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# Environmental Metabolomics in Aquatic Pollution and Toxicology

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## Introduction

Aquatic environments, particularly marine coastal areas and brackish inland ecosystems, have been increasingly threatened both directly and indirectly by a variety of anthropogenic activities, including primarily industries, agricultural operations and urban development. As a consequence of these activities, different types of toxic chemicals have been released into the environment provoking a serious impact on biota and human health [1]. Therefore, in last decades great concerns have been raised worldwide about the quality of the environments and the future of marine and coastal wildlife species as a result of aquatic pollution and several attempts have been made in order to develop effective tools for biomonitoring the environmental health status [2-4].

Aquatic ecotoxicology is a growing transdisciplinary research field that focuses on the study of the properties and behaviours of environmental contaminants in aquatic ecosystems, as well as on the assessment of their adverse effects on organisms, populations and communities. In ecotoxicological studies, both fish and aquatic invertebrates have been widely studied as sentinel organisms to predict the environment health status since they are able to efficiently metabolize and accumulate pollutants in their tissues and therefore to elicit measurable responses to toxic insults [5-15]. To date, it is well-documented that the presence in aquatic environments of pollutant mixtures, including heavy metals, polychlorinated biphenyls (PCBs), polycyclic aromatic hydrocarbons (PAHs) and pharmaceuticals, can induce toxic effects at various biological levels (e.g. molecular, cellular, biochemical, physiological), altering the normal biological performances of biota [5-9,11,13]. Taking into account that alterations at the organism level lead to changes at the population and community level, a number of biomarkers are frequently used in aquatic biomonitoring studies as early warning signals of environmental disturbances, including histological alterations [7,8,13], changes in gene and protein expression, variations in enzymatic activities or genotoxic biomarkers [5,7-10,12,13,16].

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However, due to their unidimensional nature, the conventional ecotoxicological methodologies usually applied for an estimation of the health status of sentinel organisms and their aquatic environments, could be not informative enough and cannot entirely provide the "full picture" of biological effects induced on organisms exposed to aquatic pollution. Therefore, with the purpose to overcome such limitations, an innovative and current approach is the employment of "omics" techniques, which consent the simultaneous and comprehensive evaluation of a broad number of biomolecules.

Among the "omics" techniques, metabolomics is now a wellestablished scientific field in systems biology, which refers to the identification of all endogenous low molecular weight metabolites (molecular weight <1000 Da) in cells, tissues, biofluids or whole organisms and their changes in response to physiological, developmental or pathological stimuli [17]. The value of metabolomics lies in the fact that it profiles simultaneously a wide range of metabolites involved in a variety of metabolic pathways and cellular processes, including ionic homeostasis, redox status, energy, protein and lipid metabolism and neurotransmission, providing thus a snapshot of the biological processes that are considered most proximal to a specific phenotype or disease [9,11]. In recent years, metabolomics has demonstrated to be a high-throughout approach with notable potential in the field of aquatic pollution and toxicology [9,11,17]. In particular, metabolomics based on protonic nuclear magnetic resonance spectroscopy (<sup>1</sup>H NMR), coupled with multivariate analyses and

chemometric approaches, has been revealed to be an effective and powerful tool for highlighting differences in the profile of metabolites (metabolic biomarkers) in response to environmental stressors or diseases, providing an overview of the metabolic status of a biological system [6,7,11,18-20]. Metabolite profiling, initially developed for biomedical applications, has been employed in a number of research areas such microbiology, plant science, food quality and most recently in environmental studies. Indeed, environmental metabolomics offers the potential to elucidate organism-environment interactions and it has many advantages for assessing organism health status at the molecular level. It is also well-documented that metabolomics allows the identification of new specific metabolite biomarkers of stress that are able to differentiate healthy from unhealthy organisms, as well as pollutant-exposed from unexposed aquatic organisms. Because metabolomics can provide valuable information on how xenobiotics influence physiological functions of organisms, this approach has been widely applied to experimental studies of

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selective exposure of toxic chemicals in various aquatic organisms, in order to provide insights into the mechanisms of toxicity of heavy metals, petrochemical pollutants, persistent organic pollutants (POPs), nanoparticles (NPs) and pharmaceuticals [6,7,9,11,15,18-20]. Additionally, environmental metabolomics has been successfully applied for a direct evaluation of pollutant mixture effects under field conditions, both on fish and aquatic invertebrates such as marine mussels [6,7,9,11,18,20].

Overall, the numerous biomonitoring studies that focused on the application of metabolomics as an innovative tool for environmental risk assessment demonstrate the effectiveness and high sensitiveness of the environmental metabolomics in elucidating disturbances in a variety of metabolic pathways in aquatic organisms, both fish and invertebrates, from sites with different level of environmental contamination and thus its suitability to be applied in studies of aquatic pollution and toxicology.

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