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Transcriptomic resources for environmental risk assessment: a case study in the Venice lagoon

M. Milan [a,*], M. Pauletto [a], L. Boffo [b], C. Carrer [c], F. Sorrentino [d], G. Ferrari [d], L. Pavan [e], T. Patarnello [a], L. Bargelloni [a]

[a] *Department of Comparative Biomedicine and Food Science, University of Padova, Viale dell'Università 16, 35020 Legnaro, PD, Italy*
[b] *Azienda ULSS n. 14 Chioggia, Italy*
[c] *Thetis S.P.A. – Magistrato alle Acque, Laboratorio Centro Studi Microinquinanti Organici (C.S.M.O.), Via Asconio Pediano, 9, 35127 Padova, Italy*
[d] *Ufficio Tecnico per l'Antinquinamento, Magistrato alle Acque di Venezia, San Polo 737, Riva del Vin, 30125 Venezia, Italy*
[e] *G3 Industriale, Via Milano 18, 30020 Marcon, VE, Italy*

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A B S T R A C T

The development of new resources to evaluate the environmental status is becoming increasingly important representing a key challenge for ocean and coastal management. Recently, the employment of transcriptomics in aquatic toxicology has led to increasing initiatives proposing to integrate ecotoxicogenomics in the evaluation of marine ecosystem health. However, several technical issues need to be addressed before introducing genomics as a reliable tool in regulatory ecotoxicology. The Venice lagoon constitutes an excellent case, in which the assessment of environmental risks derived from the nearby industrial activities represents a crucial task. In this context, the potential role of genomics to assist environmental monitoring was investigated through the definition of reliable gene expression markers associated to chemical contamination in Manila clams, and their subsequent employment for the classification of Venice lagoon areas. Overall, the present study addresses key issues to evaluate the future outlooks of genomics in the environmental monitoring and risk assessment.

1. Introduction

Ecosystems are complex entities subjected to sudden changes due to environmental variables, as well as to anthropogenic impact. Over the past century, the land surface has been deeply transformed by human action, leading to habitats impoverishment and worrying consequences on living organisms (Denslow et al., 2007; Vitousek et al., 1997). As a result, several countries have been compelled to implement legislations aiming to achieve sustainable management of water resources and to protect and improve the ecological status.

The European Water Framework Directive (WFD, 2000/60/EC) and the European Commission Directive (Directive, 2008/56/EC) established a legislative framework to define "good ecological status" and delineate the corresponding community actions in terms of water policy to protect and improve the ecological status of European surface waters (rivers, lakes, transitional waters, and coastal waters) by 2015. The WFD defines the ecological status as "an expression of the quality of the structure and functioning of aquatic ecosystems associate with surface waters" (Ar. 2.21) and requires site-specific evaluations of water bodies by integrating biological, physicochemical, chemical, and hydro-morphological data. Accordingly, in order to assess the ecological status of marine environments, water policies previously based on the establishment of emission limits, have been updated through the development and integration of approaches and knowledge from different disciplines able to detect changes in ecosystem functioning (Hagger et al., 2006; Solimini et al., 2006, 2009).

Recently, transcriptome profiling has benefited from technological improvements and has become more efficient and economically affordable for a wide range of species, thus overcoming the initial concerns about its high costs (Van Aggelen et al., 2010). The employment of transcriptomic techniques, such as DNA microarrays, in aquatic toxicology already allowed the identification of early biomarkers of exposure and the definition of Mode Of Action (MOA) or gene patterns associated to chemicals or environmental stressors (*e.g.* Falciani et al., 2008; Leaver et al., 2010;

* Corresponding author.
E-mail address: massimo.milan@unipd.it (M. Milan).

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Milan et al., 2013a; Pujolar et al., 2013; Schirmer et al., 2010). Therefore, international initiatives proposing to integrate eco-toxicogenomics in Environmental Risk Assessment (ERA) and in the evaluation of marine ecosystem health have been recently increased (Van Straalen and Feder, 2012; Van Aggelen et al., 2010).

Currently, the main challenges that must be addressed for a successful implementation of genomics in regulatory ecotoxicology are i) the standardization of data collection and analysis (Bourlat et al., 2013); ii) the definition of transcriptional features/perturbations reflecting the response to specific environmental conditions, and the establishment of thresholds associated to "non-significant" and "adverse" responses (Blunt et al., 2007); iii) the generation of well-defined MOA for reference chemicals; iv) the identification of links between molecular and biochemical responses and adverse alterations in survival, development, and reproduction (Van Aggelen et al., 2010); v) increasing exchange of information between scientists and end-users, such as government policy, non-government organizations and environmental regulators (Blunt et al., 2007).

The Venice lagoon constitutes an excellent case study for environmental risk assessment due to the severe impact of different anthropogenic pressures, such as the industrial activities of Porto Marghera, maritime traffic, and agricultural pollution (Apitz et al., 2007; Matozzo et al., 2010; Micheletti et al., 2011). Regulatory bodies in the Venetian region carried out extensive studies to assess the environmental status of the Venice lagoon. This area is also of major importance for bivalves production, especially Manila clams (FAO data; Turolla, 2007). Currently, to ensure that seafood from lagoon areas is absolutely safe for the consumer, one third of the Venice lagoon is classified as not suitable for shellfish harvesting (DGR 3366/2004). In addition, upper limits of whole-bodies dioxins and dibenzofurans (PCDD/Fs) imposed for bivalves commercialization (0.5 pg/g WHO-TEQ) are very stringent and far below those imposed by the European Commission (Regulation No 1259/2011) for fish and fishery products including molluscs species (3.5 pg/g WHO-TEQ).

Given the high economic importance of clam culture and harvesting in the Venice lagoon (Solidoro et al., 2003), the restrictions

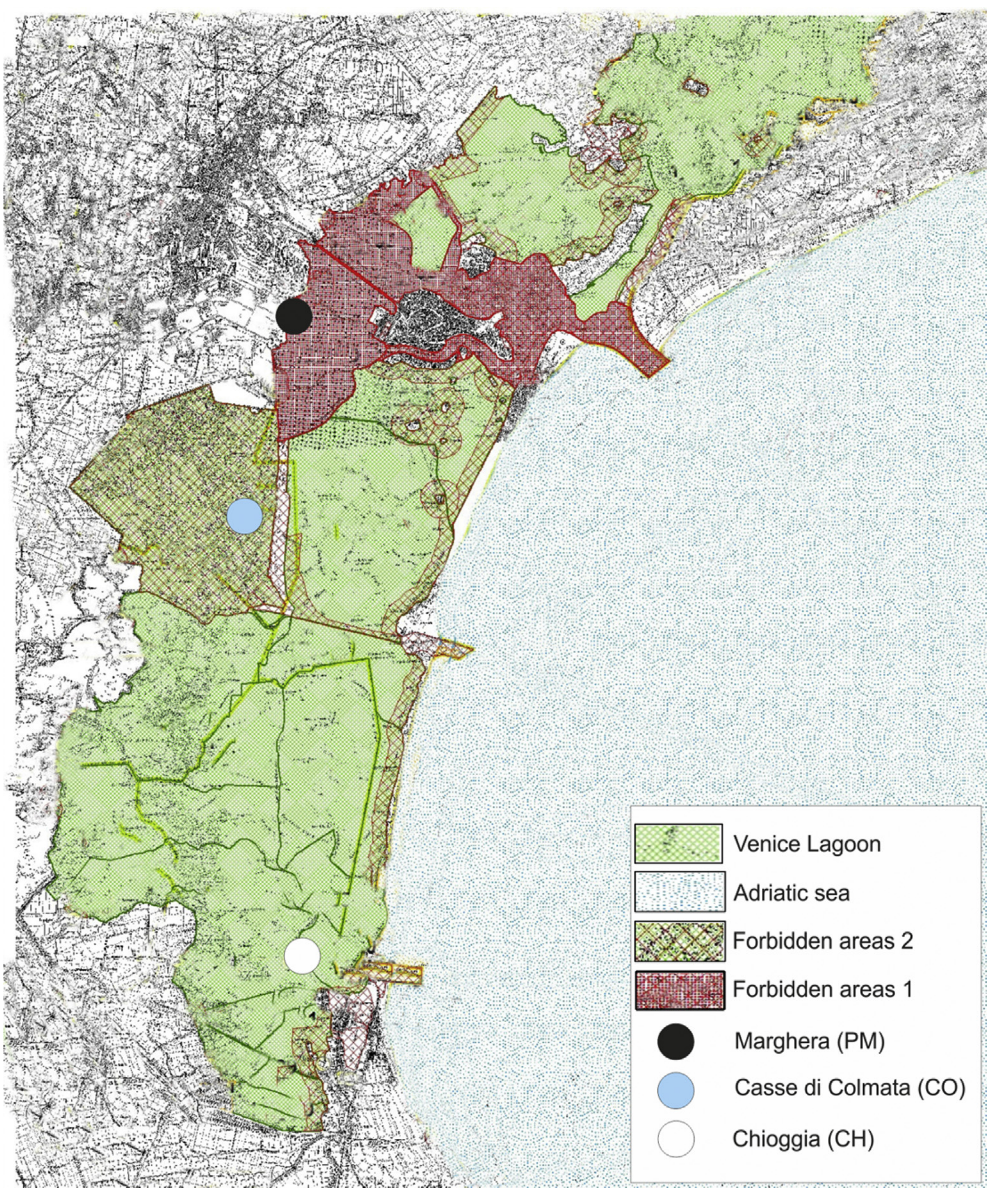

Fig. 1. Map of the Venice lagoon indicating the Manila clam sampling sites: Marghera (PM), Chioggia (CH), and Colmata (CO).