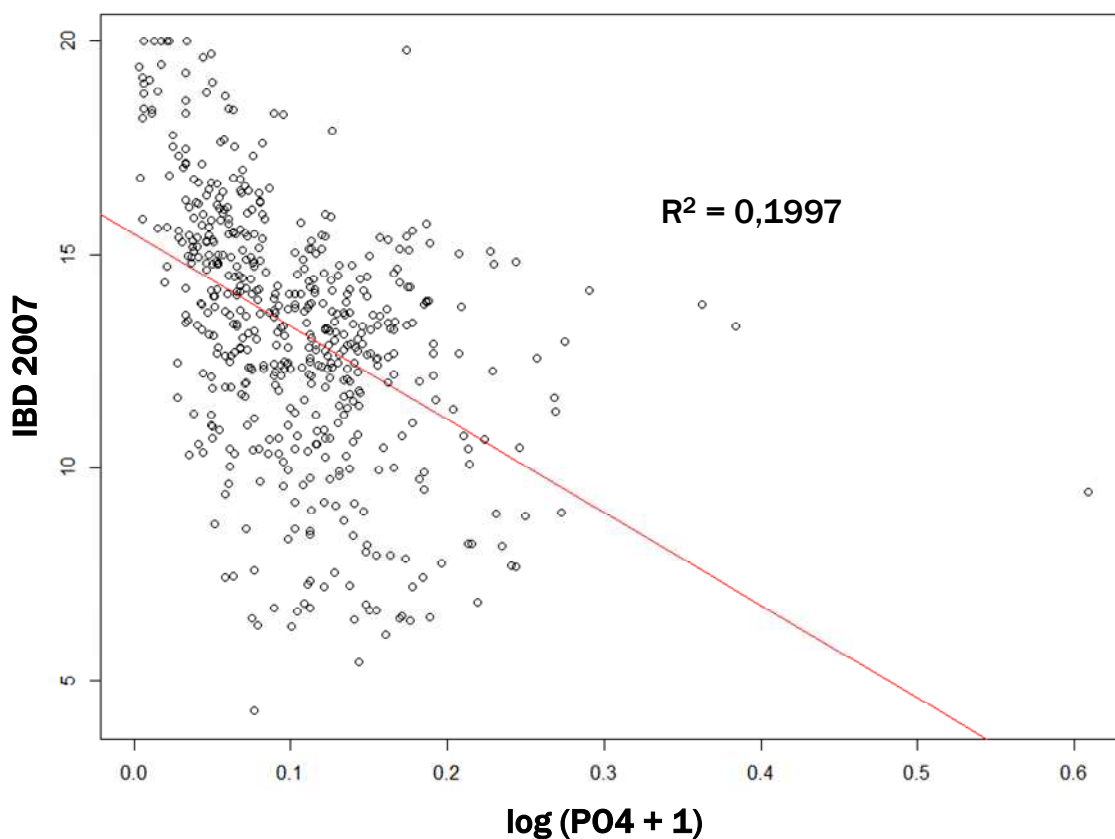


Figure 6: Linear relationship between IBD 2007 and log (PO4 + 1) (annual mean) in very large rivers

If a relationship can be drawn between these 2 variables, it looks poorer than the previous one shown in 2007 on the base of all types and all sizes of rivers. Several reasons may contribute to that result:

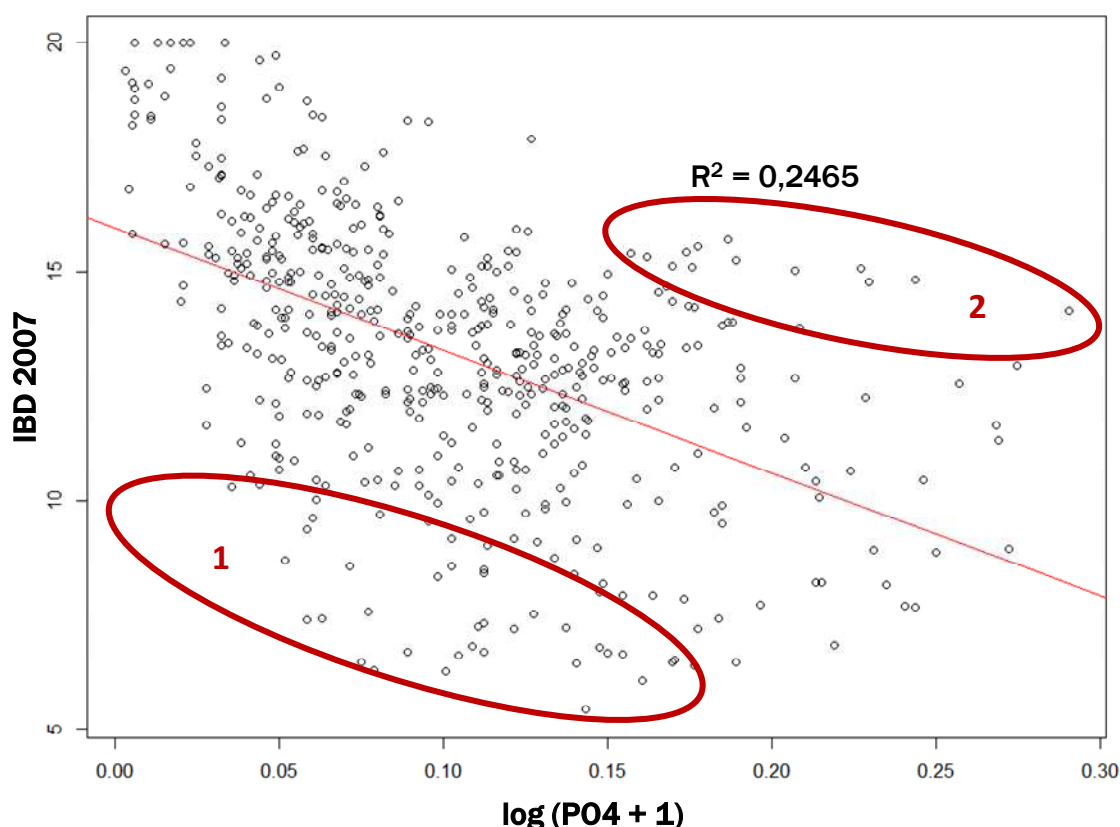
-as already said, by construction, the response of IBD 2007 is composite and integrative of 7 different pressure variables. That index being sensitive to these 7 descriptors, the response to only PO4 is not fully representative of the real level of integrated anthropogenic pressure in these 523 study cases.

- as it can be seen through the reduction of range covered by the scale of  $\log(\text{PO4}+1)$  in the 2 comparable biplots of Figure 5 and Figure 6, the selection of only VLR, even in an enlarged way (8 000 km<sup>2</sup> instead of the 10 000 km<sup>2</sup> threshold for the collective intercalibration exercise) strongly reduces the PO4 gradient covered.

- Furthermore, a benthic diatom sample is time-representative of an integrated quality of water flowed during 2 to 3 months at the maximum. Working with yearly mean is then poorly representative of the real integrated water quality to which the diatoms have been submitted during the growth of a summer biofilm.

Considering the elements listed above, we tried to remove the most outlier values of the PO4-IBD relationship presented Figure 6, in order to assess the progression of the relationship's quality with the removal of the less linked dots.

With an automatic removal of the five « worst » outliers out of 521 (under R software), the progression of the relationship is such as follows (Figure 7).



**Figure 7: Linear relationship between IBD 2007 and  $\log(\text{PO4} + 1)$  (annual mean) in very large rivers after the removal of 1% of outliers**

The automatic selection of outliers asks question, as 3 of the 5 data removed shorten by 2 the scale of PO4 values (and are perhaps not true outliers considering this IBD 2007 / PO4 relationship). .

Anyway, even if the relationship strenghtens moderately with the removal of 5 outlier values, it may be qualified of moderate. The main reasons have already been evoked :



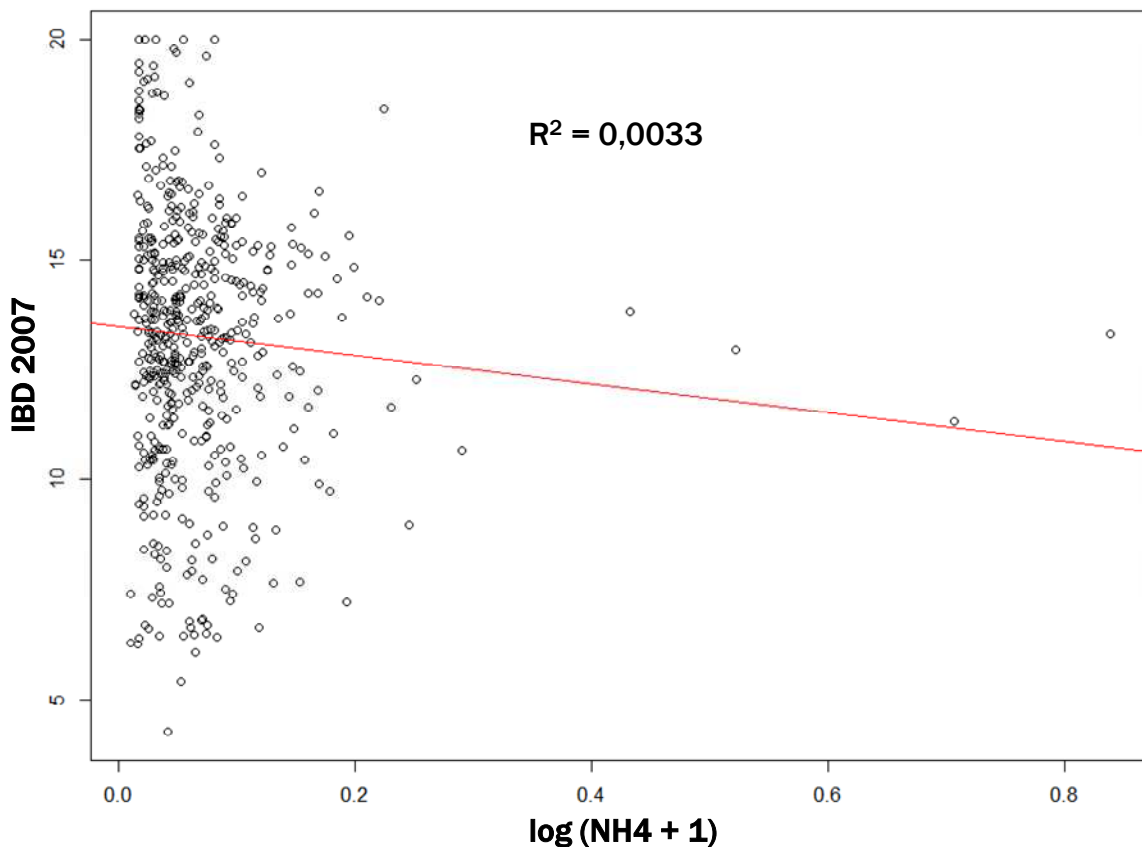
- one single pressure descriptor taken into account in that case (the biological response of IBD 2007 has been derived from 7 different descriptors)

- annual mean of PO<sub>4</sub> /vs trimestrial integration of water quality by the specific composition of benthic biofilm).

Concerning the best way to remove outlier values, one can notice that the linear regression, if really due to PO<sub>4</sub> concentrations, should cross the Y axis in a sector close to the IBD note of 20. The most abnormal dots towards this relationship are those which are figured in the sector of the ellipse Nr.1 (very low to low values of PO<sub>4</sub>, which cannot be responsible to the low level of the index note in such cases). Probably, what explains these abnormal values is high values of anthropogenic pressure due to **other hydrochemical descriptors**, like BOD<sub>5</sub>, NH<sub>4</sub>, Kjeldahl N, oxygen depletion in situ (% Sat O<sub>2</sub>).

Taken into account the problems of time-representativeness due to grab sampling in the water column, it is possible that several values inside the ellipse Nr 2 were higher at the moment of the spot sampling than the mean exposure to orthophosphates during 2 to 3 months.

We also tried to figure out the relationship between IBD 2007 and NH<sub>4</sub> values (log-transformed).



**Figure 8: Linear relationship between IBD 2007 and log (NH<sub>4</sub> + 1) (annual mean) in very large rivers**

If a relationship may exist, 1) it is quite poor without removal of the less explanatory values ; 2) other anthropogenic descriptors also make influence and generate noise ; and 3) the actual determination coefficient is very low and not satisfying.

3 main reasons can be evoked :

- The annual mean of NH<sub>4</sub> is visibly not enough representative of the real exposure condition at the precise season where the biofilms of the samples were exposed,

- In such large rivers, in general, it is difficult to find high to very high values of NH<sub>4</sub> because of big sewage plants regulations; of the dilution of sewage outlets; and of mineralization and nitrification processes in situ which drive to moderate concentrations. The gradient is then less extended than in small-to-medium-size rivers.
- Like already said, in a comparable sector as for the ellipse N<sub>r,1</sub> in the Figure 7, some very low index values cannot be explained by the low to very low values of NH<sub>4</sub>, but by other pressure parameters, and a selective removal of outliers in this precise sector would undoubtedly comfort the slope of the model and the determination coefficient.

Anyway, these relationships are a lot poorer than :

- on a national dataset composed of all types of rivers (see Figures 4 and 5),
- with an analyse based on 2 to 3 month 'mean values of nutrients (here, yearly mean values)

For sure, the use of trimestrial mean values could improve the relationships ; the removal of some low values of nutrients also, if a strong value in other anthropogenic descriptors allows us to do so.

We could also try to test the relationship between IBD 2007 and the composite pressure gradient with the precise set and weights of anthropogenic variables which have been used for its development.

Perhaps quantile regressions could also help to a better demonstration of pressure-impact relationships...

We will then continue to study these data in the next months in order to try to progress in that field.

#### **4) Links between IBD 2007 scores and the modification of autecological attributes of communities towards PO<sub>4</sub> (trophy) :**

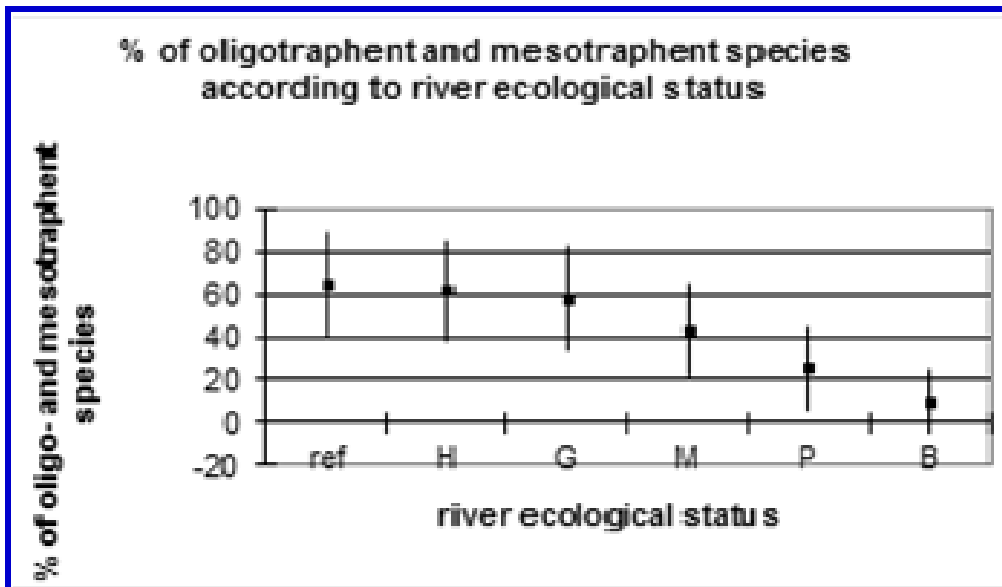
##### ***4-1) All national types of rivers mixed :***

As already described in the previous technical note, in order to validate the performance of the new index, we verified the consistency between the index notes and the repartition of the autoecological traits of the multispecific communities, following van Dam (1994), in reference condition and along the ecological status gradient summarized by the five quality classes (from high to bad status class).

These test were done on a french dataset of 845 samplings collected in all the types of rivers (from the very small ones to the very large ones).

The Figure 9 presents the percentage of sensitive species ('oligotraphent' + 'mesotraphent' species) in reference conditions and along the ecological status gradient. This graph shows:

- no significant difference in sensitive species percentage between reference condition and high status;
- a very slight but significant decrease of sensitive species between high and good status; nevertheless, the relative abundance of sensitive species still remains dominant (median value).
- a drop in the percentage of sensitive species between good and moderate status, illustrating a clear shift of communities in response to anthropogenic degradation of the aquatic environment.



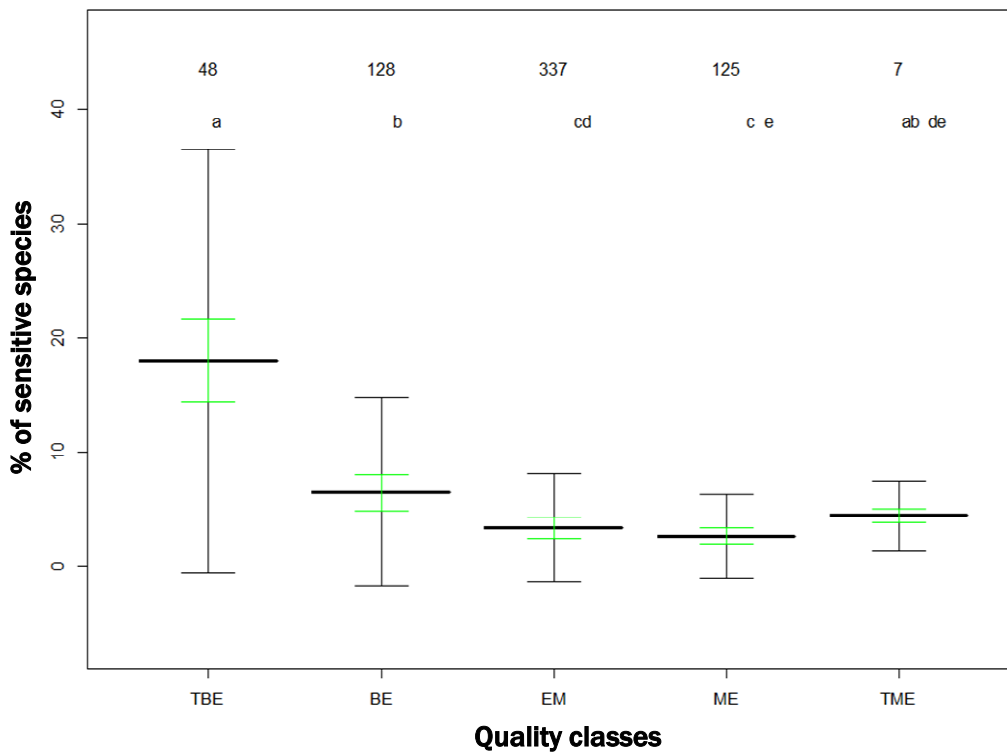
**Figure 9: Trophic characteristics of diatom communities in reference sites and along the complete gradient of water quality classes (from high to bad status)**

The limit between good and Moderate status looks then properly tuned, in agreement to the WFD normative definition of Good Status.

**4-2) All national types of rivers mixed :**

During the last 2 weeks, we just initiated a work which aims at verifying the consistence of the biological classification with IBD 2007 and the composition of communities in terms of autoecological traits, on a data subset composed of 645 diatom samplings only collected in Very Large Rivers (selection threshold: watershed > 8 000 km<sup>2</sup>).

**Trophy (P04)**



**Figure 10: Percentage of PO4-sensitive species per quality class in very large rivers (> 8 000 km<sup>2</sup>)**

Following a quite similar procedure as previously proceeded on the French complete national dataset in 2007 (Figure 9), we obtained a sensibly different level of autecological attributes per quality class (Figure 10).

For the VG and G classes, the levels of trophy-sensitive taxa are sensibly lower than for the previous composite dataset made of all types of rivers:

- The first reason is probably that in VLRs, no real reference site still exist and probably also, an inferior proportion of high status sites exist, mostly distributed in the lowest half of the class.
- Another possible reason, following the river continuum concept proposed by Vannote (1980), emerges from the assumption that the structural and functional characteristics of communities in rivers are adapted to comply with the most probable position, or mean state, of the physical system at the site. In natural hydrosystems, mean physical conditions and mean trophic condition are then at a different level at the upstream and downstream, with a normal increase of natural trophic level to the downstream. In our national dataset only composed of VLRs, none of them being out of anthropogenic influence, this natural trend is increased by diverse kinds of anthropogenic pressures (*e.g.* diffuse agricultural pollution, cumulated effects of big cities and sewage plants along the rivers...). This could also be a valid reason why we can observe such a difference of trophic autecological attributes of communities per quality class.

We could also suspect a methodological issue. In the 2 previous Figures (9 and 10), the trophic Class 7 of van Dam (from oligotrophic to hypereutrophic) corresponds to taxa which couldn't be classified in terms of trophy, or which were unknown in the biogeographical conditions of the Netherlands in 1994. In the Figures 9 and 10, in a comparable way, these taxa were regrouped with the tolerant species, that could influence the general autecological shape of communities.

We couldn't easily access again to the exact dataset, and to the exact taxa attributes' tables in Omnidia, used in 2006-2007. But, on the VLR dataset, we further investigated the distribution of undetermined species (Class 7) through the quality classes.

### Trophy (PO4)

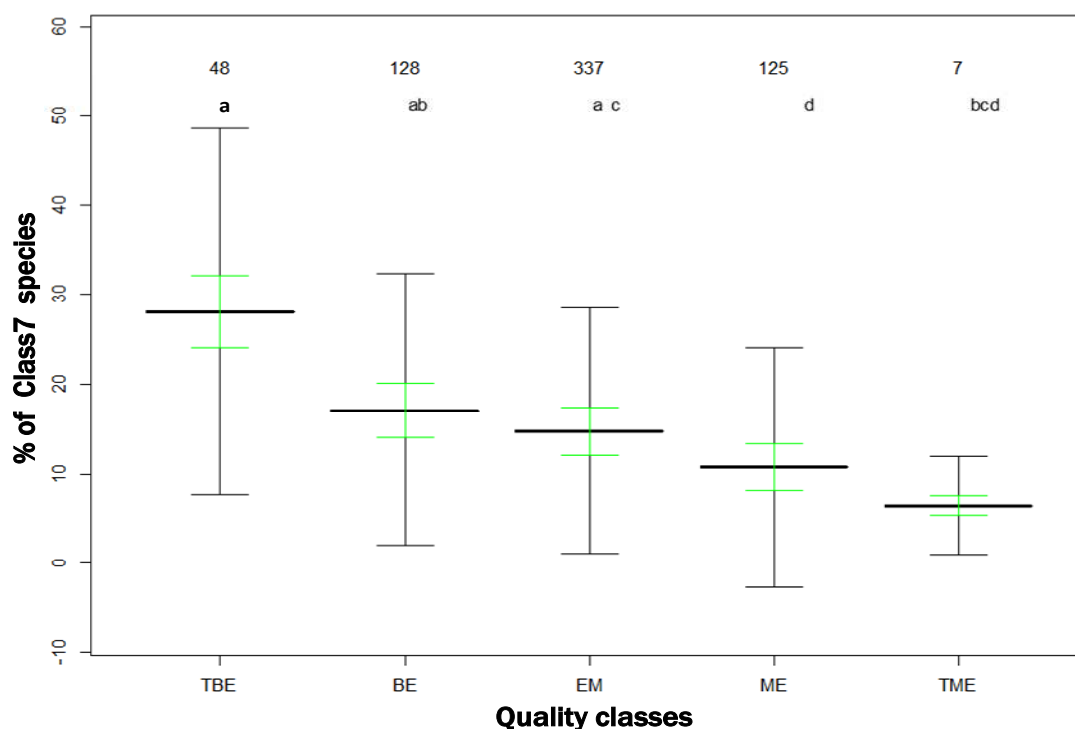


Figure 11 : Distribution of Class 7- species (undetermined towards trophy) along the 5 quality class gradient

It is possible to observe that a very significant quantity of undetermined taxa exists in our VLRs dataset and that their distribution is not balanced between classes, but very significantly stronger in high (> 28 % in mean) and in good status (> 17% in mean), than in poorer status classes.

This gradient of undetermined species works at the inverse of the gradient of sensitive species. It quite strongly influences the results presented in Figure 10, with the effect of artificially diminishing the level of trophy-sensitive taxa in the H and G status classes.

Probably, in the Netherlands, van Dam never found most of the species contributing to the typical communities of High and of Good status-VLRs in France (the best quality rivers being Dordogne and Durance, which directly come from high elevation places in the “Massif Central” and Alps respectively. At the inverse, he has qualified the autecology of most of the tolerant taxa that we also find in France, illustrating the fact that this category of species is easier to be found into hydrosystems of the Netherlands.

In order to solve this biogeographical problem, the only efficient solution would be to do again the type of analysis done by van Dam on the complete French dataset, which covers more diverse types of natural conditions than this one of Netherlands which is a flat country, and to derive the autecological characteristics of a sensibly increased number of species.

If we are aware of the interest of doing again such a work, it is impossible to achieve it in a short time and to present modified results in such a way, because our national chemical dataset needs a quite heavy post-processing before being available in that purpose.

We also tried to see what shape would show the results by proceeding in an inverse way by summing the increase of trophy-tolerant species (PO4), including the undetermined species of trophic Class 7 (Figure 12).

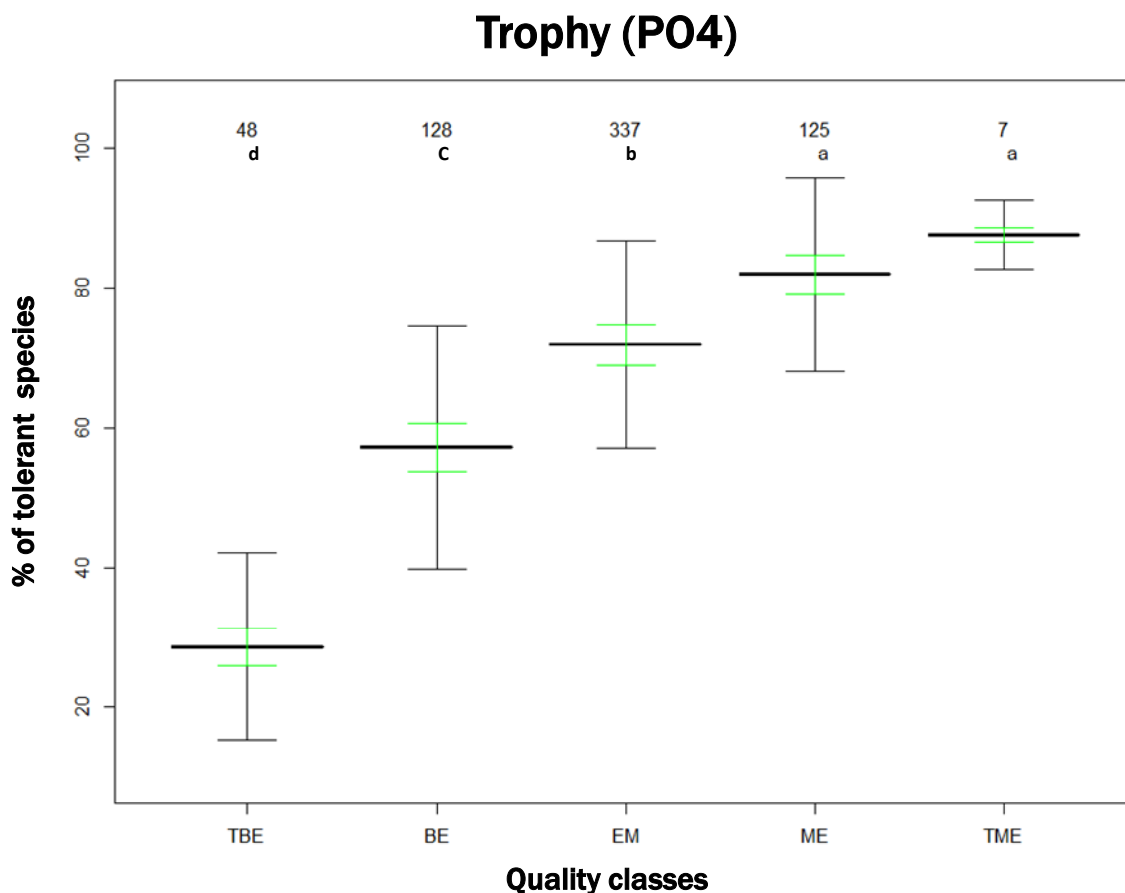


Figure 12: Percentage of PO4-tolerant species per quality class in very large rivers (> 8 000 km2)  
To be modified

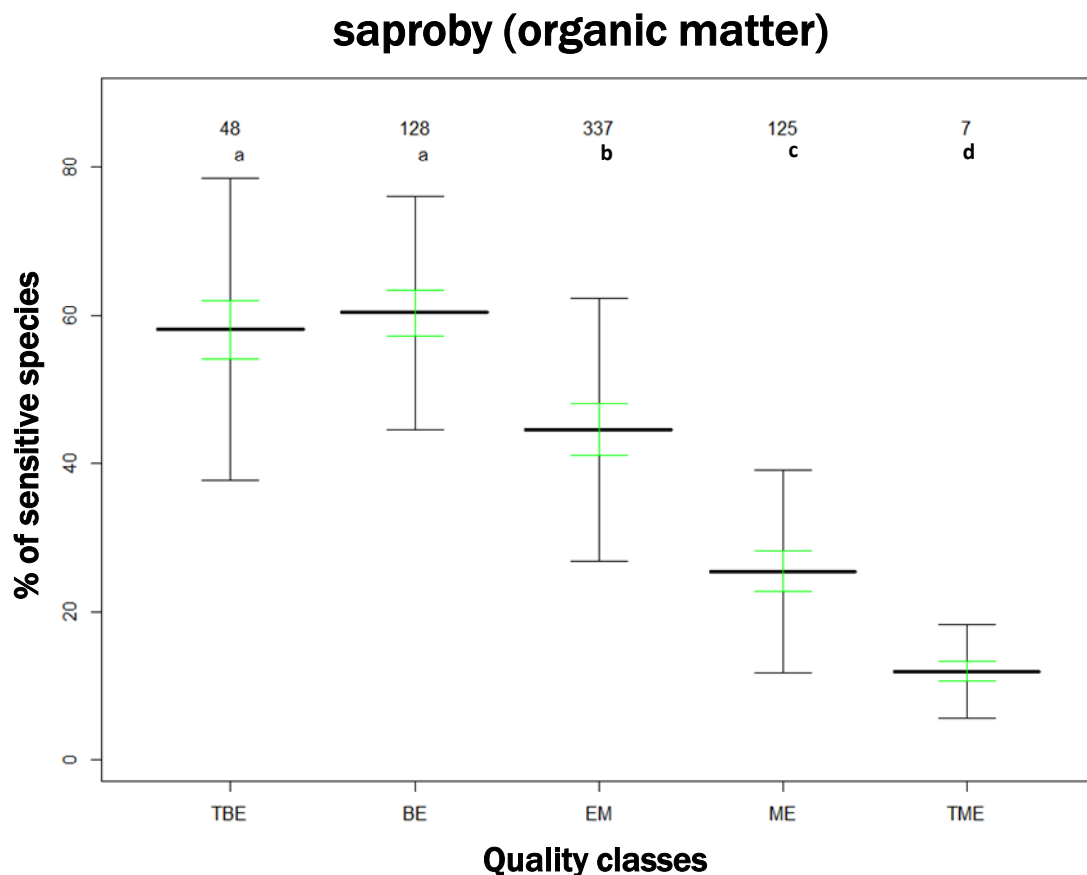
Even if an important percentage of underlying unknown species (see Figure 11) has also been cumulated here with the trophy-tolerant species, increasing in an exaggerated way the percentage of tolerant species in the high and the good status classes, the overall evolution of the community composition follows a more understandable shape. Taking into account that most of the 28 % (in mean) of taxa in the High Status Class and most of the 17% (in mean) of taxa in the Good Status Class have been misclassified in that analyse, the result of autecological traits per class would be rather comparable to the one obtained with the French dataset mixing all the types of rivers (Figure 9), passing from sensibly less than 50 % of tolerant taxa in the Good Class to sensibly more than 50 % in the moderate class, with significant differences in between.

The first thing we have to do now (the necessary content being not inside my computer right now...) will be to remove the "Class 7" species (unknown autecology with respect to the trophy) of the VLR countings before drawing again the Figure 12. That is not a completely satisfying way of doing (a non-negligeable part of the species enters in that case, and it would be preferable to define their ecological preferences than to remove them), but such an autecological analysis needs an important pre-processing of a lot of chemical data and will necessarily take time.

For the mid-term, in order to solve the unknown ecological characteristics of a substantial part of the communities that can be found in the french natural conditions, it will be important to process again a comparable analysis as the one done by van Dam twenty years ago, but at least on our French complete dataset.

**5) Links between IBD 2007 scores and the modification of autecological attributes of communities towards organic matter (saprobity) :**

We followed the same framework as for trophy, on the base of the 645 diatom samplings collected in Very Large Rivers exclusively (selection threshold: watershed > 8 000 km<sup>2</sup>).



**Figure 13 : Percentage of saprobity-sensitive species per quality class in very large rivers (> 8 000 km<sup>2</sup>)**

We firstly tried to figure out the distribution of saprobly-sensitive species, regrouping the Class 1: "Oligosaprobous" and the Class 2: "Bêta-mesosaprobous" of van Dam (1994).

Results of this first analysis (see Figure 13) globally show:

- no significant difference in the proportion of sensitive species between the "High" and the "Good" quality classes, this category of taxa being dominant in the community (mean level close to-to-more than 60 %).
- a significant decrease of sensitive species between the "Good" and the "Moderate" classes, which becomes dominated by tolerant species (mean level of sensitive species becoming inferior to 50 %).
- a significant and severe drop in the proportion of sensitive taxa for the "Poor" and the "Bad" classes.

Again, these results somewhere comfort the choice of the G/M threshold in the case of the French quality assessment system.

But we can also detect something illogical in the general shape of distribution of sensitive taxa, by seeing the inversed repartition of saprobic traits between the "High" and the "Good" classes.

This result lets us think about the same kind of artefact that previously for the trophic attributes of species.

Similarly to the case of trophic classification, a "Class 6" regroups the taxa which could not be attributed to a saprobic class because of insufficient data, or because they did not exist at all in the Netherlands countings. This drives to a lack of ecological information for a biogeographical reason.

We tried to figure out the repartition of such unknown taxa per quality class (Figure 14).

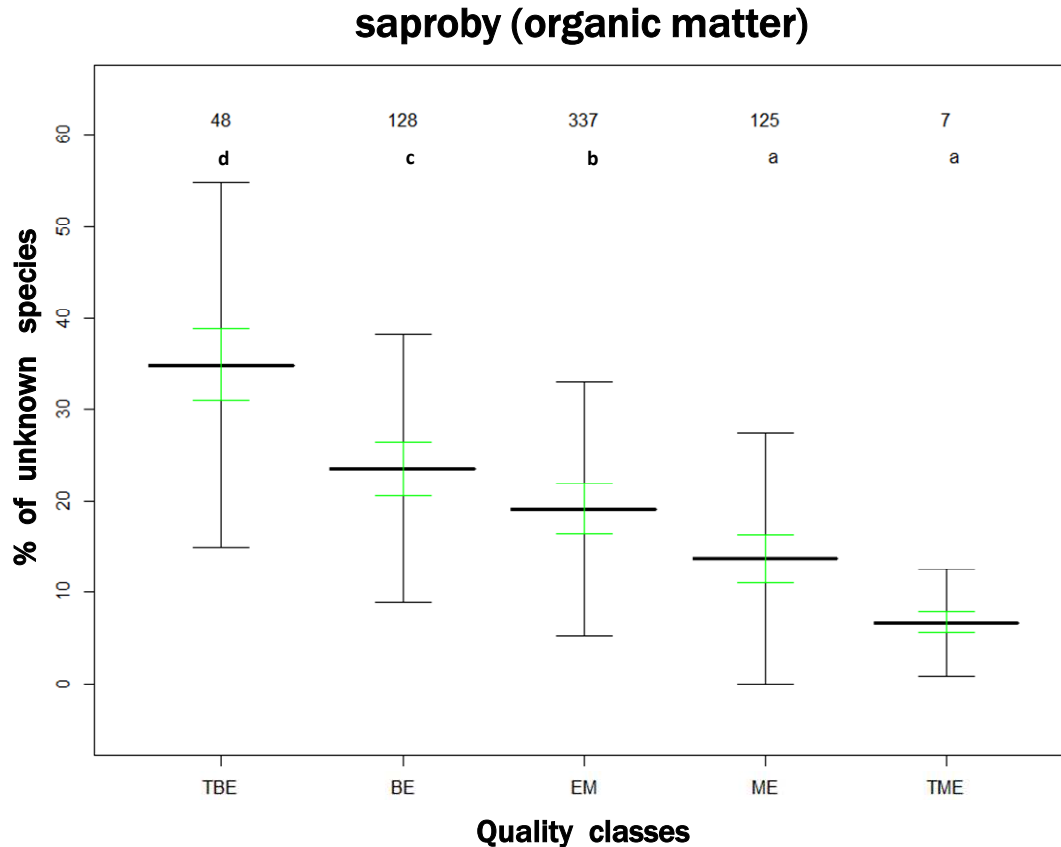


Figure 14: Distribution of unknown species (undetermined towards saprobly) along the 5 quality-class gradient

As for the trophy, an important part of taxa in the VLRs diatom communities have an unknown saprobic classification and furthermore, their distribution is not balanced between classes, but very significantly stronger in high (> 35 % in mean) and in good status (> 23% in mean), than in the poorer quality classes.

This gradient of undetermined species works at the inverse of the gradient of sensitive species. It quite strongly modulates the results presented in Figure 13, with the effect of artificially diminishing the level of "saprobly-sensitive taxa" in the H and G status classes.

The reason is similar as previously explained for trophy. Probably, in the Netherlands, van Dam never found most of the species contributing to the typical communities of High and of Good status-VLRs in France (the best quality rivers being Dordogne and Durance, which directly come from high elevation places in the "Massif Central" and Alps respectively). These sites host saprobly-sensitive species from mountainous areas that probably do not exist in the plain rivers of the Netherlands. At the inverse, he has been able to qualify the saprobic autecology of most of the tolerant taxa that we also find in France, illustrating the fact that this category of species is easier to be found into hydrosystems of the Netherlands.

Similarly as for soluble phosphorus, we then tried to express the distribution of saprobic autecology in an inverse way, by showing the increase of the tolerant taxa through the gradient of quality classes. This is not a completely satisfying way of doing, but is quicker to be done than a deep revision of autecological characteristics of taxa for a better fit to French biogeographical conditions.

The results are presented in Figure 15. In that case, we previously removed the "Class 6" taxa of the countings (unknown autecology with respect to saprobly), before processing the analysis.

### Saprobly (organic matter)

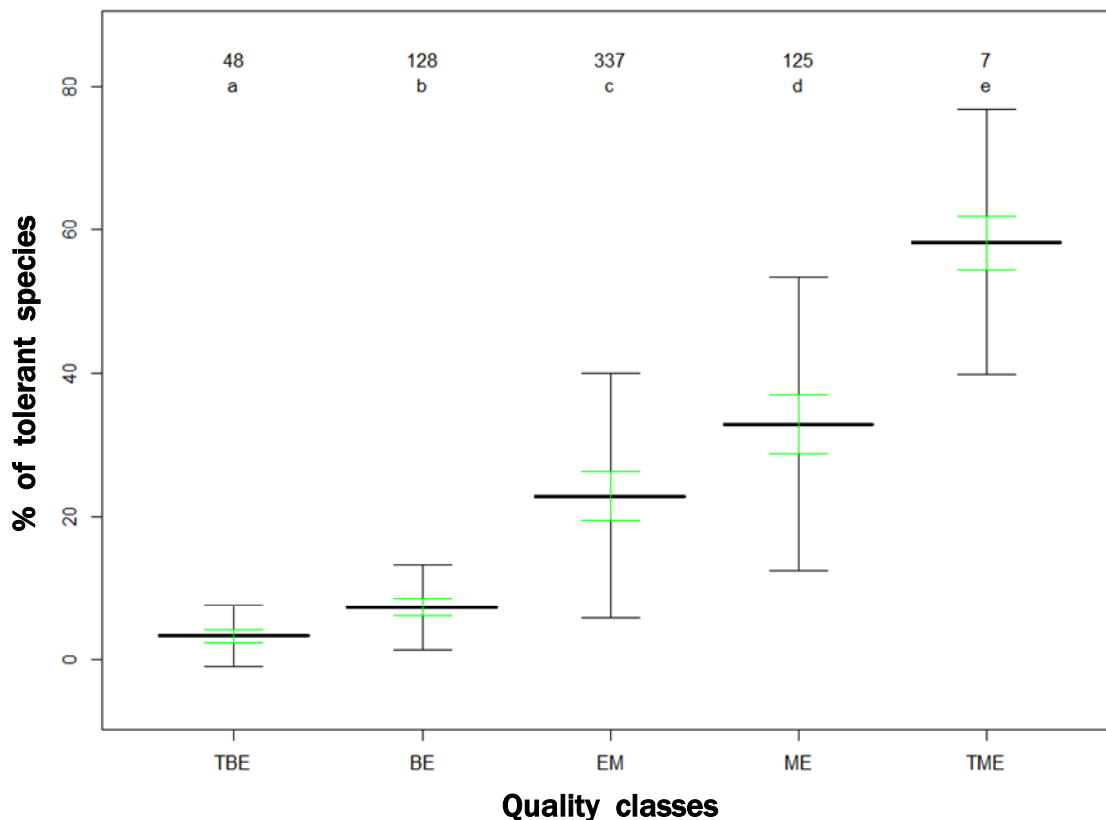


Figure 15: Percentage of saprobly-tolerant species per quality class in very large rivers (> 8 000 km<sup>2</sup>)

The results are quite similar to those concerning soluble phosphorus:



- the difference between high and good status is also slight, even if significant.
- the difference between good and moderate is also significant, but sensibly more important (from a mean of 6% to a mean close to 25 %), showing a sensible modification of the diatom communities with the increase of that kind of pressure.

Here again, the results shown in the Figure 5 reinforce the level of switching between the Good and the Moderate quality classes in the french ecological status assessment system, with respect to the normative definition of good status edicted by the WFD.

## **6) Stage conclusion :**

In comparison with analyses done on a complete national dataset, testing pressure-impact relationships on a selected VLRs data subset drives to the reduction of the internal anthropogenic pressure gradients, altering the illustration of pressure-impact relationships between the abiotic quality gradient and the national diatom index' response (in our case, IBD 2007).

These preliminary results:

- illustrate a quite satisfying adjustment of the G/M threshold in the French condition towards several pressure criteria,
- confirm the response of IBD 2007 with respect to a composite alteration gradient rather than to single pressures,
- allow to think that the investigation of pressure-impact relationships on an annual mean basis is not the most accurate way of doing, at least in the case of the diatom compartment,
- point out that autecological traits compiled by van Dam provide interesting results, but would need adaptation and extension towards a more complete list of french species to be fully usable in our national biogeographical context.

During the next few months, we intend to work further in order to deepen these approaches on the basis of the French "VLRs" data subset.

Other preliminary results that we have to investigate further, show that the distribution of diverse other attributes of species (N-trophy, motility...) towards the quality gradient give quite similar patterns.

We will then continue and improve the data analysis in order to better investigate the correspondence between chemical dataset, autecological attributes of diatom communities and French classification of ecological status based on diatoms.

Beside the intercalibration exercise, which is the main collective objective at that stage, it could be interesting to work those issues in a collaborative way, on a composite dataset of very large rivers originating from several participating E.U. Member States.

Done in Irstea Cestas, 21/12/2015

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