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# Several benthic species can be used interchangeably in integrated sediment quality assessment

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## ABSTRACT

The selection of the best management option for contaminated sediments requires the biological assessment of sediment quality using bioindicator organisms. There have been comparisons of the performance of different test species when exposed to naturally occurring sediments. However, more research is needed to determine their suitability to be used interchangeably. The sensitivity of two amphipod species (Ampelisca brevicornis and Corophium volutator) to sediments collected from four different commercial ports in Spain was tested. For comparison the lugworm, Arenicola marina, which is typically used for bioaccumulation testing, was also tested. Chemical analyses of the sediments were also conducted. All species responded consistently to the chemical exposure tests, although the amphipods, as expected, were more sensitive than the lugworm. It was found that C. volutator showed higher vulnerability than A.brevicornis. It was concluded that the three species can be used interchangeably in the battery of tests for integrated sediment quality assessment.

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# 1. Introduction

# Dredging is routinely carried out in harbors and navigational channels. However, oxygenation and mixing during the operations may form new chemical bonds causing an influx of contaminants ([Casado-Martinez et al., 2007a\)](#page--1-0). This necessitates a pre-dredging study of sediment contamination status to assess the possible its toxic impact.

A common tool for testing sediment quality is toxicity bioassays conducted in natural and controlled conditions. Toxicological results together with chemical and physical characterization of the sediments form the basis of a weight-of evidence approach, acknowledged as a powerful tool for assessing sediment quality (e.g., [Casado-Martinez et al., 2007b,](#page--1-0) [2008;](#page--1-0) [Chapman et al., 2002;](#page--1-0)

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# [Chapman and Anderson, 2005;](#page--1-0) [Crane, 2003](#page--1-0); [DelValls and Conradi,](#page--1-0) [2000\)](#page--1-0).

The amphipods Corophium volutator and Ampelisca brevicornis have been used for sediment quality characterization in various Spanish ports ([Casado-Martınez et al., 2006;](#page--1-0) [Chapman et al.,](#page--1-0) [2002;](#page--1-0) [DelValls et al., 1998;](#page--1-0) [Riba et al., 2003\)](#page--1-0). Generally, amphipod species have been widely used in sediment quality analysis because of their abundance, high sensitivity to environmental contamination and high tolerance to many other environmental factors [\(Long et al., 2001\)](#page--1-0). According to [Casado-Martinez et al.,](#page--1-0) [\(2007a\)](#page--1-0), C.volutator and A.brevicornis provide similar toxicity responses in highly contaminated sediments but different responses in less contaminated ones. More evidence is required to determine that these two species can be used interchangeably for sediment quality assessments. The lugworm Arenicola marina is also used for sediment quality assessment because of its bioaccumulation capability [\(Casado-Martinez et al., 2007a,](#page--1-0) [2008;](#page--1-0) [Morales-Caselles et al., 2008](#page--1-0); [Ramos-Gomez et al., 2011](#page--1-0)).

The aim of this paper is to compare the sensitivity of C.volutator, A.brevicornis and A.marina to toxic exposure in acute toxicity bioassays to determine the extent they can be used interchangeably in the battery of tests for integrated sediment quality analysis,

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including dredged environments. The test battery includes a number of bioassays designed to characterize sediment toxicity and is an intrinsic part of the weight-of-evidence approach.

## 2. Materials and methods

### 2.1. Sediment sampling

Sediment samples were from four commercial ports along the Spanish coast: Huelva (H1, H2, H3), Santander (S1, S2, S3), Barcelona (B1, B2, B3) and Cadiz (CA1, CA2, CA3, CA4) (Fig. 1). The site CA2 is located in a fishing dock. CA3 and CA4 have a high accumulation of contaminants; B1-B3 and S2 are subject to intense shipyard and traffic activities. Huelva is affected by historic mining activities.

At each site, sediments (three samples per site) were collected by 0.025  $m<sup>2</sup>$ Van Veen grab from approximately the top 20 cm and transported in a cool container to the lab, where they were homogenized with a Teflon $<sup>®</sup>$  spoon and</sup> sieved through a 2 mm mesh to eliminate debris. The sediments were then subsampled for physical characterization and chemical quantification, following Spanish recommendations for dredged materials ([CEDEX, 2008\)](#page--1-0). The sediment samples were stored at  $4^{\circ}$ C in the dark and hermetically closed for at most two weeks.

Field related work complied with quality assurance recommendations per [ASTM \(1991a,b\)](#page--1-0); laboratory tests were conducted with appropriate quality control (blanks, controls, ambient conditions, etc.) as outlined by [Morales-Caselles et al.](#page--1-0) [\(2008\)](#page--1-0) and [Riba et al. \(2003\)](#page--1-0).

#### 2.2. Sediment analysis

The total concentrations of select metals (Hg, Cd, Pb, Cu, Zn, As, Ni and Cr), 7 polychlorinated biphenyls (PCB: 28, 52, 101, 118, 138, 153, 180), polycyclic aromatic hydrocarbons (PAH: anthracene, benz(a)anthracene, benzo(ghi)perylene, benzo(a)pyrene, chrysene, fluoranthene, indene (1,2,3-cd) pyrene, pyrene and phenanthrene), organoclorine pesticides (POC: HCH, aldrin, DDT, dieldrin, endosulfan, ensosulfansulphate, endrin, endrin-aldehyde, heptachlor, heptachlor epoxide, hexachloro-1,3 butadiene, hexachlorobenzene, lindeno) and tributyltin (TBT) were measured to determine the chemical content of the sediments. Concentrations of Cd, Pb, Cu, Zn, Ni and Cr were determined by microwave acid digestion in Teflon vessels and quantified using atomic absorption spectrometry. Cold vapor technique and hydride generation were used for Hg and As, respectively. Both metals were determined by atomic absorption spectrometry. Results were expressed as  $mg \, kg^{-1}$  dry weight. PCBs and POCs and 9 PAHs were extracted with cyclohexane and dichloromethane. PCBs and POCs were quantified by gas chromatography with electron capture detection (by US EPA method 8080) and 9 PAHs recommended by OSPAR were detected by HPLC with fluorescence detection (US EPA method 8310). TBTs were extracted with hexane and tropolone 0.05% and analyzed by selective ion monitoring gas chromatography-mass spectrometry (SIM GC/MS) Organic compound concentrations were expressed as  $\mu$ g kg $^{-1}$ dry weight.

The accuracy of all analytical procedures was verified using the reference materials MESS-1 NRC and CRM 277 BCR for metals, and NRC-CNRC HS-1 for PCBs and PAHs, with a percentage of recovery higher than 90%. Detection limits ranged between 0.001 and 0.008 mg-kg—1 and 10–20 µg-kg—1 dry weight of sediment<br>for metals and PAHs, respectively, and were 0.5 µg kg<sup>–1</sup> dry weight of sediment for PCBs and 2  $\mu$ g kg $^{-1}$  dry weight of sediment for TBTs.

Organic carbon content (TOC) in sediments was determined according to [\(MAPA, 1998\)](#page--1-0) and expressed as percentage. For granulometry analysis, recommendations by [Thain and Bifield \(2001\)](#page--1-0) were used: coarse particle size exceeded 2 mm, sand size ranged from 0.063-2 mm and fine particles had size  $<$  0.063 mm. Sediments with the percentage of sand  $>$  than 80% were considered sandy; those

where percentage of fine particles (henceforth fines) exceeded 80%—muddy. For the rest of the cases, when sand was a prevailing substratum ( $>60\%$ ) it was classified as more sandy; if mud prevailed  $(>60%)$ —more muddy; if there was approximately equal percentage of sand and mud, then the sediment was sandy– muddy.

#### 2.3. Toxicity tests

Three independent 10 day static acute sediment toxicity tests were conducted using two crustacean amphipods (A.brevicornis and C.volutator) and a polychaete (A.marina). The percentage of mortality after exposure was selected as the toxicity endpoint.

#### 2.3.1. Amphipod test

Bioassays using the crustacean amphipods were conducted according to the procedures described in [\(Casado-Martinez et al., 2006;](#page--1-0) [Morales-Caselles et al.,](#page--1-0) [2008](#page--1-0); [Riba et al., 2003](#page--1-0)). Reference species of A.brevicornis and C.volutator were collected from clean areas of the coasts of Cadiz ([Casado-Martinez et al., 2007b](#page--1-0)) and Galicia [\(Morales-Caselles et al., 2007](#page--1-0)), respectively, by sieving the sediment through a 0.5 mm mesh.

The amphipods were transported to the laboratory in containers with seawater. In the laboratory, they were placed in 11 L aquariums with clean seawater and sieved sediment from the same locations and were acclimated for 7 days. During acclimation, aeration was provided, natural photoperiod was selected and no food was supplied.

Prior to the toxicity tests, approximately 200 g of sieved sediment from the different study sites were placed in 5 replicates in 2 l glass beakers and with about 800 ml of overlying clean seawater. When the sediments settled down, aeration was provided. After 12 h, 20 amphipods were introduced into each test beaker. Natural photoperiod was selected and no food was provided during the experiment.

Temperature (20  $\pm$  1 °C), pH (7.9–8.2), salinity (34–35‰) and dissolved oxygen ( $\geq$  90%) in the seawater were controlled daily. Concentrations of total ammonia in interstitial water were measured by indophenol blue adsorption method using a Technicon TRAACS 800 autoanalyzer and monitored at the beginning (day 2) and at the end (day 8) of the bioassays. They were below 16 mg/l in all samples.

#### 2.3.2. Polychaete test

A.marina were collected from a clean area situated on the Cantabrian coast of north Spain [\(Casado-Martinez et al., 2008\)](#page--1-0) by hand-digging and immediately transported to the laboratory in cool boxes containing clean seawater (the time between the collection and the arrival was about 24 h). In the laboratory, the lugworms were placed in aquariums with 5 cm of sieved sediment from the survey area and acclimated for 7 days.

Prior to the test, 5 cm layers of sieved sediments were placed in 201 aquariums (three replicates) and a clean seawater was added. When the sediment settled aeration was provided. 12 hours later, 10 A.marina were introduced into each replicate. During the acclimation and experimental periods, the natural photoperiod was selected, no food was provided and water was renewed on day 5.

Bioassay was performed following to the protocol used by [Thain and Bifield](#page--1-0) [\(2001\).](#page--1-0) The temperature (17  $\pm$  1 °C), pH (7.8–8.4), salinity (34–35%) and dissolved oxygen ( $\geq$  85%) in seawater were controlled daily.

#### 2.4. Sediment categorization

The degree of sediment contamination was expressed as categories derived from the Spanish dredged material management framework [\(CEDEX, 2008](#page--1-0)) [\(Fig. 2](#page--1-0)). The framework uses a weight-of-evidence methodology by linking



Fig. 1. Map showing locations of the selected ports.

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