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Sediment toxicity assessment in the Lagoon of Venice (Italy) using *Paracentrotus lividus* (Echinodermata: Echinoidea) fertilization and embryo bioassays

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Abstract

The capacity of two toxicity bioassays (fertilization and embryo toxicity tests) to discriminate sediment toxicity using the sea urchin *Paracentrotus lividus* was tested in five stations with different levels of pollution in the Lagoon of Venice. Two stations were located in estuarine sites, two in the industrial zone, and one in a site at the top of our quality gradient (reference). Elutriate was chosen as sediment matrix to assess the potential effects of bioavailable pollutants in the water column as a consequence of sediment resuspension (dredging and dumping, fishing gear, etc.). An experimental design based on Quality Assurance/Quality Control procedures (QA/QC) was adopted in order to set the methodological basis for an effective use of these bioassays in monitoring programs. Results revealed both higher embriotoxicity than spermiotoxicity in all stations and the efficacy of combined use of both toxicity bioassays in discriminating differing pollution/bioavailability between stations and periods. The good representativeness of the integrated sampling scheme and the standardization of all experimental phases yielded high precision of results. Clear Toxicity Fingerprints were evidenced for the investigated sites through the combined use of both bioassays. A good fit between ecotoxicological data and chemical contamination levels was found, except for unnatural sediment texture.

Keywords: Sediment toxicity; Lagoon of Venice; Sea urchin; Elutriate; Methodological research

1. Introduction

In lagoonal environments, sediments represent the major repository of integration and accumulation of many toxic substances. These substances may affect benthic and water column organisms, the latter mainly because of frequent artificial sediment resuspension (e.g. dredging operations, fishing gear used in clam harvesting, boating) such as that occurring in the Lagoon of Venice (Rosselli et al., 2002; Facca et al., 2002).

The use of biological indicators has become very important in evaluating the quality of sediment and its potential effects on the lagoonal ecosystem, due to resuspension phenomena. The choice of biological indicators must be based on reliable scientific knowledge and, possibly, on the availability of standardized procedures. The development of methods and their validation for the environment of interest requires iterative steps such as set-up, evaluation of reproducibility, sensitivity and discriminatory capacity with pure substances. As far as sediment is concerned, it is also necessary to evaluate the reliability of the methods used for the individual matrices (pore water, elutriates, whole sediment) in order to define their field of applicability (Volpi Ghirardini et al., 2003).

Toxicity bioassays have been applied worldwide to contribute to the assessment and monitoring of sediment quality, since only the responses of living systems are able to integrate the complex effects of contaminants. The internationally recognised peculiar roles of toxicity bioassays (Nendza, 2002) are related to their ability to provide quantifiable information about the potential for biological

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damage (toxic hazard) caused by bioavailable multi-factorial contamination, to give responses which are not restricted by a predetermined list of contaminants, to have a spatial–temporal resolution that is usually considered better than that of other bioassessment tools (MacDonald et al., 1997; Wells, 1999). In particular, in lagoonal ecosystems, toxicity bioassays might contribute to the proper management and recovery of areas subjected to pollution, since regulatory authorities must have reliable methods for characterizing sludges and selecting disposal procedures for contaminated materials.

Among the recognised methods for biomonitoring coastal marine and estuarine environments (ICES, 1997; US EPA, 2000), sea urchin toxicity bioassays are well-known to be sensitive and reliable. Standard procedures using critical stages of the sea urchin life-cycle (i.e. fertilization, embryo development) are enforced in some countries (Environment Canada, 1992; US EPA, 1994a, 1995; ASTM, 1995). Sea urchin toxicity bioassays are applied worldwide to assess and monitor sediment toxicity by the exposure of gametes and embryos to aqueous phases such as pore water (Carr and Chapman, 1995; US EPA, 2000) and elutriates (Beiras, 2002; Nendza, 2002).

Sea urchin toxicity bioassays using the north Atlantic and Mediterranean autochthonous species are being studied in some Italian research laboratories (Arizzi Novelli et al., 2001). Two methods have been developed in our laboratories for the most common sea urchin species *Paracentrotus lividus* (Lamarck, 1816) with reference to standard procedures, the sperm cell toxicity or fertilization test (Volpi Ghirardini and Arizzi Novelli, 2001) and the embryo toxicity test (Arizzi Novelli et al., 2002). Both procedures were tested for intralaboratory reproducibility, comparability with Atlantic sea urchin data using a reference toxicant (Volpi Ghirardini and Arizzi Novelli, 2001) and reliability using organic (Volpi Ghirardini et al., 2001), organometallic (Arizzi Novelli et al., 2002) and inorganic toxicants (Arizzi Novelli et al., 2003). The present work is part of a study using an integrated approach (toxicity bioassays, field bioaccumulation indicators and chemical analyses) to evaluate the quality of sediments in a coastal lagoon such as the Lagoon of Venice (Italy). This research was focused on a methodological study conducted in order to provide data on the effectiveness of the above-mentioned sperm cell and embryo toxicity tests using *P. lividus* to discriminate sediment bioavailable contamination. Five stations, representing different types and levels of pollution, were selected and elutriates were chosen according to their potential effects on sensitive biological components (i.e. gametes, embryos and larvae) that could be exposed to pollutants made bioavailable in the water column as a consequence of sediment resuspension. The aims were: a) to evaluate the replicability of all procedures (sediment sampling method, sediment treatments and preparation of elutriates, toxicity tests results); b) to rank sites (make a classification among sites) for toxicity and to take into account possible temporal variations (summer–winter sampling campaigns) at each site; c) to evaluate a possible fit between ecotoxicological and chemical assessment approaches. This work seeks to form the methodological basis for providing reliable toxicological data to contribute to sediment quality assessment and monitoring in transitional environments.

2. Materials and methods

2.1. Sampling stations

The Lagoon of Venice can be considered to be an ideal environment for testing a method's performances (i.e. discriminatory ability, resolution power, correspondence with contamination levels, etc.) and their utility for quality assessment and monitoring. Indeed, it is a very complex transitional environment characterized by high macro- and micro-variability and by multifaceted dynamics, severely influenced by man (Ravera, 2000). It includes estuarine and marine environments, pristine salt marshes and human environments like the city of Venice and the industrial district. Point and non-point sources contributing to the pollution flowing into the lagoon include: industrial discharges, untreated and treated municipal wastewaters (from Venice and Mestre urban areas, respectively), streams, agriculture, motorboat traffic and atmospheric deposition. Marked gradients of sediment contamination, from the industrial district to the sea inlets, were shown by a recent monitoring program (Venice Water Authority, 1999).

Five sampling sites located along a pollution gradient were chosen (Fig. 1). The main criteria used in choosing the sampling areas were differences in contamination levels and availability of information on chemical–physical characteristics and particularly on pollutants loads (DRAIN Project). Particular attention was given to the creation of a link with other projects (particularly the DRAIN Project) and co-ordination with research groups of the same Orizzonte 2023 Project. After an initial survey on selected areas, the definitive sampling sites were chosen on the basis of population abundance of Nereid polychaetes (*Hediste diversicolor*), used for simultaneously studying organic micropollutant's bioavailability in sediments (Volpi Ghirardini et al., unpublished data).

Two stations were chosen in estuarine areas used to study pollutant loads in the Lagoon: the estuaries of the Dese (DE) and Marzenego (Osellino Canal—OS) rivers. Both stations are influenced by freshwater carrying agricultural runoff; OS is also affected by multifactorial contamination from urban source (treated and untreated wastewaters from the city of Mestre), industrial source due to the vicinity of Porto Marghera industrial district. A possible leaching of contaminants from an uncontrolled landfill built over the Campalto saltmarshes, just behind the Osellino Canal, is supported by the impressive confining works aimed to prevent the release of leachates at this area, started after the conclusion of this study (CVN, 1999). To the best of our knowledge, no information on surface sediment contamination of both sampling sites was available before this research.

Two other stations were chosen in canals located inside the industrial district of Porto Marghera: the South Industrial Canal (SA) and the Lusore–Brentelle Canal (BR). The latter was expected to be extremely polluted (Spoladori, 2004), it also receives freshwaters from the mainland and was used to study