



# Response of free-living marine nematodes to the southern Yellow Sea Cold Water Mass



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## ABSTRACT

The Yellow Sea Cold Water Mass is a remarkable seasonal hydrographic event in the bottom water of the Yellow Sea. In order to reveal the response of free-living marine nematodes to this event, community structure and biodiversity indices of nematodes were studied in June and November 2013. The dominant species were *Dorylaimopsis rabalaisi*, *Spilophorella* sp., *Daptonema* sp., *Sabatieria* sp. and *Parasphaerolaimus* sp. In terms of trophic structure, epigrowth feeders were the most dominant group. Correlation analysis showed that Shannon–Wiener diversity index had significantly negative correlation with sediment silt–clay percentage, organic matter content and water content. Results of BIOENV indicated that sediment phaeophorbide content, water content, bottom water salinity and temperature were the most important factors related to nematode community. In conclusion, community structure and biodiversity indices of nematodes were consistent in the two sampling seasons.

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## 1. Introduction

Free-living marine nematodes are the most diverse and numerically dominant metazoans in marine ecosystems (Heip et al., 1985; Balsamo et al., 2010, 2012; Appeltans et al., 2012). Due to their unique reproductive strategies (small size, high abundance and fast turnover rates), many studies demonstrated that marine nematodes have a global effect on the energy conversions within the pelagic–benthic system, via the ingestion and stimulation of production of microorganisms (Zhang and Zhou, 2003). Besides, the ecological and practical advantages associated with the use of nematodes in benthic studies make them suitable bioindicators in environmental monitoring using changes of community structure and biodiversity indices (Schratzberger et al., 2000; Balsamo et al., 2012; Semprucci and Balsamo, 2012; Semprucci et al., 2014). By virtue of their dominance, universality and robust bodies, nematodes are the most promising component of meiofauna for assessing effects of natural and anthropogenic disturbances on marine environment (Sandulli and de Nicola-Giudici, 1990; Sandulli and Nicola-Giudici, 1991; Bongers and Ferris, 1999). Biodiversity indices is suggested as the ultimate measure of ecosystem health (Leonard et al., 2006). Conventional measures of species diversity such as Shannon–Wiener diversity index ( $H'$ ), Pielou's evenness index ( $J'$ ) and Margalef's richness index ( $d$ ) are the most widely used ecological parameters for assessing biodiversity indices.

For the mixed impact of unique topography and other thermal and dynamic factors, the Yellow Sea has formed a unique hydrological phenomenon: under the seasonal thermocline covering a basin scale of low-temperature water, namely the Yellow Sea Cold Water Mass (Ho et al., 1959; Yu et al., 2006). The Yellow Sea Cold Water Mass is a seasonal water mass. In spring, with the emergence of the thermocline, the Yellow Sea Cold Water Mass begins to form. Until late spring, with the development of the thermocline, the Cold Water Mass is fully formed, lasting from July to August for the prosperity of the thermocline. During autumn, with weakening of thermocline, the Cold Water Mass is declining. By December, the thermocline and Cold Water Mass almost disappear simultaneously. The Yellow Sea Cold Water Mass affects phytoplankton growth and reproduction to a certain degree. Moreover, macrobenthos density is relatively low in this area due to the impact of the Cold Water Mass (Ho et al., 1959; Zhang et al., 1996; Yu et al., 2006; Wang, 2001; Zhang, 2012).

Several studies on free-living nematode assemblages have been reported in the southern Yellow Sea (e.g. Liu et al., 2005, 2007; Huang et al., 2006). However, there are limited studies on meiofauna and nematode community in the southern Yellow Sea Cold Water Mass area (Wang et al., 2011; Xu et al., 2015; Liu et al., 2015). Although basic information of free-living marine nematode community structure is essential to understand baseline benthic conditions of the southern Yellow Sea Cold Water Mass, response of marine nematodes to the formation and evolution of the southern Yellow Sea Cold Water Mass is more important. The purposes of the present study are to analyze the response of marine nematodes to the formation and evolution of the southern

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Yellow Sea Cold Water Mass through analysis of community structure and biodiversity of free-living marine nematodes.

## 2. Materials and methods

### 2.1. Field sampling

Sediment samples were collected in the southern Yellow Sea Cold Water Mass (YSCWM, according to Yu et al., 2006; Ho et al., 1959) and its adjacent area (G3, F4) during two cruises in June and November 2013 by R/V “Dongfanghong II”. A grid of 8 sampling stations was chosen, located at 33°–37°N and 121°–124.5°E (Fig. 1).

Undisturbed sediment samples were collected with a modified 0.1 m<sup>2</sup> Gray-O’Hara box corer (Jonasson and Olausson, 1966). Three cores of sediment for meiofauna (a cut-off syringe with a 2.9 cm inner diameter and 8 cm deep into the sediment) were carefully taken from three boxes respectively at each sampling station. All samples were fixed on board with 5% buffer formalin solution in seawater. Sediment samples were also collected and frozen at –20 °C for the analysis of organic matter, grain size, Chlorophyll-a (Chl-a) and Phaeophorbide (Pha). Sediment grain size was measured by a laser particle size analyzer (Master Sizer 3000). Chl-a and Pha contents were determined with spectrophotofluorimetry following the protocol given by Lorenzen and Jeffrey (1980) and modified by Liu et al. (1998) for wet sediment. The organic matter content of the sediment was measured by the (K<sub>2</sub>Cr<sub>2</sub>O<sub>7</sub>–H<sub>2</sub>SO<sub>4</sub>) oxidation method (National Technology Supervision Bureau of China, 1992; Mudroch et al., 1997). Conductivity/temperature/depth (CTD) profiles were obtained for water depth, bottom water salinity and temperature by a Seabird CTD in situ.

### 2.2. Analysis of meiofauna and marine nematodes

Meiofauna samples were stained with 3–5 ml Rose Bengal for more than 24 h. Then all the samples were washed on a sieve with the pore size of 0.5 mm for the upper size limit and a sieve with the pore size of 0.031 mm for the lower size limit (SCOR Working Group 76, 1994). Meiofauna were later extracted from the remaining sediment using centrifugation in Ludox-TM with a specific gravity adjusted to 1.15 (Heip et al., 1985; Warwick et al., 1998). Each sample was washed into a lined Petri dish and nematodes, copepods and other taxa were sorted and counted under a stereoscopic microscope (Higgins and Thiel, 1988). After being counted, all meiofauna samples were fixed with 5% formalin solution.

Nematodes were transferred into the 9:1 solution of 50% alcohol:glycerol in block cavity to slowly evaporate to pure glycerol,

then mounted into permanent slides (Huang and Zhang, 2004; Zhang, 2005). Nematodes were identified to species level following the pictorial keys by Platt and Warwick (1983, 1988), and Warwick et al. (1998) and the NeMys online identification key using a compound microscope (Olympus BH-2) with bright-field illumination (Heip et al., 1985; Somerfield and Warwick, 1996).

### 2.3. Nematodes feeding type

Based on the characteristics of buccal morphology, Wieser (1953) devised a classification of feeding types for nematodes. According to this classification, 4 groups of feeders were defined as follows: no buccal cavity or a fine tubular one-selective deposit (bacterial) feeders (1A); large but unarmed buccal cavity-non-selective deposit feeders (1B); buccal-cavity with scraping tooth or teeth-epistrate or epigrowth (diatom) feeders (2A); buccal cavity with large jaws-predators/omnivores (2B).

### 2.4. Statistical analysis and ecological indexes

Diversity indices of marine nematode community were calculated using Shannon–Wiener diversity index ( $H'$ ), Pielou’s evenness index ( $J'$ ), Margalef’s richness index ( $d$ ) and Simpson index ( $\lambda$ ) as follows:

$$H' = -\sum (N_i/N) \log_2(N_i/N)$$

$$J' = H' / \log_2 S$$

$$d = (S-1) / \log_2 N$$

$$\lambda = \sum (N_i/N)^2$$

where  $S$  is the number of species at each station,  $N$  is the total number of individuals, and  $N_i$  is the number of individuals of the  $i$ th genus ( $i$  from 1 to  $S$ ).

The differences of nematode abundance between two sampling seasons and among all sampling stations were tested with Independent-Samples T-test and one-way analysis of variance (ANOVA) using SPSS 19.0. To further examine nematode community structure, non-metric Multi-Dimensional Scaling (MDS) was performed. BIOENV analysis was used to link the multivariate biotic patterns of nematodes to suites of environmental variables. Analysis of similarity percentages was used to find the dominant species. K-dominance curves were plotted for the comparison of species diversity of the sampling stations. The above analyses were performed using the PRIMER v6.0 (Plymouth Routines in Multivariate Ecological Research), a multivariate statistical package

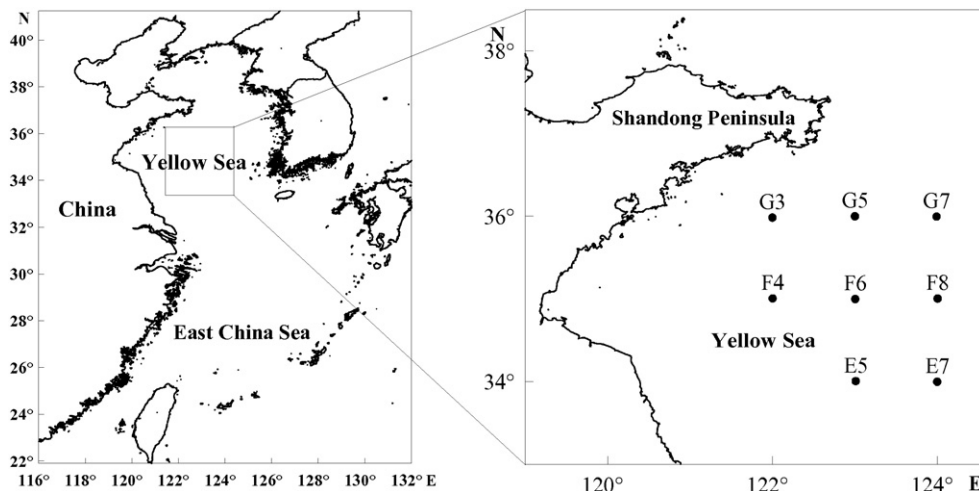


Fig. 1. Sampling stations in the southern Yellow Sea Cold Water Mass.

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