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# Nematode biomass and allometric attributes as indicators of environmental quality in a Mediterranean harbour (Ligurian Sea, Italy)

Valentina Losi\*, Mariapaola Moreno, Luigi Gaozza, Luigi Vezzulli, Mauro Fabiano, Giancarlo Albertelli

University of Genoa, Dipartimento di Scienze della Terra, dell'Ambiente e della Vita (DISTAV), Corso Europa 26, 16132 Genoa, Italy

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# ABSTRACT

Nematode biomass and allometric attributes (size spectra, body length and width, morphotypes) were measured and related to the environmental quality of sediments of the Genoa-Voltri commercial harbour (Ligurian Sea, NW Mediterranean) to investigate their possible use as ecological indicators. The sediment quality was defined by measuring the level of organic enrichment (quantity and biochemical composition of sedimentary organic matter) and oxygen stress (redox potential).

Nematode biomass spectra (NBS) proved to be extremely valuable in determining differences in the environmental quality of sediments on a hundred-metre spatial scale. High peaks of the NBS were observed in the more organic-rich and oxygen-stressed stations probably in relation to a lower diversity of the nematode communities in these stations, with the predominance of tolerant genera such as *Paracomesoma* and *Sabatieria*. Among allometric variables, the length was found to be negatively correlated with oxygen concentrations and positively with TOM percentages, whilst the morphotype length/width ratio (*L*/*W*) resulted negatively related to oxygen concentrations and organic matter quality (protein:carbohydrate ratio), suggesting that these allometric attributes represent indicators of the functional adaptation of nematodes to the changing environmental conditions.

We suggest that in contrast to time-consuming and expertise-requiring nematode taxonomic analysis, biomass and allometric attributes analysis can provide a simpler but comparable tool to assess sediment quality and environmental heterogeneity of harbour ecosystems.

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## 1. Introduction

Harbours are generally enclosed areas characterised by low hydrodynamism and oxygen availability in the water column, high rate of sedimentation, high anthropogenic pressure and concentrations of pollutants (e.g., Estacio et al., 1997; Guerra-García and García-Gómez, 2004; Jiang and Falconer, 1985). The problems associated with harbours ecosystems are becoming increasingly important (Jiang and Falconer, 1985), causing significant damage to water and sediments quality and, subsequently, to marine life and ecosystems, and to human health (Mestres et al., 2010). Particularly, harbours are receptor of multiple contaminants owned to their related activities and the result is a wide combination of several contaminants, such as toxic compounds, heavy metals, hydrocarbons, organic matter, mineral or organic particles either in their organic or inorganic form. Moreover, the environmental disturbance within harbours may change rapidly over spatial scales of a few metres, depending on various factors like the localisation

E-mail addresses: valentina.losi@unige.it, losivalentina@gmail.com (V. Losi).

and magnitude of pollution source, river inputs, tidal regime, water circulation, harbour position, shape and size (Fabiano et al., 2006).

The relevant importance of sustainable development has led to research into methods for environmental management in harbours (Grifoll et al., 2010) and to include the environmental topics in port development and management policies. Therefore the identification and assessment of environmental risk areas within harbours may be helpful for the development of good planning and monitoring programmes, leading to better management of harbour ecosystems (Moreno et al., 2008b).

The biological indicators play a fundamental role in the monitoring of the environmental conditions, as the Water Framework Directive (WFD, Directive 2000/60/EC) and the Marine Strategy Framework Directive (MSFD, Directive 2008/56/EC) have pointed out, recommending their use in the water body quality status assessment. The use of physico-chemical or abiotic variables to detect changes or impact in environmental conditions often is problematic and many researchers have recognised the need to measure environmental pollution and impacts using biological rather than physico-chemical indicators (e.g., Fabiano et al., 2004a,b; Vezzulli and Fabiano, 2006; Goodsell et al., 2009).

The meiofaunal community has proven to be extremely useful in assessing the effects of anthropogenic disturbance in marine

<sup>\*</sup> Corresponding author. Tel.: +39 010 353 8584.

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sediments (e.g., Coull et al., 1981; Warwick et al., 1988; Coull and Chandler, 1992; Austen and Somerfield, 1997; Fichet et al., 1999; Lee et al., 2001), and due to its peculiar characteristics (small size, high abundances, presence of typical tolerant genera), it represents the most significant metazoan component of the fauna in harbour sediments (Fichet et al., 1999). Within meiofaunal taxa, nematodes offer a variety of possibilities for assessing changes in community structure, due to their high structural and functional diversity, constituting the most diverse and numerically dominant metazoans in aquatic habitats, with a wide distribution varying from pristine to extremely polluted habitats. For these reasons, nematodes have already been employed in biomonitoring studies and turned out to be suitable indicators for pollution-induced disturbances of benthic ecosystems (Ferris and Ferris, 1979; Coull and Palmer, 1984; Coull and Chandler, 1992; Bongers and Ferris, 1999; Höss et al., 2011; Moreno et al., 2011; Losi et al., 2012).

Previous studies on nematode communities in different harbours and marinas allowed defining the general environmental quality state and identifying a spatial differentiation, where present, among areas at different "vulnerable" level, highlighting the relationship between physical-chemical variables and benthic assemblages and representing an important tool in the monitoring programmes to evaluate the environmental health (Marin et al., 2008; Moreno et al., 2008a,b, 2009; Nanajkar and Ingole, 2010; Liu et al., 2011). These studies are based on nematode analysis at genus/species level, requiring a quite considerable amount of work, money and high taxonomic skills. For these reasons nematodes are not regularly implemented in monitoring studies. However, since nematodes are known responding to the environmental disturbance both in terms of genera/species and morphotypes, in the present study the response of some parameters based on nematode biomass and allometric attributes to the environmental conditions was investigated, in order to test the accuracy level of results based on these parameters compared with those obtained with taxonomic identification. Body size in fact is related to many aspects of animals, such as their life history, physiology, energy requirement and biotic and abiotic interactions (Peters, 1983; Calder, 1984). Although, at first glance, nematodes may appear structurally very similar, looking more carefully, they show a relatively wide range of morphometrical adaptations and remarkable differences in size and body proportions (Tita et al., 1999; Vanhove et al., 2004). Therefore, assuming that similar shapes correspond, to some extent, to similar fitness constraints, morphometric characterisation becomes a useful descriptor of ecosystems (Schwinghamer, 1983).

Even though direct adaptation to sediment particle size is often reported as the main cause of the variation in body size of the infauna (Schwinghamer, 1981; Snelgrove and Butman, 1994; Steyaert et al., 1999), many studies have rejected this hypothesis (e.g., Duplisea and Drgas, 1999; Leaper et al., 2001). In fact many sediment characteristics are probably confounded with sediment grain size (Snelgrove and Butman, 1994) and factors such as organic content, water content, redox potential, porewater oxygen concentrations, seem to be strongly related with infauna body size (Fleeger et al., 2011).

One of the most extensive and powerful generalisation that can be used in ecological studies is represented by the study of the biomass distribution over size (Peters, 1983), useful in describing and comparing benthic communities (Sprules and Munawar, 1986; Drgas et al., 1998; Duplisea and Drgas, 1999; González-Oreja and Saiz-Salinas, 1999; Duplisea, 2000). Indeed, while in undisturbed sediments the benthic size-spectra resulted to be conservative (Drgas et al., 1998; Duplisea and Drgas, 1999; Duplisea, 2000), in stressed ecosystems variations in macro- and meiobenthic biomass spectra are reported (González-Oreja and Saiz-Salinas, 1999; Vanaverbeke et al., 2003). Nematode body size has been found to be sensitive to environmental changes: Soltwedel et al. (1996) detected changes in nematode body size due to changing food availability, Vanaverbeke et al. (2003) reported alterations in nematode biomass size spectra (NBS) as responses to oxygen stress and phytoplankton sedimentation events. Another informative parameter is represented by the nematode shape, which was suggested to be related with the available food but also with the biogeochemical conditions of the sediment (e.g., oxygen stress) (Tita et al., 1999; Soetaert et al., 2002; Vanaverbeke et al., 2004).

In order to calculate the biomass and morphotypes, only nematode length and width, robust parameters measured using non-destructive methods (Soetaert et al., 2002), are required. Thus, in comparison with the taxonomic identification, biomass and allometric attributes can provide an easy way for monitoring changes in the sediments due to anthropogenic or natural stress, earning time, energy and money (Vanaverbeke et al., 2003), with relevant implications in environmental management studies, which often requires a quick and economic response.

The aim of the present study was to evaluate the possible use of nematode biomass and allometric parameters to assess the environmental quality of harbour ecosystems. In order to reach this aim:

- (1) biomass, allometric parameters, NBS, width, length and length/width spectra relative to three harbour stations characterised by different level of environmental quality (in terms of organic enrichment and oxygen stress), were compared;
- (2) the information obtained by these analysis was contrasted with the structural analysis results;
- (3) the relationships between nematode morphometrics and environmental proprieties were investigated.

The null hypothesis tested are that nematode size and shape (i) do not show differences between stations with different environmental quality; (ii) give information not comparable with the one based on taxonomic identification; (iii) are unrelated to the examined environmental proprieties.

#### 2. Materials and methods

#### 2.1. Study site and sampling design

Data were collected in the industrial Genoa-Voltri harbour, an important container and oil terminal located to the west of Genoa (Ligurian Sea, NW Mediterranean). Genoa-Voltri is a heavily polluted harbour, characterised by high concentrations of organic compounds (e.g., proteins, carbohydrates, PAHs), covering an area of 34.5 ha with a depth of 7-12 m and sediment composed of fine sand and silt. Samples were collected on four occasions (June, July, November 2002 and February 2003) from three sampling stations, located over a distance of 1000 m. These stations were situated at: the inner part of the harbour (station I), the middle (station M) and the outer parts (station O), close to the open sea (Fig. 1). At each station, nine PVC-core (inner diameter, 3.6 cm) sediment samples were taken by scuba divers, three for chemical analysis (quantity and biochemical composition of sedimentary organic matter, redox potential and polycyclic aromatic hydrocarbons (PAHs)) three for meiofauna and nematodes and three for total bacterial counts.

A detailed description of study site, sampling, experimental design and sample processing techniques and methods can be found in Salvo et al. (2005) and Moreno et al. (2008a).

Only data related to the surface sediments samples (0-2 cm) were examined in this study.

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