Rome, 14 Apr 2016

In the following I report the requested comments, as I was required as an appointed referee, in order to support the review process of the manuscript:

A Generalization of the TRIX trophic index to the Adriatic Sea basin.

Author(s): E. Fiori et al. - MS No.: nhess-2016-69 - MS Type: Research article

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A. With reference to the criteria recommended by the Editor, and expressing an overall judgment, this work is surely to be considered as an excellent work, both for its Scientific Significance and for the related Scientific Quality. It represents undoubtedly a serious contribution to the understanding "of natural and man-made hazards" in coastal-marine areas, to say the study of coastal-marine Eutrophication, by proposing new ideas and useful methodological approaches. In this respect, the interest for the work presented here, derives mainly from the mathematical approach adopted, which allowed to process and compare long historical data series available for the Adriatic, to the output of simulation models. The results are discussed in an appropriate and balanced way (i.e. clarity of concepts and discussion, including appropriate literature references).

As for the **Presentation Quality**, in this case also the judgement is to be considered "good": results and conclusions are presented in a clear, concise, and well-structured way (number and quality of figures/tables, appropriate use of technical and English language, simplicity and good understanding of the language used).

B. Now I enter into the merits of the work, not only to contribute to the discussion, but also to point out the need for some clarification in the text.

1. "The final results of this paper present a new TRIX index formulation that could be used to assess the GES of coastal and marine waters in terms of eutrophication status." (rows 80-82 of the manuscript).

The reference to the Marine Strategy Framework Directive is all too clear. However, it should be noted that both the EU Directives, the MSFD and also the WFD 2000/60 EU, in general give priority to the Biological Elements and they assume an approach based on DPSIR model (Driving forces, Pressures, Status, Impacts, Responses). Under this aspect the TRIX has been defined as a linear combination of the four fundamental trophic status indicators (chlorophyll, nitrogen, phosphorus and oxygen deviation from saturation). As such, TRIX represents a mixing of pressure (N and P) and impact (Chl-a and oxygen deviation) indicators and finally it does not result fully consistent with the MSFD "philosophy", nor with the WFD requirements.

Nevertheless, it can work as an excellent control variable and, in this light, it has already been considered by the Italian legislation that received the WFD directive, just as a variable supporting the judgment of ecological status. (Cfr. Decree 260/2010 "Classification criteria" at Cap. A.4.3.2: Technical criteria for classification on the basis of physico-chemical and hydromorphological quality elements in support. In particular Table 4.3.2./c provides class boundaries, expressed in terms of TRIX units, that cannot be exceeded if the resulting ecological status, based on the related Biological Quality Elements, belongs to the "good"class.)

2. "However, the general methodology for constructing TRIX for the different areas has not been clarified. In order to apply TRIX to different areas a precise evaluation of the scaling parameters is required." (rows 73-75 of the manuscript).

In the cited work of Giovanardi and Vollenweider (2004), on the meaning of the TRIX it is reported:

"...a purely numeric scale that scores the trophic properties of surface and subsurface seawaters, station by station and/or sequential in time, would be preferable to some preconceived categorical denominators."

Clearly this definition is limitative in the sense that it provides only one facet, while the broad array of tropho-dynamic processes and the related biological diversities that characterize any marine-coastal area, will have to be added by appropriate research.

On the other hand, **the approach is general, not geographically bound**, but on which the mentioned arrays can be built as a platform.

On this basis it was defined the trophic scale of the TRIX values, also reported in the present manuscript as Table 1 (pag. 23). The aim was to define a general reference system in order to make comparisons between different sea areas and / or between different seasonal periods on the same sea zone. It is clear that these comparisons will be possible,

only if the measuring tool used (i.e. the TRIX Index), was built with a unique criterion, namely, always using the same scales to define the range of variation of each component.

On the other hand (rows 216-227: Sensitivity analysis), the authors make abundantly clear why they have chosen different scales for TRIX components, depending on different areas and/or different temporal series. The pursued aim is certainly to be appreciated, being finalized not only to test the index used in different areas (affected and not affected directly by river inputs), but also to demonstrate the goodness of the general approach followed in the formulation of TRIX Index.

3. "Numerical simulations can therefore represent an important support for monitoring activities, i.e. they will allow to extend the use of TRIX to much larger areas where in situ sampling activities are difficult to implement. " (Rows 276-278 - Conclusion)

I think this is really the main scientific merit of all this work, that comes not only from the analysis of long time series of "real" data, but also by the possibility of generating simulated time series as output from numerical models. In this regard I wish to emphasize another merit of the work, it presents the results of the simulation models, without going too over the more technical and mathematical aspects and referring the interested reader to appropriate literature.

This has certainly helped to ease the reading of the work, allowing even those who are not expert in mathematical modeling to understand the results and the objectives pursued by the numerical simulations.

One last observation. The authors used the results of the models (i.e. the values of the components of the TRIX provided by simulation models), to describe the behavior of the index TRIX as such, in its temporal and spatial variability. But they omitted to consider the potential changes of the percentage contributions of the components of trix, in time and space.

This datum could be very interesting. The weight of each component in the index formulation is in fact to be related to the so-called "efficiency" of the marine and coastal systems to use nutrients to produce phytoplankton biomass. Of course, to concur in the formulation of the TRIX may prevail the components relating to the active phytoplankton biomass (Chl and Oxygen deviation from 100%), or nutritional factors, (N and P). In the latter case, the system does not use all the available nutrients and thus shows little "efficiency."

As discussed in the paper mentioned above (Giovanardi and Vollenweider, 2004), coastal systems of the Tyrrhenian and Adriatic Seas they differ primarily because of their peculiar "efficiency" in utilizing nutrients. But perhaps these aspects deserve to be treated and deepened, following the same approach already followed here, in a later work.

4. The TRIX index is based on four state variables (n), which are directly related to productivity: chlorophyll-a (Chl, $mg m^{-3}$), oxygen as the absolute percentage deviation from oxygen saturation (DO, %), dissolved inorganic nitrogen (DIN, $mg m^{-3}$) and total phosphorous (TP, $mg m^{-3}$). In particular, $DIN = NO_3 + NO_2 + NH_4$ and DO = abs(100-Ox), where Ox is the oxygen saturation. (Rows 96 -100).

Some imprecision may lead to confusion, as it has already happened on other occasions.

Is it correct to indicate the units of measurement expressed as weights (mg m⁻³) for Chlorophyll and Nutrients. But be aware that the relation $DIN = NO_3 + NO_2 + NH_4$, as it is reported in the text, is stoichiometrically incorrect. It would be correct if the units were all expressed in mM m⁻³. The correct form is therefore the following: $DIN = N-NO_3 + N-NO_2 + N-H_4$,

that is to say, at risk of becoming pedantic, DIN, Dissolved Inorganic Nitrogen in weight, corresponds to the sum of Nitric Nitrogen, Nitrous Nitrogen and Ammonia Nitrogen.

Recalling the very positive overall assessment of the work, as already expressed at the beginning, I believe that the authors should take into account these observations and adjust the text of the manuscript, where necessary.