A ring-test to improve the correct use of diatoms to evaluate a water body strongly affected by urban waste water and pesticides. Case study Ribosc River (Trento, Italy)

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After the first ring-test exercise on identification and counting protocols of benthic rheophilous diatoms held in Trento (Italy) in November 2012 (Torrisi *et al.*, 2013) a second intercalibration exercise was organised in March 2014. The aim of this ring-test was to deepen taxonomic problems in biomonitoring rivers using diatoms in order to decrease uncertainty of results in the application of the Intercalibration Common Metric Index or ICMi in Italy (D.M. 260/2010).

This second ring-test was attended by 51 experienced diatom analysts working in different Regional Environmental Protection Agencies. Each diatom analyst examined a slide prepared from a single sample of epilithic diatoms collected according to EN 13946 (CEN, 2003) in a station located in Ribosc stream (595 m a.s.l.), a watercourse belonging to "Calcareous southern Alps and Dolomites" hydroecoregion flowing in Trento Province (Northern Italy), affected by urban waste water and pesticides (e.g. pyrimethanil, carbaril).

The current ring-test was evaluated based on two different counting modules: 1) counting of the pre-selected target species: *Nitzschia dissipata* (Kützing) Grunow, *Navicula cryptotenella* Lange-Bertalot and *Fistulifera saprophila* (Lange-Bertalot et Bonik) Lange-Bertalot along a single transect with a defined length (12 microscopic fields) and 2) quantitative evaluation of the species composition by means of their relative abundance (up to 400 valves counted following the requirements of EN 14407 (CEN, 2004). Last but not least, influence of pesticides contamination on diversity and ecological guilds of river diatoms was analysed.

The results of evaluation of target species counts pointed out that the error rate for the first two species was 27% and 29%, respectively. The most probable sources of such bias are (i) the specific differences in the counting protocol of the different operators, e.g. counting of broken frustules, girdle views and teratogenic forms; and (ii) identification errors and taxonomic uncertainty. The species from the Nitzschia dissipata complex for example such as Nitzschia dissipata, N. media Hantzsch, N. recta Hantzsch and N. rectiformis Hustedt were often confused. also because their species identity is not clearly delimited and their determination characteristics overlap according to the common identification guides used as reference. Also Navicula cryptotenella was sometimes misidentified as Navicula antonii Lange-Bertalot. The error rate for Fistulifera saprophila scaled up to 49%. In this case, the variability of the counting, in addition to the sources of error mentioned above, was probably due to the low number of valves present in the fields considered for the exercise and to their small size and less silicified valves. Comparisons of quantification of the whole diatom assemblage showed that the most problematic taxa belonged to genus Achnanthidium. As confirmed by observations to SEM, in this sample was present only Achnanthidium minutissimum (Kützing) Czarnecki often confused with A. eutrophilum (Lange-Bertalot) Lange-Bertalot and A. saprophilum (Kobayasi et Mayamaea) Round et Bukhtiyarova. Furthermore, the sample contained significant proportion of teratogenic species and large number of motile species (22 species) such as Navicula and Nitzschia, which typically proliferate in nutrient rich waters contrary to those belonging to the high profile guild (13 species) that are exposed to dissolved pesticides to a greater extent being thus more sensitive. This could explain their lower abundance (Rimet & Bouchez, 2011).

References

- CEN, 2003. Water quality Guidance standard for the routine sampling and pretreatment of benthic diatoms from rivers. EN 13946. Comité Européen de Normalisation, Brussels, 14 pp.
- CEN, 2004. Water quality Guidance standard for the identification, enumeration and interpretation of benthic diatoms from rivers. EN 14407. Comité Européen de Normalisation, Brussels, 12 pp.
- D.M. 260/2010 Ministero dell'Ambiente e della Tutela del Territorio e del Mare. Decreto 8 novembre 2010, n. 260. Regolamento recante i criteri tecnici per la classificazione dello stato dei corpi idrici superficiali, perla modifica delle norme tecniche del decreto legislativo3 aprile 2006, n. 152, recante norme in materia ambientale, predisposto ai sensi dell'articolo 75, comma 3, del medesimo decreto legislativo. Supplemento ordinario alla Gazzetta Ufficiale n. 30 del 7 febbraio 2011 Serie generale.
- Rimet F. & Bouchez A., 2011 Use of diatom life-forms and ecological guilds to assess pesticide contamination in rivers: Lotic mesocosm approaches Ecological Indicators 11 (2):, 489-499
- Torrisi M., C. Monauni, R. Zorza, V. Della Bella, M. Siligardi, C. E. Wetzel, L. Ector, 2013 Ring-test exercise on identification and counting protocols of benthic rheophilous diatoms at Trento Province (Italy). 32ème Colloque de l'Association des Diatomistes de Langue Française & 7th Central European Diatom Meeting 16-20 sept. 2013.