Ecotoxicology and ecosystems health

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Summary. - There is mounting evidence that traditional investigations are no longer sufficient either to assess the ecological characteristics of a resource or to better understand the dynamics of an ecosystem. Reliable experimental data to be used for further investigation as well as for environmental corrective action are obtained using the traditional quantitative and qualitative biological methods. In fact, environmental health is connected with well functioning global and local ecological cycles, that also assure renewable, good quality natural resources. There is, therefore, a qeen need for new, adequate techniques based on biological parameters that will allow for a better understanding and protection of the environment. The use of biological indexes and indicators and toxicology tests have been also prescribed by a recent Italian law on water protection. Differences between toxicology, environmental toxicology and ecotoxicology, often used as equivalent terms, are also analyzed. Furthermore a new classification is proposed and mapping of ecobiotic and toxicologic data on studying waters in river basins.

Key words: biological indicators, ecotoxicology, water, ecosystems.

Riassunto (*Ecotossicologia e salute degli ecosistemi*). - Storicamente, la sorveglianza delle condizioni dell'ambiente è stata condotta tramite indagini di laboratorio che hanno fatto ricorso, in via pressoché esclusiva, a metodiche chimiche, fisiche ed alle analisi microbiologiche. Attraverso tali metodiche si riesce, infatti, a fronteggiare utilmente l'esigenza di definire la qualità delle *risorse* ambientali in relazione ai possibili *usi* cui esse sono destinate ed a predisporre, a tal fine, standard, obiettivi di qualità, strategie operative per la sicurezza e per la prevenzione sanitaria. Tuttavia, nel tempo è sorta l'evidenza di come l'applicazione di dette metodiche, da sole, non riesca a fornire un quadro sufficientemente accettabile dello stato di qualità ecologica complessiva di un ambiente. E' nata così la consapevolezza che il buono stato (anche utilitaristico) delle risorse è legato al funzionamento dei cicli ecologici locali e globali che ne consentono la rinnovabilità, il mantenimento qualitativo e - immediatamente o a seguito di semplici trattamenti - una pluralità di usi. L'inserimento nella normativa di settore degli indicatori biologici ed ecotossicologici rappresenta un importante progresso verso una gestione sostenibile delle acque. Viene trattata la distinzione terminologica fra tossicologia, tossicologia ambientale ed ecotossicologia ed è lanciata una proposta di classificazione e rappresentazione grafica dei dati eco-biotici e tossicologici nello studio delle acque nei bacini idrografici.

Parole chiave: indicatori biologici, ecotossicologia, acqua, ecosistemi.

Introduction

Traditionally environmental surveillance has been conducted by laboratory investigations, using chemical and physical methods as well as microbiological analysis.

Through these skilled investigations many goals can be reached: define the environmental quality (according to the destined use of the resource) assess standards, quality goal, operative strategies for safety and health prevention [1].

However, during the time it has become evident that these analytical methods by themselves were not able to give satisfying information on the state of the overall ecological quality of a given environmental compartment. The experience developed in the past few years, also showed that to focus investigations on characteristics for "prevalent and present" use of the resource it is non protective, during the time, for resource itself [2].

For instance, ammonia, nitrate and phosphate nutrients are quite desirable in water intended for agriculture, but this has contributed indeed to contamination of groundwater as well as eutrophication of lakes, rivers and marine coastal environments. Other possible use of the resource were spoiled too like fishing, bathing, drinking water, aquaculture.

We progressively became aware that a good status of the environment is strictly connected with well functioning global and local ecological cycles, that also

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assure a renewable, good quality of natural resource; then the need of new, adequate methods of investigations that will allow a better understanding and protection of the environment.

The use of traditional analytical methods is spoilt also by the massive production and diffusion of "brand new" chemicals; which makes more difficult their investigations; many pollutants are active at levels that cannot be traced by ordinary instruments or in case of episodic input.

Furthermore for years we studied pollution instead of quality, as a result there is a big lack of data regarding the "normal status" of a natural resource, a very helpful information to define a referring point to study the degree of variation from the natural quality.

More and more there is the need of new, multidisciplinary approach of investigation to improve our skills, that will consider the association of traditional analytic methods with new techniques, based on biological indexes and indicators, traditional toxicology tests, environmental toxicology and ecotoxicology. These new techniques are also considered in the more recent Italian and European law on water protection [3-5].

Both analytical and biological techniques have their own specific role, even if they give their best when used in association.

The traditional analytical, physical-chemical analysis gives information on:

- characterization of abiotic and mass components of ecosystem;

- presence and quantity of pollutants.

Whereas biological indicators are used on the field and are expressed by index, they are helpful to [6]:

- to characterize the "normal" status and the pathologic deviation;

- to disclose "hidden" or episodic pollution, so it is possible to use specific analytical analysis;

- to disclose all changes due to no direct polluting agents (damage of habitat, biodiversity);

- to give results testing not only the instant status but along a given period.

Biological approach

The use of biological techniques will then be helpful on answering the following question: *how much is it deviated from a good ecological status?*

In our opinion toxicology test is the connection-point of the two techniques physico-chemical analysis and biotic indexes.

In practice, sometimes the terms ecotoxicology, toxicology or environmental toxicology may be used as equivalent terms. Actually, they are specific fields with own scientific identity and peculiar testing procedures. They all provide investigation tools able to give us different information that, mutually, can be used for a better understanding of environmental conditions. Traditional toxicology is characterized by the use of a single species-test, it is always related to pure toxic single substance or a mixture of several ones considered in standard laboratory conditions.

The main advantage is to accomplish toxicity data in laboratory; the limit is that the toxic profile will not reflect all the dynamic processes in evolution in natural habitat, like synergy and/or antagonism occurrence, that can enhance or mitigate the toxicity tested in laboratory.

However this method has operative application too, especially in outlining priority for planning further investigations, prevention acts or even restoring actions [7, 8].

Environmental toxicology is helpful in identifying, even with predictive methods, the damage (caused by pollution or human pressures) occurred in living organism at the different trophic and habitat level in the considered natural resources.

It is specific on studying biological community or population of interest, instead of just only certain predetermined species (species-test).

In case of deviation from a normal standard assessed by biological indices, environmental toxicology is of use for a more incisive understanding of the lack of balance among biological communities at the ecosystem level.

Once the "agent" responsible for the damage has been individuated, it is possible to remodel the path along mass and energy flow to characterize the targets species.

Through these processes it is possible to assess the mechanism of action of pollutants, the compromised biological functions (growth, development, breathing, etc.) the degree of the damage on involved species (age, percentage of population) and, finally, a qualitative measure of ecosystem damage.

Compared to traditional toxicology, environmental toxicology can assess not only the effects of "poisoning" but also the exceeding biological tolerance (or the limiting factor) due, for example to nutrients or organic substances concentrations.

Ecotoxicology is often used in papers as an analogue of environmental toxicology and, to be sincere, is nowadays an abused, more then properly used, term, to reveal something different and new from what we have been discussed until this point.

We do believe also that this term can be specifically characterized in a future development of knowledge and diagnosis in environmental issue.

An ecotoxicological diagnosis is supposed to take in consideration all unbalances occurred on ecosystemic relationships due to polluting and/or pressure factors.

An ecosystem is characterized by an energy flow supporting a mass cycle. Its more striking characteristic is its own complexity, the content of negative enthropy in an interdependent relationships net.

The ecosystemic integration, allowing a dynamic order, is regulated by living organism sensitivity able to

react to different stimuli of "telemediators" of chemical (smells), physical (vision, color, shapes, electromagnetic, acoustic or electric) nature.

Considering such complex system, that we still cannot deeply understand and manage, we still consider environmental change phenomena in terms of "concentrations", dose-response, exposure and so on, that is with quantitative evaluations.

An effective ecotoxicology should take (hopefully will soon take) in greater consideration the qualitative aspects of ecological processes, giving attention to possible interference phenomena injuring telemediators and sensorial systems.

The latter can be injured even by very small traces of pollutant, producing an impairment of multiple physiologic abilities of living organism (hunting, hiding, to locate partners, moving in the environment). Pollutants interfering with this system of communication could be more dangerous of others that are not conflicting, producing only limited damages even at higher doses.

At the end, ecotoxicology should be the science that, hopefully in the immediate future, will investigate injuries to ecosystem caused not only by toxic substances and environment alteration but also by impairment of signals of telemediators. Only information, rather than mass, differently by traditional pollution phenomena, may be involved.

Planning environmental interventions

In everyday control activities we know that ordinary standards for chemicals are not enough to show hidden pollutants, whose identification calls for a number of specific and expensive analyses. In these cases, biological methods can help the chemical testing, as they reveal the existence of toxicity problems. We know this is particularly useful when assessing the contamination of drinking water, groundwater, liquid wastes, eluates from solid matrices: once the chemical composition of an environmental sample is known, it is possible to draw a check list of priorities for addressing recovery activities, focusing the attention on substances that show the greatest toxicity at the observed concentrations. This may lead to the planning of the environmental intervention. At this purpose, two elucidating cases may be reported. A tannery was planned to close down because its treatment system was not working efficiently and required a long, complex restoration. The liquid waste it produced rendered inactive the municipal water treatment plant close by, which could not be used in the case of an emergency. The factory was supposed to close down for good. At that point, tests were adopted to assess the toxicity of the waste studying the active sludge by monitoring cyliated protozoa. The results showed that hexavalent chromium and ammonia were largely responsible for the toxicity in the sludge. It was sufficient to do a partial cleaning, taking away only chromium and ammonia to make the waste compatible with the performance of the wastewater plant. This new system was tried for six months, enough to save the activity of the factory. The same approach was used to clean the liquid waste from an olive oil production plant, a very common problem in Mediterranean areas, which experts said was not possible to solve. In this case, the appropriate dilution dose for NOEC (no observed effect concentration) for protozoa was found and by reaching an acceptable minimum level just below NOEC, we made the treatment plant work.

Ecotoxicological tests used in Italy

First among European countries, the Italian legislation has recently included the ecotoxicological tests in the framework directive for the protection of surface and ground waters [3, 4], thus anticipating the European Waterframe Directive of year 2000. The introduction of test on aquatic organisms (invertebrates, fishes), and on bioluminescent bacteria represent the innovative approach which contributes, together with the classic chemical analyses to the definition of the limit values of pollutants.

These tests are briefly summarized below.

Water courses, lakes

Biota. - Priority tests: toxicity test on *Daphnia magna*, mutagenesis and teratogenesis tests, algal growth tests, bioluminescent bacteria tests; additional tests: BCF tests to detect priority, organic and inorganic, micropollutants (i.e. DDT, Cd) on fish and macrobenthic organisms

Sediments. - Priority tests on sediments on extracted sediments, total sediments, interstitial water (Onchorynchus mykiss, Ceriodaphnia dubia, Daphnia magna, Selenastrum capricornutum, Chironomus tentans, Chironomus riparius, bioluminescent bacteria) [9-14].

Coastal sea waters - transition waters

Biota. - Priority tests: BCF tests on heavy metals, PAH, PCB, organochlorine and organophosphorous pesticides; additional tests: short-, long- term tests on *Ostrea edulis, Crassostrea gigas, Mytilus galloprovincialis, Donax trunculus, Tapes decussatus, Tapes philippinarum* and on selected aquatic organisms, mainly from autochtonous fauna. The availability of standard protocols is a fundamental requirement

Sediments. - Priority tests: bioassays on different taxonomic groups integrated with chemical analyses on bioaccumulative compounds; additional tests: bioassays to reveal long- short- term effects to integrate analytical data.

Emission thresholds for wastewater

Toxicity tests over a 24 h (immobilisation) on *Daphnia* magna. Additional short term tests on freshwater invertebrates (*Ceriodaphnia dubia*, *Selenastrum capricornutum*) and saltwater invertebrates (*Artemia* salina), bioluminescent bacteria etc. [14, 15].

An operative integrated approach

As a proposal, an estimation of the environmental quality of water courses through an integrated project at the watershed level may be suggested, which includes a new classification of the toxicity potential of wastewater effluents in accordance with the EBI (extended biotic index) and IFF (river functionality index) indexes. It would be possible to design an evaluation of the toxicity of wastewaters based on the definition of five classes of toxicity through appropriate tests. The cartographic representation of the whole information may easily lead to a rapid comprehension of the environmental impact of the effluents in the different sections of a receiving water *course*: the point are depicted with bars and arrows, the discharged water volumes may be quantified through the shapes of *round areas* and coloured in order to reflect



Fig. 1. - Thematic map of the Tevere (Tiber) watershed partially classified through the EBI (extended biotic index) classification and wastewater effluent classification of toxicity.

different ranges of toxicity. Furthermore, other and integrated index (number of people in surrounding urban areas, secondary sewage, industrial, effluents) may be also specified.

As an example, the following figure reports the classification of a real portion of the Tiber river watershed (Latium region, Italy) where the environmental quality evaluation of the main water course through the EBI index has been recently conducted [16]. The map (Fig. 1) shows the biological quality together with the inclusion of hypothetical wastewater effluents: large volumes of lightly toxic effluents may not change aquatic environment quality.

But a very little volume per day of an high toxic pollutant, at insufficient dilution in the river flow may damage at significantly level the river quality.

Open questions

There are still several problems that need to be solved. Just to mention a few:

- the need to standardise analytical procedures to get more reliable results. A good solution would be the use of the Ring Tests;

- the improvement of the procedures in the case of synergic or antagonistic effects of pollutants;

- the definition of the proper use of single or specific tests. For instance, wine, which is not considered a contaminant of concern, is known to kill Daphnia, so it is advisable to use other indicators in the testing;

- the difficulty of testing the long-term toxicity of volatile compounds;

- the assessment by multiple testing procedures which are reliable for a large group of contaminants and for different environmental matrices, sea water included;

- the assessment by quick and simple tests (even if a little rough, very useful because rapid and providing a first diagnosis).

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