MEIOBENTHOS AND TAXOCENE OF NEMATODES UNDER CHANGES IN ECOLOGICAL CONDITIONS IN THE COASTAL ZONE OF THE NORTHWESTERN PART OF THE BLACK SEA

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 Abstract: Based on studies conducted in June - August 2023 at two marine sites in the in front of Odessa Bay (Cape Lanzheron and Cape Maly Fontan), an assessment was made on the response of meiobenthos and nematodes to the destruction of the dam of the Kakhovka reservoir (06 June 2023) in the coastal waters of the NW Black Sea. In the sublittoral zone (0.5 m) of Cape Lanzheron, the abundance of meiofauna varied from 4000 to 169830 ind./m², averaging 28094 ± 10982.2 ind./m². A low occurrence of meiobenthos taxa, a species diversity index, and a high percentage of dominance of Nematoda and Oligochaeta were noted. An increase in the number of r-strategists and a predominance of non-selective detritivores were recorded. According to the values of the maturity index (MI) and the index of trophic diversity (ITD), the water area of Cape Langeron during the study period changes the status of ecological quality (EcoQ) from Good (I-III survey) to Poor (IY-Y survey). The increase in eutrophicity in the waters of Cape Maly Fontan in the sublittoral zone (1 - 5 m), caused by the consequences of the disaster, contributed to an abnormally sharp increase in the abundance of meiobenthos, especially nematodes. The density of meiobenthos varied from 114000 to 9708310 ind./m², averaging 1753412 ± 261350.7 ind./m². Low MI values and an increase in ITD values due to the noticeable dominance of one trophic group (1B) indicate increased stress in the area of Cape Maly Fontan. The response of nematode assemblages to environmental changes, both in terms of density and diversity, has demonstrated significant potential for persistence and recovery. The increase in the density of Harpacticoida and juvenile Bivalvia during the last survey (08/24/2023) may be evidence of their ability to quickly recover when environmental conditions improve.

In general, the status of the ecological quality of the environment (EcoQ) of the study area can be classified as (Bad) in the area of Cape Lanzheron and (Poor) in the area of Cape M. Fontan.

Key words: meiobenthos, nematodes, ecological quality status, coastal area of NW Black Sea

1. INTRODUCTION

The consequences of anthropogenic impacts on marine ecosystems are most clearly not only determined by chemical and physical parameters, but also by biological ones. Currently, ecologists use dozens of different methods for assessing the quality of the environment based on biological indicators. Among the benthic community of organisms, it is the meiobenthos, a unique complex of small bottom animals 0.1 – 3 mm in size that is an informative and practically

method for assessing the state of marine ecosystems under conditions of environmental pollution (McIntyre, 1969; Giere, 2009). Meiobenthos, having high fertility, as a rule, a short life cycle and a rapid growth rate, is capable of rapid restructuring of its structure and functional indicators. The response to environmental stress is manifested both in the indicators of the entire meiobenthos as a whole, and at the level of its large taxonomic ranks and directly at the species level (Vorobyova, 1999).

Meiofaunal features are good indicators of environmental conditions and changes caused by anthropogenic activities (Vincx and Heip, 1987; Balsamo *et al.*, 2012; Alves *et al.*, 2013). Biological indicators are considered more relevant for assessing environmental quality than physicochemical or abiotic variables alone (Semprucci *et al.*, 2012). Meiobenthos is also able to be the first of the zoobenthos to restore its populations (Heip *et al.*, 1985; Coull *et al.*, 1992; Danovaro *et al.*, 2008; Moreno *et al.*, 2008; 2011).

Nematodes are one of the dominant groups of meiobenthos, which plays an important role in marine ecosystems. They can tolerate even the worst polluted biotopes; being often among the last surviving organisms in such places. Free-living nematodes are the most abundant and diverse marine metazoan taxon in various sediments (Heip *et al.*, 1985; Lambshead *et al.*, 2003).

The possibility of using meiobenthos in assessing the quality of the marine environment has been indicated by Black Sea researchers (Gomoiu, 1985; 1987; 2005; Vorobyova, 2000; Ürkmez *et al.*, 2014; Muresan *et al.*, 2019; Muresan *et al.*, 2019; Kulakova, 2022).

The Odessa Marine Region (OMR) is one of the most ecologically vulnerable water areas of the NW Black Sea. Being bordered in the north by the mouth of the Small Adzhalyk Estuary, and in the south by the Sukhoi Estuary, it is of particular interest. Its hydrological conditions are formed under the influence of urbanized runoff of the Odessa metropolis and waters coming from the Dnieper-Bug estuary (Garkavaya *et al.*, 2006).

The shallowest part of the OMR is the Odessa Bay, which is the most susceptible to anthropogenic pressure. The salinity of the waters of the OMR water area is formed mainly under the influence of the Dnieper (93.4%), Southern Bug (5.7%), Ingul (0.5%) and Ingulets (0.3%) discharges. The water of the estuaries' spreads in a thin layer over the surface of sea water and forms three water masses in the OMR: estuary with a salinity of 3 – 7 ‰, surface sea water 12 – 17 ‰ and bottom sea water 7 – 18.5 ‰ (Vinogradov, Khutornoy, 2013).

Apart the rivers as sources of substances dangerous for marine ecosystems, the coastal zones are also point sources of pollution. In the first place comes the influence of economic activities of ports, shipping, dredging, etc., recreational load and discharges of domestic wastewater (Vinogradov *et al.*, 2012; 2014). Also, large-scale protection work was carried out on the Odessa coast from the late 50s to early 60s. Concrete blocks of piers, concrete traverses and breakwaters were installed, covering 18.7 km of the coastline. They form basins, most of which have poor water exchange with the open sea. The beaches of the narrow coastal zone were reclaimed. Such large-scale water engineering construction introduced a number of changes in the morphometry of the coast, hydrology, temperature regime, concentration of dissolved oxygen, content of nutrients, the rate of production processes, qualitative and quantitative characteristics of the

flora and fauna of the narrow coastal zone (Zelinsky *et al.*, 1978).

One more factor should be included among the above factors causing a risky ecological situation in this part of the Black Sea. This is the destruction of the dam of the Kakhovka reservoir (06 June 2023). Thus, the Black Sea was flooded by the Dnieper waters that entered through the Dnieper-Bug estuary. Approximately two days after the dam explosion, a wedge of fresh water reached the sea coast causing a severe impact on the Black Sea ecosystem (Afanasyev 2023; Vyshnevskyi, 2023; Tuchkovenko *et al.*, 2023; Minicheva *et al.*, 2023).

This event affected the state of marine fauna, in particular the meiobenthos. The purpose of this study was (1) to study the structure of the meiobenthos community (quantitative indicators, biodiversity, (2) to study the use of meiobenthos and free-living marine nematodes as indicators of pollution in an area heavily damaged by the destruction of the Kakhovskaya hydroelectric power station dam.

2. MATERIAL AND METHODS

Study area and sampling sites

The studies were carried out in the coastal zone of the northwestern part of the Black Sea (Cape Lanzheron 30°76'587"S; 46°47'561"W; and Cape Maly Fontan 30°77'196"S; 46°44'855"W). Due to limited access to the study areas during this period due to military operations, it was possible to take samples of bottom sediments on Lanzheron Beach (Cape Lanzheron) in the upper sublittoral zone at a depth of 0.5 m at one station (point), a total of five sample in three replicates each: I (06/13/23), II (06/15/23), III (06/22/23), IV (06/26/23) and V (07/06/23). Samples were taken using a hand core (2.8 cm in diameter and 50 cm in height) with a depth of 5 cm into the soil (15 samples in total). In the area of the Biological Station (Cape Maly Fontan), sampling was carried in the period: I (06/29/23), II (07/15/23); III (07/28/23), IV (08/24/23) in the sublittoral at a depth of 1, 2, 3, 4 and 5 m on a loose (sand mixed with broken shells) and hard substrate, respectively (stones overgrown with algae). At each depth, samples were taken on hard and soft substrates in a total 40 samples. Samples were taken using a diver with a 10x10 cm frame lined with mill gas (64 µm). A total of 55 samples were collected (15 samples in Lanzheron plus 40 at Cape M. Fontan) (Fig. 1).

In the laboratory, the meiobenthos samples were washed through a system of sieves with mesh sizes of 1 mm, 0.250 mm, 0.100 mm. To capture meiofauna, a nylon mill sieves with a mesh size of 64 µm was placed on the lower sieve. The suspension with organisms was sorted from the sediment by flotation and transferred to a container, the sample volume was adjusted to 100 cm3 with distilled water with the addition of 4% formaldehyde and Rose-Bengal (0.2 g. l-1). Later on, the samples were sorted in a Bogorov chamber under stereomicrosope.

Fig. 1. Scheme of location of sampling sites in the coastal zone of the NorthWestern part of the Black Sea.

The number of organisms was recalculated for the entire sample (100 cm³) and for 1 m² of the bottom surface (Vorobyova, 1999). All meiobenthos groups were subjected to quantitative accounting. Each sample contained from several hundred to several thousand meiobenthos organisms. The biomass of nematode species was determined using nomograms (Chislenko, 1968).

To identify the nematodes, these were removed from the sample and placed in Seinhorst's liquid (Seinhorst, 1959) (distilled water – 70 parts, 96% ethanol – 29 parts and glycerol – 1 part) to clarify them. After a while, they were placed on a glass slide in a drop of glycerol-gelatin and covered with a coverslip (Platonova, 1968; Vincx, 1996). The preparations were studied using a Konus 5625 Biorex3 light microscope. For preliminary identification of nematodes, the works of Filipjev (1918-1921), Platonova (1968) and the NeMys database (Bezerra *et al.*, 2021) were used. The nomenclature of the species was verified and validated using the web portal https://www.marinespecies.org/ and the 'WoRMS Taxon Match Tool' (WoRMS Editorial Board, 2022).

For environmental quality assessment based on meiobenthic population, several pollution indicators were taken into account: total density, biomass, species richness, Abundance-Biomass Comparison plots (ABC) (Warwick, 1986), Nematode-Copepod Ratio (Raffaelli and Mason, 1981) and several ecological indicators based on diversity: Shannon-Weaver diversity, (H'), (Shannon and Weaver, 1963), evenness, (J') (Pielou, 1969), abundance of species, the socalled 'k- dominance' curves (Lambshead *et al.*, 1983) and on ecological strategies: index of trophic diversity, ITD; maturity index, MI; ecological status of environmental quality, EcoQ.

For the initial assessment of the trophic structure of nematode communities, the classification of Wieser (Wieser, 1953) was used. Index of trophic diversity (ITD), based on the proportion of each trophic group, and was calculated according to Heip *et al.*, (1985). A maturity index (MI) derived from the life history characteristics of nematode genera was

calculated for each accession according to Bongers (1990) and Bongers *et al.* (1991; 1995).

EcoQ status was assessed by the abundance (richness) of meiobenthic taxa as proposed by Danovaro *et al.* (2004) modified according to the WFD classes, while the thresholds reported by Moreno *et al.* (2011) and Semprucci *et al.* (2014a, b) were applied to evaluate EcoQ with nematodes.

MDS, ANOSIM, SIMPER and cluster analyzes were performed using the Primer® v.6 software package (Clarke and Warwick, 1994; Clarke and Gorley, 2006). To determine the structure of sand sediments, we used the method proposed by Strakhov (1953).

3. RESULTS

The deposits of the subtidal zone of the study areas are represented by medium- and coarse-grain sands, often with a mixture of shell detritus (on average up to 22%). The composition of the sand fractions was formed of mediumgrained sand (on average more than 40%), fine-grained sand – 23% on average of the sediment, and coarse-grained sand – 15%. The sand fineness modulus (*Mdk*) varied from 0.13 to 2 mm, with an average of 0.46 mm.

3.1. Cape Langeron (Langeron beach)

Meiobenthos. In the upper subtidal zone of 0.5 m, ten upper-level taxa of meiobenthos were found: eumeiobenthos (Foraminifera, Nematoda, Harpacticoida, Halacarida, Turbellaria, Kinoryncha) and pseudmeiobenthos (Oligochaeta, Polychaeta, Bivalvia, Amphibalanus). Each sample contained 6 to 8 taxa, among which Nematoda dominated as occurrence with a percentage ranging from 14% to 61% (Fig. 2a, b). The second most common species were Harpacticoida (73%) and Turbellaria (60%), and abundances that varied from 3 to 29% and from 6 to 35%, respectively. Halacarida and Kinoryncha were recorded with minimum occurrence (6.7%) and minimum abundances (10%). Among the temporary component of the meiobenthos, Oligochaeta and Polychaeta were noted with occurrence (60% each). The percentage of Oligochaeta abundance from the total meiobenthos was significant and varied from 0 to 68%. Polychaeta represented 1 to 16% of the meiobenthic community. During all periods of surveying, dominance in numbers of nematodes and oligochaetes was observed.

The values of the Shannon-Wiener (H') and Pielou's evenness (J') species diversity indices were generally low and varied from 0.59 to 1.35 (average 1.0 ± 0.12) and from 0.62 to 0.91 (average 0.77 \pm 0.05), respectively. Their low values were recorded in samples IV and V (Fig. 3a).

The abundance of meiofauna over the entire period of research varied from 4000 to 169830 ind./m2, averaging 28094 ± 10982.2 ind./m2; biomass from 40.9 to 9586.4 mg/m2 on average 331.8 \pm 427.8 mg/m². The minimum abundance of meiofauna was noted in survey II (9533 \pm 8972.4 ind./m²). It reached its maximum in survey III (66230 \pm 8972.4 ind./m²) (Fig. 3b).

When comparing meiofaunal structural features within each of the five surveys, average similarity values ranged from 31.2 to 61.9% (SIMPER analysis). The average similarity value in the taxocene of survey I was 52.9%. Among the seven taxa detected, Nematoda, Polychaeta, Oligochaeta and Turbellaria were the dominant ones, contributing 100% cumulatively to the average similarity. Their relative contribution to intracomplex similarity was 46.2, 33.4, 10.4 and 10%, respectively.

Taking into account the taxonomy of survey II, the average similarity value was similar to the previous survey and amounted to 50.4%. Of the eight meiofaunal taxa noted, Nematoda, Harpacticoida, Oligochaeta, and Turbellaria were dominant, contributing 100% cumulatively to the average similarity. Their relative contribution to intracomplex similarity was 40.8, 34.9, 13.1 and 11%, respectively.

For taxocene survey III, the average similarity was 60.1%. The dominant of the eight taxa are Nematoda, Turbellaria, Harpacticoida, and Oligochaeta, making 93.2% of the cumulative contribution to the average similarity. The relative contribution of each of them was 32.4 27.1, 26.8 and 7.1%, respectively.

Fig. 2. Frequency of occurrence (**a**) and relative abundance of meiofauna taxa. Mean values of replicated samples (n = 3) are shown (**b**).

Fig. 3. Meiofaunal univariate measures. (**a**) Shannon-Wiener (H') and Pielou's evenness (J'); (**b**) meiofauna abundance (N, ind./m2) and biomass (B, mg/m2) in the sublittoral zone of 0.5 m during the study periods.

In taxocene survey IY, the number of taxa decreases to six. The average similarity value was 61.9%. The number of dominant taxa making a relative contribution to intracomplex similarity is also reduced. These are Nematoda (54.6%), Harpacticoida (29.8%) and Oligochaeta (15.7%). Their cumulative contribution to the average similarity was 100%.

A decrease in the value of average similarity to 37.2% was noted in the taxocene of survey Y. Of the six taxa, only two taxa made a cumulative contribution to the average similarity. These are Nematoda and Bivalvia. They also made the greatest relative contribution to the intracomplex similarity (84.6% and 15.4%, respectively).

When comparing taxonocenotic complexes between surveys, the highest values of the level of difference were identified when comparing surveys I and Y (54.3%) and the lowest between III and Y (39.4%).

For a comparative assessment of the structure of meiofauna during different periods of surveying, *k*-dominance curves were used. K-dominance curves indicate the dominance of meiofaunal taxa in survey II, with 5 taxa accounting for 90% of their dominance. There is a noticeable decrease in diversity in survey IY: there is a pronounced dominance of only two taxa – up to 80%, which indicates a change in environmental conditions during this period (Fig. 4).

Nematodes. The nematofauna of the study area is represented by 24 species from 16 genera, 17 families and 5 orders (Table 1). The orders Enoplida and Monhysterida dominated in the number of species. It should be noted that the species diversity of nematodes was low in all periods studied (Fig. 5a). The Shannon index (H') varied from 0.2 to 1.2 (average 1.04 \pm 0.12) with a maximum value in the first surveys. The Pielou evenness index (J) values were high, ranging from 0.5 to 0.9 (mean 0.8 ± 0.01) also in the first surveys (Fig. 5b).

The results of comparison of quantitative parameters of nematodes during different survey periods revealed significant differences in their structure (ANOSIM, $R = 0.646$; $P = 0.1\%$). MDS plots show these differences. If in the first surveys (I, II, III) we see their scatter, then the last ones (IY, Y) are grouped (Fig. 6).

Fig. 4. Spatial variation of *k*- dominance curves of meiobenthos assemblage for different sampling dates (surveys).

Fig. 5. Contribution (%) of nematode orders: (**a**) Shannon-Wiener (H'), Pielou's evenness (J') (**b**) in the sublittoral zone of 0.5 m during the study periods.

Table. 1. Mean relative abundance (Ab,%), and biomass (B,%), frequency of occurrence (F,%), feeding types (FT) and coloniser-persisters classes (c-p%) in the sublittoral zone 0.5m of the Lanzheron beach.

Fig. 6. Multi-dimensional scaling (MDS) ordination for transformed square root nematode abundance on a two-dimensional scale at each station location.

The quantitative indicators of nematodes were not high and varied over a wide range from 0 to 35000 ind./m2, averaging 8756 ± 4353.1 ind./m2. The *k*-dominance curves showed the highest species diversity in the first two surveys (Fig. 7).

When comparing nematode structural features within each of the five surveys, average similarity values ranged from 18.2 to 83.6% (SIMPER analysis). The average similarity value in the taxocene of nematodes from survey I was 40.9%. Among the ten species detected, *Viscosia glabra*, was the dominant species, contributing 92.9% cumulatively to the average similarity. Their relative contribution to intracomplex similarity was 56.7 and 36.3%, respectively.

In the taxocene of survey II, the average similarity was low and amounted to only 18.2%. Of the eleven species noted, *Enoploides cirhatus* and *Enoplus littoralis* were dominant, making a 100% cumulative contribution to the average similarity. Their relative contribution to intracomplex similarity was 63.9 and 36.1%, respectively.

In the taxocene of survey III, the average similarity value increases to 60.1%. The dominant of the seven species are *Daptonema conicum* and *Enoplus littoralis*, making 100% of the cumulative contribution to the average similarity. The relative contribution to the intracomplex similarity of each of them was 77.8 and 22.2%, respectively.

In taxocene survey IY, the number of species decreases to four. The average similarity value was 83.6%. The three dominant species made relative contributions to intracomplex similarity. These are *Daptonema conicum* (42.9%), *Metachromadora arenaria* (28.7%), and *Gammarinema ampulocauda* (28.5%). Their cumulative contribution to the average similarity was 100%. The average similarity value in the Y survey taxocene was 63.2%. Of the three species, *Daptonema conicum* (100%) made a cumulative contribution to the average similarity. When comparing taxonocenotic complexes between surveys, the highest values of the level

of difference were identified when comparing surveys, I and Y (97.7%) and I and IY (100%). Low – between III and IY (65.4%).

Based on the type of nutrition, four trophic groups of nematodes were discovered. The dominant ones at most stations were $1B =$ non-selective deposit feeders (54.6%) and $2B =$ predator feeders (34.9%). Among the eight nematodes (1B), the highest abundance was observed for *Daptonema conicum* and *Sabatieria pulchra*. From group 2B, out of seven representatives, *Viscosia glabra* and *Enoplus littoralis* dominated in abundance (the former predominated in the IY and Y surveys, the latter in the I-III surveys. The index of trophic diversity (ITD) varied from 0.35 to 1 (average 0.65 \pm 0.906), reaching maximum values in IY and Y surveys (Fig. 8).

Representatives of the c-p1 class dominated (36%), with *Daptonema conicum* dominating in abundance. Representatives of the c-p2 and c-p3 classes, on the contrary, dominated in the first schemes, such as, *Sabatieria pulchra*, *Metachromadora arenaria* (c-p2), and *Viscosia glabra* (c-p3). Classes с-р4, с-р5 were recorded in surveys II – IY. The maturity index (MI) ranged from 1 to 4 (mean 2.13 ± 0.27). Its maximum values were noted in surveys II and III (Fig. 9).

To indicate disturbances in the structure of the nematode community in these studies, the Abundance-Biomass Comparison plots method was used. K-dominance curves for meiofauna abundance and biomass show a clear change in taxon structure from the first to the last survey (Fig. 10). Thus, in the first two surveys, when the supply of organic matter had not yet affected the meiofauna, *k*-dominance curves for species biomass and abundance (ABC curves) showed a stable undisturbed community of the meiofauna (unpolluted) (the biomass curve is located above the abundance curve).

Relatively large taxa of organisms predominated: Polychaeta, Oligochaeta and Turbellaria. We then observe a sharp increase in the abundance of Oligochaeta in survey III, which determined an increase in biomass over abundance despite the fact that Nematoda, Turbellaria and Harpacticoida were also present.

Fig. 7. Ranked species *k*-dominance curves for the free-living nematode species in the different period survey.

Fig. 8. Trophic composition of the nematode community. Based on the feeding guilds of Wieser (1953): 1A = selective deposit feeders, 1B = non-selective deposit feeders, 2A = epistrate feeders, 2B = predators (**a**) and Trophic Diversity Index (ITD) (**b**).

Fig. 9. Percentage of nematode coloniser-persisters classes revealed in the study area (**a**) and Maturity index (MI) (**b**).

In the IY and Y survey we see that the abundance and biomass curves overlap, indicating the complex, unstable environment in which the meiofauna evolves.

A high density of oligochaetes was noted during survey III (06/22/2023), as well as a general decrease in the density of pseudomeiobenthos during the last two surveys. This may be due to significant fluctuations in salinity from 4.5 to 15.4‰ (Fig. 10).

K-dominance curves for nematode abundance and biomass (ABC) also show changes in their structure from the first to the last survey (Fig. 11).

In surveys I and II, the abundance and biomass curves coincide, but in surveys III and IY they overlap, which indicates environmental instability. In the latter, we observe the dominance of density over biomass (the biomass curve is located below the abundance curve). As well as a reduction in the species diversity of nematodes.

3.2. Cape Maly Fontan (Hydrobiological station)

Meiobenthos. Representatives of the permanent component (eumeiobenthos) of the meiobenthos (Foraminifera, Nematoda, Harpacticoida, Halacarida, Ostracoda, Turbellaria) and representatives of the temporary component (pseudomeiobenthos) (Oligochaeta, Polychaeta, Bivalvia, Balanus) were found in the meiobenthos. In each survey there were found between 5 to 8 taxa. Among eumeiobenthos, Nematoda was the dominant taxon in terms of occurrence (100%). The second most common were Harpacticoida (93.5%) and Halacarida (84.8%). Among the temporary component of the meiobenthos, Polychaeta (97.8%), Bivalvia (89.1%) and Balanus (80.4%) dominated in occurrence. The Shannon index (H') varied from 0.50 to 1.90 (average 1.29 \pm 0.06) and the Pielou evenness index (J) from 0.29 to 0.91 (average 0.67 ± 0.03 (Fig. 12).

The results of comparison of the structure of the meiobenthos community during different survey periods did not reveal significant deviations. The average value of rank similarity between all stations (ANOSIM, R-statistics) was only 0.011 with a significance level of 0.56%. A comparison of stations at different depths also showed a low difference between meiobenthos communities.

Fig. 10. K-dominance curves for biomass and abundance (ABC curves) of meiobenthos (**a**-**e** corresponds to I-Y survey) and relative abundance of eu- and pseudomeiobenthos in the sublittoral zone 0.5m on the Lanzheron beach (**f**).

The average value of rank similarity between all stations (R-statistics) was only 0.103 with a significance level of 1.4%. There was a difference in the structure of meiobenthos between the loose and hard substrates (ANOSIM, $R =$ 0.396; $P = 0.1\%$). On the MDS graph we see: stations with hard substrate are grouped at the top left and less similar meiobenthos communities on loose soils are grouped on the right (Fig. 13).

A special feature of the meiobenthos in the Maly Fontan is the high density of all its representatives throughout the entire study period. It varied from 114000 to 9708310 ind./ m2 and biomass from 1581.6 to 55157.5 mg/m2, averaging 1753412 \pm 261350.7 ind./m² and 19815.6 \pm 1734.8 mg/m². The numbers and biomass of meiobenthos increased from the first sample to the last one (from 1336163 to 2083333 ind./m2 and biomass from 152614 to 23828.5 mg/m2) (Fig.14).

The share of eumeiobenthos averaged 77% and pseudomeiobenthos – 23%. The main share in the total number of meiobenthos was determined by Nematoda. Their density varied from 657675 \pm 263589.6 ind./m² to 1303665 \pm 689914.0. ind./m². They reached their maximum in the II survey.

Fig. 11. K-dominance curves for species biomass and abundance (ABC curves) nematods in the sublittoral zone 0.5 m on the Lanzheron beach $(I - Y$ survey).

Fig. 12. Frequency of occurrence meiofauna taxa (**a**) and diversity indices: Shannon-Wiener (H'), Evenness (J') (**b**) in the sublittoral zone 1-5 m on the Biostation beach.

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Fig. 13. Multi-dimensional scaling (MDS) ordination for transformed Fourth root meiofaunal abundance on a two-dimensional scale at each station location.

Fig. 14. Average density and biomass of meiobenthos (**a**) and average density and biomass eu- and pseudomeiobenthos in the sublittoral zone 1 – 5 m on the Biostation beach (**b**).

Harpacticoida were subdominant in abundance, varying from 93248 \pm 18672.9 ind./m² to 347500 \pm 92590.0 ind./m². Their numbers increased noticeably in the III and especially in the IV survey.

In the pseudomeiobenthos, juveniles of Bivalvia dominated in density (from 84905 \pm 21104.3 ind./m² to 351667 \pm 101753.3 ind./m². There is a tendency for their density to increase from the first survey to the last. The number of Polychaeta was distributed evenly in all surveys and ranged from 121545 \pm 28344.7 up to 161667 \pm 38882.8 ind./m2 (Fig. 15).

When comparing the structural features of the meiofauna within each of the four surveys, their average similarity values were large and ranged from 67.6 to 71.4% (SIMPER analysis). Nematoda, Polychaeta, Harpacticoida and Bivalvia were the dominant taxa detected, contributing 73% cumulatively to the average similarity. The values of the level of difference when comparing taxonocenotic complexes between surveys were low (from 30.6 to 31.9%). High values of average similarity were also noted when comparing meiofauna at different depths (from 61.3 to 75.2%).

When comparing the diversity of meiofaunal taxa in different survey periods, *k*-dominance curves showed the greatest diversity in surveys I and III (Fig. 16).

K-dominance curves indicate the dominance of six taxa, which determine 90% of their dominance. These are Nematoda, Polychaeta, Harpacticoida, Ostracoda, Bivalvia, and Balanus. In the second survey, there was a reduction in dominant taxa. In survey IY, there is a pronounced dominance of three taxa (up to 90%) – Nematoda, Harpacticoida and Bivalvia, which indicates a change in environmental conditions during this period.

Fig. 15. Relative abundance of eumeio- and pseudomeiobenthos in the different period survey in the sublittoral zone 1 – 5 m on the Biostation beach.

Fig. 16. Ranked species K-dominance curves for the taxon's of meiobenthic in the different period survey.

Nematodes. The spatial distribution and species diversity of nematodes in the Biostation area were analyzed based on materials from survey I (06/29/2023). The nematofauna in the Biostation area is represented by 33 species from 6 orders (Table 2). The orders Monhysterida, Enoplida and Chromadorida dominated in the number of species. Shannon index (H') values ranged from 0.8 to 2.3 (average 1.91 ± 0.13) (Fig. 17).

Fig. 17. Diversity indices: Shannon-Wiener (H'), species richness (**a**) and percentage of nematode orders (**b**) in the sublittoral zone 1 – 5 m on the Biostation beach.

Table 2. Relative abundance (Ab%), biomass (B%), frequency of occurrence (F%), feeding types (FT) and coloniser-persisters classes (c-p%) in the sublittoral zone $1 - 5$ m on the Biostation beach.

The results of comparison of quantitative parameters of nematodes between stations revealed significant differences in their structure (ANOSIM, $R = 0.676$; $P = 0.8\%$). The difference between stations by substrate is shown on the MDS graph. The similarities within stations can be explained by the similar nematofauna and environmental conditions (Fig. 18).

The proportion of nematode density from the total meiobenthos on rocky substrates at the studied depths varied from 11.2 to 35.4% (on average 21.4%), while on loose soils it ranged from 17 to 89.2% (on average 55.7%). The average density of nematodes on rocky substrate was 195751 \pm 53642.4 ind./m². This is almost 6 times less than on loose soils, where it was recorded on average 1117589 \pm 451476.6 ind./m2 (Fig. 19).

Fig. 18. Multi-dimensional scaling (MDS) ordination for transformed Fourth root nematode abundance on a two-dimensional scale at each station location.

Fig. 19. Distribution of the density and biomass of nematodes on hard and soft substrates.

Based on feeding type, four trophic groups of nematodes were identified and the index of trophic diversity (ITD) was calculated based on their dominance in the stations (Fig. 18). Epistrate feeders (2A) dominated at most stations (39%). Of the 11 representatives of this group, *Metachromadora arenaria* and *Neochromadora poecilosomoides* ranked the first ones Subdominant group was constituted of non-selective deposit feeders (1B) (31%).

The leaders in abundance were *Daptonema maeoticum*, *Valvaelaimus euxinus*, *Daptonema conicum*, and *Axonolaimus setosus*. Selective deposit feeders (1A) accounted for 19%. Of these, *Anoplostoma viviparum* dominated. Group 2B = predators made up 11% with dominance in numbers of *Viscosia glabra*, *Enoploides brevis*. The index (ITD) in the study area varied from 0.34 to 0.66, averaging 0.43 ± 0.03 (Fig. 20).

 The MI values in the study area varied from 1.75 to 2.41 (on average 2.15 \pm 0.06). Representatives of the c-p1 class made up 4%, however, the number was high (*Daptonema conicum*). Class c-p2 was the most common, accounting for 67%.

Fig. 20. Trophic composition of the nematode community. Based on the feeding quilds of Wieser (1953): 1A = selective deposit feeders, 1B = non-selective deposit feeders, 2A = epistrate feeders, 2B = predators (**a**) and Trophic Diversity Index (ITD) (**b**).

The leaders in numbers were *Metachromadora arenaria*, *Neochromadora poecilosomoides*, *Daptonema maeoticum*, *Valvaelaimus euxinus*, *Anoplostoma viviparum*, *Axonolaimus setosus*. These are species that typically live in highly eutrophic environments. Among the class c-p3, which made up (23%), *Chromadora nudicapitata*, *Viscosia glabra*, *Chromadorita demaniana* turned out to be were noted.

Classes c-p4 (very sensitive to stress) (3%) and c-p5 (3%) were also recorded, but their numbers were not high. Among c-p4 we can note *Pomponema multipapillatum*, *Oncholaimus dujardinii*, *Oncholaimus campylocercoides*. Among c-p5 – *Enoplus littoralis* (Fig. 21).

Meiofauna abundance and biomass *k*-dominance curves (ABC) indicate the complex, unstable environment in which meiofauna develop (abundance and biomass curves overlap) (Fig. 22).

Fig. 21. Percentage of nematode coloniser-persisters classes revealed in the study area (**a**) and Maturity index (MI) (**b**).

Fig. 22. K-dominance curves for meiobenthos biomass and abundance (ABC curves) in the sublittoral zone 1 – 5 m on the Biostation beach.

The results of comparing the quantitative parameters of nematodes between stations with different substrates revealed significant differences in their structure (ANOSIM, $R = 0.676$; $P = 0.8\%$). K-dominance curves for abundance and biomass (ABC) demonstrate disturbances in the structure of the nematode community both in soft and hard substrates. In the graphs (Fig. 23), the curves of abundance and biomass overlap, which indicates environmental instability, stress, when the size structure is being restructured.

Nematodes, due to certain biological characteristics (high productivity, short life cycles), quickly respond to environmental changes, reducing species diversity, increasing numbers and decreasing biomass (Lambshead *et al.*, 1983; Warwick *et al.*, 1994).

Nematode-Copepod Ratio. As the bottom eutrophicates or silts, the ratio of the density of nematode and harpacticid settlements changes – the proportion of nematodes in the meiobenthos increases, and the harpacticid decreases (Warwick, 1981).

Fig. 23. K-dominance curves for nematod biomass and abundance (ABC curves) on different types of substrates in the sublittoral zone 1 – 5 m on the Biostation beach.

High N/H values were noted in the second and third surveys (July 15 and July 28), where they reached values of 12.4 and 13.7, respectively, indicating the predominance of nematodes in the meiobenthos. Already in the fourth survey (August 24), we observe an increasing role of harpacticoids in the total number of meiobenthos. The N/H indicators decreased and amounted to only 2.1, which indicates an improvement in the environment for their development (Fig. 24).

Fig. 24. Dynamics of nematode-copepod ratio indices in the sublittoral zone of 1 – 5 m on the Biostation beach.

A special feature of the meiobenthos in this area is the high density of all its representatives throughout the entire study period.

Nematodes (56%) and harpacticoids (10%) dominated in eumeiobenthos in terms of density; in pseudomeiobenthos, juvenile bivalve mollusks (10%) and polychaetes (8%) dominated. Meiobenthos density varied from 1336163 to 2083333 ind./m2 and biomass from 15261.4 to 23828.5 mg/m2 (average 1736027 ind./m2 and 19,815.6 mg/m2). It increased almost 8 times compared to the regional average (period 2012 – 2019) (Fig. 25).

The response of meiobenthic assemblages (especially nematodes) following environmental change, both in terms of density and diversity, demonstrated significant potential for persistence and recovery in the Biostation area in the 1 – 5 m subtidal zone.

Using the ecological quality (EcoQ) classification based on threshold values of taxonomic richness of meiobenthos

(Moreno *et al.* 2011), the study area falls into Moderate status (Table 3). According to the indicators of the Shannon diversity index (H'), maturity index (MI), c-p%, index of trophic diversity (ITD) of nematodes, the region of the Biostation can generally be classified as poor (Poor) status and the Lanzheron region can be classified as bad (Bad).

According to the values of the maturity index (MI) and the index of trophic diversity (ITD), the water area of Cape Langeron during the study period changed the status of ecological quality of the environment (EcoQ) from Good (I-III survey) to Poor (I-Y survey) (Fig. 26).

Fig. 26. Status of ecological quality of the environment (EcoQ) in the upper sublittoral zone of Cape Langeron during the study.

4. DISCUSSION

The anthropogenic eutrophication in the northwestern part of the Black Sea affects the structure and quantitative indicators of meiobenthos. The consequences of anthropogenic impacts on marine ecosystems can be assess using different biological indicators. Internal restructuring of hierarchical structure of ecosystems is reflected by their component biocenoses (cenoses, communities). Biological indicators can be used to assess the consequences of anthropogenic impact (stress) – acute and chronic. In the context of acute stress induced by the destruction of the

dam of the Kakhovka reservoir (06 June 2023), we analyzed the main trends in the development of meiobenthos and, in particular, of taxocene Nematoda. Increased deposition of suspended matter has led to changes in the granulometric composition of the sediment that became finer as with the siltation of the soil. Shifts in the abiotic parameters of the marine environment entail significant changes in the structure and functioning of the entire benthic community of organisms.

It has already been noted in the literature that meiobenthic organisms respond to the state of the environment by changing their overall density and the ratio of different groups. Thus, after 1983, increased dominance of nematodes has been observed in the Northwestern Black Sea. They often make up 85 – 100% of the total meiobenthos, i.e. there is a change in its qualitative composition. (Vorobyova, 1999; Vorobyova, Kulakova, 2009). The authors noted that in areas of the NWBS with different anthropogenic loads, the qualitative composition of meiobenthos and its quantitative indicators differ significantly from each other (Vorobyova *et al.*, 2017).

After the destruction of the platinum of the Kakhovskaya hydroelectric power station, the entry of fresh polluted water into the marine ecosystem led in the first days to a dramatic decrease of salinity of the sea on the Odessa coast down to 3.95‰, a decrease in the oxygen saturation to 75% of the surface layer, an increase in the concentration of ammonia nitrogen to 13.8 MPC, and the occurrence of acute lethal toxicity. An increase in the concentration of chlorophyll a and the massive development of blue-green planktonic algae were registered. The salinity of bottom water varied from 4 to 17‰. (Minicheva *et al.*, 2023).

Meiofauna of the sublittoral zone of Cape Langeron. It should be noted that the species diversity of nematodes was low in all periods studied in the coastal waters of Lanzheron Beach. The Shannon index (H') values varied from 0.2 to 1.2.

Table 3. The threshold values used to evaluate EcoQ according to Danovaro *et al.*, (2004); Moreno *et al.*, (2011) and Semprucci *et al.*, (2014a, b) and EcoQ status of the studied water areas.

The quantitative indicators of nematodes were not high and varied in a wide range from 0 to 35000 ind./m2, averaging 8756 \pm 4353.1 ind./m². Low numbers of meiobenthos and nematodes are characteristic of the upper sublittoral zone of 0.5 m, which is constantly exposed to waves and substrate mixing. The sediments were generally characterized by coarse and medium particle sizes, which was associated with tidal activity that washed sand away from fine particles. Sediment grain size is an important environmental factor that influences the species structure of meiofauna (Wieser, 1960; Giere, 1993; Vanaverbeke *et al.*, 2002).

When comparing the structural features of the meiofauna within each of the five surveys (SIMPER analysis), a decrease in the average similarity value of 61.9 to 31.2% was noted from the first to the last survey. K-dominance curves also indicate this. There is a noticeable decrease in diversity in survey IY: there is a pronounced dominance of only two taxa – up to 80%, which indicates a change in environmental conditions during this period. Nematodes, due to certain biological characteristics (high productivity, short life cycles), quickly respond to environmental changes, reducing species diversity, increasing numbers and decreasing biomass (Lambshead *et al.*, 1983; Warwick *et al.*, 1994).

When comparing nematode structural features within each of the five surveys, average similarity values ranged from 18.2 to 83.6% (SIMPER analysis). A change in the species structure of nematodes was detected. Thus, in the first survey, *Viscosia glabra*, *Sabatieria pulchra* were dominant (92.9% cumulative contribution to the average similarity). In the latter, these are *Daptonema conicum* (42.9%), *Metachromadora arenaria* (28.7%) and *Gammarinema ampulocauda* (28.5%). These similarity analysis results complement the ABC results. That is, a replacement occurred in the structure of the taxocene of nematodes. K-strategist nematodes were replaced in the last two surveys by r-strategists (short generation period, small size, presence of numbers, resistance to disturbances is their characteristic features).

Dominance at most stations was recorded by $1B = non$ selective deposit feeders (54.6%) (*Daptonema conicum* and *Sabatieria pulchra*) and an increase in the index of trophic diversity (ITD) to 1 in IY and Y surveys, which may indirectly indicate poor environmental conditions. The predominance of class c-р1 (36%) with dominance in abundance of *Daptonema conicum* and a decrease in the maturity index (MI) were also noted in the latest surveys.

The influx of freshwater and an increase in organic matter on the coast of Lanzheron beach after the destruction of the dam at the Kakhovskaya hydroelectric station negatively affected the composition of meiobenthos. The low occurrence of its groups and the high percentage of dominance of Nematoda and Oligochaeta suggests the sensitivity of other meiofaunal groups to a dynamic habitat compared to the above-mentioned organisms. (Heip *et al.*, 1985; Vranken and Heip, 1986; Coull and Chandler, 1992).

Cape Maly Fontan (Biostation beach). When comparing the structural features of the meiofauna within each of the four surveys in the subtidal zone at a depth of $1 - 5$ m, the values of their average similarity were high and ranged from 67.6 to 71.4% (SIMPER analysis). Nematoda, Polychaeta, Harpacticoida and Bivalvia were the dominant taxa detected, contributing 73% cumulatively to the average similarity.

Among the nematode species, epistrate feeders (2A) (39%) dominated at most stations. Of the 11 representatives of this group, *Metachromadora arenaria* and *Neochromadora poecilosomoides*. Subdominant group of non-selective deposit feeders (1B) (31%) (*Daptonema maeoticum*, *Valvaelaimus euxinus*, *Daptonema conicum*, *Axonolaimus setosus*).

The MI values in the study area were recorded to be low, varying from 1.75 to 2.41 (on average 2.15 ± 0.06). Class c-p2 was the most common, accounting for 67%. (*Metachromadora arenaria*, *Neochromadora poecilosomoides*, *Daptonema maeoticum*, *Valvaelaimus euxinus*, *Anoplostoma viviparum*, *Axonolaimus setosus*). These are species that typically occur in highly eutrophic environments (Herris and Bongers, 2009). Classes c-p4 were also recorded, but their numbers were not high (*Pomponema multipapillatum*, *Oncholaimus dujardinii*, *Oncholaimus campylocercoides*). Species classified in this class are very sensitive to pollutants and other disturbances in the marine meiobenthos (Warwick 1986; Bongers, 1990). Increased food supply also promotes the rapid proliferation of species with higher reproductive potential, resulting in reduction in index of MI maturity (Bongers *et al.*, 1991).

A feature of the meiobenthos in this area is the high density of all its representatives throughout the entire study period compared to previous years (Fig. 25). High abundance of meiobenthos (especially nematodes) can be explained, firstly, by a decrease in the anthropogenic factor in the coastal zone (closing beaches during the war period), which contributed to improved conditions for the development of meiobenthos due to its ability to quickly restore numbers and biodiversity and secondly, by an increase in eutrophicity in the sublittoral zone (1 – 5 m), caused by the consequences of the destruction of the dam. The concentrations of phosphates and silicic acid, important nutrients, on the Odessa coast in June at a seawater salinity of 4‰ were 2–3 times higher than the longterm average values. High concentrations of phosphates were also noted in July 2023 in the M. Fontan area, which may be associated with the destruction and decomposition of dead organic matter of phytoplankton during the period of its mass development (Minicheva *et al.*, 2023). The response of nematode assemblages to environmental changes, both in terms of density and diversity, has demonstrated significant potential for persistence and recovery.

It is necessary to note the increase in the density of harpacticoids and representatives of pseudomeiobenthos during the last survey (08/24/2023), which may be the result of improved environmental conditions for their development by this time, i.e. (Fig. 15).

The use of meiobenthos and free-living marine nematodes turned out to be an effective tool for assessing the ecological state of the study area, which was heavily damaged by the destruction of the Kakhovskaya hydroelectric power station dam. This conclusion can be supported by studies carried out in the Mediterranean harbor area (Ancona, Adriatic Sea, Italy) (Baldrighi *et al.*, 2023). A multi-benthic approach, which was used to identify different environmental conditions, showed that meiofauna were likely to be more sensitive to the effects of environmental features and pollutants than macrofauna.

5. CONCLUSION

The influx of freshwater and an increase in organic matter in the coastal waters of Cape Lanzheron after the destruction of the dam at the Kakhovskaya hydroelectric station affected the composition of the meiobenthos. The low occurrence of its groups and the high percentage of dominance of Nematoda and Oligochaeta suggests the sensitivity of other meiofaunal groups to a dynamic environment compared to the above-mentioned organisms. According to the values of the maturity index (MI) and the index of trophic diversity (ITD), the water area of Cape Langeron during the study period changes the status of environmental quality of the environment (EcoQ) from Good (I-III survey) to Poor (IY-Y survey).

The increase in eutrophication in the waters of Cape Maly Fontan in the sublittoral zone $(1 – 5 m)$, as consequences of the disaster, contributed to an abnormally sharp increase in the quantitative indicators of meiobenthos (especially nematodes). Nevertheless, the response of nematode assemblages to environmental change, both in terms of density and diversity, has demonstrated significant potential for persistence and recovery.

The increase in the density of harpacticoids and representatives of pseudomeiobenthos during the last survey (08/24/2023) may be evidence of their ability to quickly recover when environmental conditions improve.

In general, the status of the ecological quality of the environment (EcoQ) of the study area can be classified as bad (Bad) in the Langeron area and poor (Poor) in the Biostation area.

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