Water is not only a commercial product, but also a common good and a limited resource that must be protected and used sustainably, in terms of both quality and quantity. However, its use in a wide range of sectors, such as agriculture, industry, tourism, transport and energy, puts pressure on this resource [1–5]. For example, the European community has established two main legal frameworks for the protection and management of freshwater and marine water resources through a holistic ecosystem-based approach, namely the Water Framework Directive and the Marine Strategy Framework Directive [6,7]. These regulations establish a framework for the protection of inland surface waters, transitional waters, coastal waters and groundwater. This framework aims to prevent and reduce pollution, promote sustainable use, protect and enhance the aquatic environment and mitigate the effects of floods and droughts. The main objective is to ensure that all waters achieve a good ecological status. To this end, new minimum drinking water quality requirements have been established, emphasizing the importance of the presence of new emerging pollutants [8]. It is therefore important to implement methodological approaches to assess the ecological risk posed by chemical pollutants discharged into different aquatic environments [9]. Furthermore, the environmental risk management approach requires that the cumulative effect of all impacting anthropogenic activities be considered at different levels of an organization [10,11]. The need to detect the biological effects of emerging pollutants even at low concentrations and in complex mixtures has increased the need for studies on the relationships between exposure to contaminants and alterations to various biochemical and cellular processes in organisms, in order to use them as biomarkers of exposure and early responses to different classes of new emerging contaminants [12–16]. Biomarker measurements in bioindicator organisms have become valuable tools for environmental monitoring from the perspective of surveillance, hazard assessment or documentation of the remediation of aquatic environments [12,17]. Moreover, biomarkers are an essential component of aquatic environment monitoring programs in several countries, supporting commonly used chemical monitoring techniques [18–20].

Biomarkers include a variety of measures of the molecular, biochemical, cellular and physiological responses of specimens of key species to exposure to contaminants or physical stressors [21]. They act at the individual level, as for the classic ecotoxicological bioassays, but also provide mechanistic information on the effects of pollutants. For this purpose, certain criteria must be fulfilled: sensitivity, modulability, dose dependence and robustness to natural fluctuations [17]. Biomarkers must respond to pollutants within the expected environmental concentration range [17,19,21]. The validity of any biomarker thus depends on its ability to accurately separate the influence of natural variability from the toxic effects of different pollutant classes or from the generic stress caused by different anthropogenic factors [22]. To detect the effects caused by different stressors in bioindicators, one should not measure a single parameter, but a suite of biomarkers [21].
Biomarkers should not only be diagnostic but also prognostic of such high-level effects to provide a reliable early warning of the health status of a selected bioindicator [12].

Predictive toxicology has largely adopted the Adverse Outcome Pathways (AOP) model [19], a toxicological conceptual framework in which different orders of information are interlinked: from the chemical properties of pollutants to biochemical, cellular, and physiological effects, and high-level consequences such as reproduction and mortality. The future, therefore, lies in allocating suites of robust biomarkers in specific AOP frameworks. To this end, in recent years, new techniques have been developed for environmental applications [17]. Molecular techniques such as genomics, proteomics and metabolomics are being used to develop new biomarkers [23]. Furthermore, according to the theory of molecular systems biology [17], organisms react to all kinds of perturbations, including environmental changes and thus chemical pollution, through a cascade of high-throughput molecular processes, such as global gene expression, proteome, and metabolome remodeling [23]. The particular set of genes and/or proteins and/or metabolites involved will depend, at least in part, on the type of chemical. The particular response pattern may represent a fingerprint for a exposure to a specific pollutant, allowing a simple identification of the biomarker, but also providing information on all cellular processes taking place during the environmental perturbation [24,25]. The measurement of biomarkers is also important due to the use of sentinel species or key bioindicator species [12]. Indeed, the selection of a sentinel or bio-indicator species must be justified by a recognized link with the structure or functioning of the ecosystem being monitored. Various factors must therefore also be considered, such as the importance of the selected species within trophic chains, also in the light of the emergence of new problems such as the increase in invasive and highly competitive species that alter aquatic ecosystems. Measuring biomarker suites has been shown to be useful in assessing the impact of alien species in aquatic environments [24] and for detecting the health status of endangered species living in the impacted environment [19]. The use of an integrated approach of bio-indicators and biomarkers in ecological risk assessment nowadays makes it possible to investigate possible stress situations related to different time scales. The use of such investigative methodologies in an integrated ecotoxicological approach toward the biomonitoring of aquatic ecosystems offers a sensitive and specific tool for assessing exposure to emerging pollutants and the potential damage they exert on organisms living in each environment, allowing for short-term interventions and the development of appropriate sustainable environmental management programs [19,20,25]. The implementation of an integrated approach poses the need to improve the ability to discriminate contaminants (particularly the emerging ones) and to investigate the link between biomarker response and adverse effects at the organism level, including processes such as growth, reproduction, and mortality.

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References

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