



Integrating Taxonomic and Functional Responses of Free-Living Marine Nematodes for Coastal Pollution Assessment and Ecosystem Health Monitoring

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Abstract

Free-living marine nematodes are widely used as bioindicators, yet comparative studies integrating taxonomic, functional, and multivariate community responses across pollution gradients remain limited. This study evaluated changes in nematode assemblage responses along a regional coastal disturbance gradient and investigated their applicability for ecological monitoring. Fifteen sampling stations were classified into high, intermediate, and low disturbance levels. The nematode assemblages were studied at the genus level through diversity indices, trophic guild structure, c-p strategy, and functional diversity. High-disturbance sites showed significant reduction in genus richness, Shannon diversity, and Maturity Index compared with low-disturbance reference sites. Opportunistic bacterivores and c-p 1–2 group taxa, particularly members of Xyalidae and Comesomatidae, dominated highly disturbed assemblages, while predator–omnivore taxa declined from 18% at low-disturbance sites to 4% at high-disturbance sites. Functional richness and Rao's quadratic entropy had more significant reductions than taxonomic diversity, where the functional trait space of severely disturbed sites was 23% that of reference sites. Multivariate analyses further indicated strong associations between nematode community restructuring and environmental disturbance. Overall, the findings indicated that the inclusion of functional trait metrics with the Maturity Index can enhance sensitivity and ecological interpretation of nematode-based coastal pollution assessment and monitoring of ecosystems.

These results demonstrate that the functional trait-based approach provides enhanced sensitivity for determining ecological damage compared with traditional taxonomic indicators. Integrating the maturity index measurement with the functional trait approach in the monitoring strategy is thus effective and feasible in coastal pollution assessment and management.

Keywords: *marine nematodes; bioindicators; colonizer-persisted index; coastal pollution; multivariate ordination; sediment ecology; ecosystem health monitoring*

1. Introduction

Coastal marine ecosystems form dynamic interfaces between terrestrial and oceanic processes and are among the most productive yet vulnerable ecosystems globally (1). They support commercially important fish and invertebrate populations, maintain high biological and functional biodiversity, and contribute to essential biogeochemical cycles such as nitrogen cycles, denitrification, and carbon sequestration. However, rapid industrialization, intensive agriculture, urban expansion, and increasing wastewater discharge have intensified anthropogenic pressures on coastal environments, resulting in the persistent accumulation of heavy metals, organic contaminants, and excess nutrients in marine sediments (2-4).

The ecological consequences of these stressors occur across multiple levels. The geochemistry of sediments can be affected by inputs of contaminants, which can also lead to oxygen depletion by increased microbial respiration of organic matter and change sediment structure and habitat complexity (5-7). These physicochemical shifts can subsequently restructure biological communities by favoring disturbance-tolerant and opportunistic taxa while decreasing sensitive organisms, ultimately simplifying community organization and altering ecosystem functioning (8). Therefore, reliable biological assessment methods are needed to assess ecological responses to cumulative and episodic disturbances and to inform the effective management of coastal resources.

Traditional environmental monitoring programs have relied primarily on physicochemical indicators, including dissolved oxygen, nutrient concentrations, turbidity, sediment characteristics, and contaminant levels (1). While these variables can give important information on environmental exposures, they may not be comprehensive enough to represent the cumulative biological impacts of long-term exposure to multiple stressors associated with complex contaminant mixtures and changing environmental conditions (9, 10). Biological indicators are used alongside physicochemical measurements and incorporate organismal and community-level responses to environmental change (11). The potential benthic bioindicators that have been given considerable attention include free-living marine nematodes due to their ecological importance, abundance, diversity, and sensitivity to environmental disturbance.

Marine nematodes are among the most abundant metazoan elements of the marine meiofaunal communities, ranging from 105 to 106 individuals m⁻² across a broad spectrum of sediment habitats. They occupy diverse trophic positions, including microbial grazers, deposit feeders, epistrate feeders, predators, and omnivores (2). Their short generation times enable assemblages to change quickly in response to environmental changes, which can make them of potential value in the detection of acute and chronic disturbance (12). Furthermore, many aquatic nematode taxa do not have extensive dispersal, and local assemblage structure may closely reflect local environmental conditions, which results in relatively high spatial resolution for ecological assessment (13).

The development of nematode-based biomonitoring has been strengthened by life-history approaches, especially

the colonizer–persister (c–p) framework developed by Bongers (1990). In this framework, nematode taxa are divided into groups based on ecological strategy; colonizers (c–p 1–2) have small body size, a tolerant response to environmental stress, and rapid reproduction, and dominate environments that are stressed and/or resource-enriched (14). Colonizer taxa, particularly c–p 1–2 groups, generally exhibit rapid reproduction and relatively high tolerance to environmental stress and resource enrichment, whereas higher c–p groups are characterized by slower life cycles and greater sensitivity to environmental disturbance. The relative representation of these strategies can be obtained by the Maturity Index (MI), which combines the taxon-specific c–p values with the relative abundance and represents the community-level disturbance.

An alternative functional approach can be obtained from trophic guild analysis, which groups nematodes based on feeding mode and buccal morphology (15). The trophic classification system developed by Wieser (1953) and subsequently modified by other researchers distinguishes selective and non-selective deposit feeders, epistrate feeders, and predators/omnivores. Trophic shifts can signify energy pathway changes, resource depletion, food-web complexity, and ecosystem functioning changes (16). For example, organic enrichment may favor bacterivorous and deposit-feeding taxa, whereas severe environmental stress may lead to a decrease in higher trophic levels like predators and omnivores.

More recently, functional trait-based approaches have expanded nematode biomonitoring beyond conventional taxonomic measures by incorporating ecological characteristics related to life-history strategy, feeding mode, morphology, and environmental tolerance. Functional diversity metrics, such as functional richness (FRic), functional evenness (FEve), and Rao's quadratic entropy (Q), provide information on the distribution of ecological traits within communities. Such metrics may capture processes like 'convergence of traits', 'loss of functional redundancy' and 'environmental filtering' that are not necessarily reflected in taxonomic diversity. Thus, the functional approaches can supply complementary information related to ecosystem degradation and ecological restructuring under anthropogenic stress.

Despite these methodological advances, important gaps remain in the integrated assessment of marine nematode responses to coastal pollution. Previous studies have commonly examined taxonomic diversity, trophic structure, life-history strategies, functional traits, or environmental relationships separately. Fewer studies have integrated these dimensions across clearly defined pollution gradients while accounting for habitat-related confounding factors, such as sediment characteristics (17). Furthermore, very few studies have been conducted to directly compare the responses of conventional taxonomic indicators with functional diversity measurements under the same sampling design. Addressing this gap is important for determining whether multi-indicator nematode assessments offer more information for coastal ecosystem monitoring than single ecological indicators do.

Accordingly, the present study investigates free-living marine nematode communities along a coastal disturbance gradient, using a combination of measures of taxonomic diversity, trophic guild structure, coloniser–persister life-history strategies, functional trait diversity and multivariate community–environment relationships. Specifically, the study aims to: (1) characterize nematode community composition and taxonomic diversity across disturbance levels; (2) quantify changes in trophic group, c–p strategy, and structure of the functional traits; (3) identify the environmental variables most strongly associated with nematode community using multivariate analyses; and (4) compare the responses of taxonomic and functional indicators to anthropogenic disturbance.

2. Materials and methods

2.1 Study Area and Site Selection

The study was conducted in a coastal embayment where there is a gradient in anthropogenic stress from coastal industrialized and urbanized areas to relatively undisturbed marine protected areas. Industrial activities, municipal wastewater discharge, and diffuse agricultural runoff are sources of input to the region. Fifteen sampling sites were chosen and classified as high disturbance (HD, $n = 5$), intermediate disturbance (ID, $n = 5$), and low disturbance reference (LD, $n = 5$). In the 12 stations located within 500 m of industrial discharge areas or municipal effluent-influenced depositional zones (12), contamination was found to be heavier than in other stations. The contamination was heavier in 12 stations located within 500 m of industrial discharge areas or municipal effluent-influenced depositional zones (12). ID stations were placed in transitional areas where contamination from diffuse sources from urban and agricultural activities was present, and LD stations in a protected area with a low anthropogenic influence. Differential GPS was used to collect geographic coordinates.

2.2 Sediment Collection and Processing

During austral autumn and compare samples from Ho Chi Minh city, to minimize seasonal variation, sampling was carried out. At each station, three replicate sediment cores (5 cm inner diameter, 12 cm depth) were taken at a depth of 3–8 m with SCUBA-assisted push coring (18). Sediment cores were subdivided into 0–2, 2–5, and 5–10 cm after a period of less than 30 min. Nematode analysis was done on the top 0–2 cm of sediment. Samples were fixed in 4% buffered formaldehyde, decanted, and sieved through a 38 μm mesh. Nematodes were isolated by density gradient centrifugation in Ludox HS-40 and mounted on permanent glycerol slides.

Simultaneous sediment physicochemical analyses were conducted on parallel samples. Laser diffractometry was used to measure the grain size following organic matter and carbonate removal. The total organic carbon (TOC) and total nitrogen (TN) were measured by CHN elemental analysis. Heavy metals (lead, copper, zinc, cadmium, chromium and nickel) were determined by ICP-MS following microwave digestion with acids. All analyses included both certified reference materials and procedural blanks.

2.3 Taxonomic Identification and Functional Classification

Nematodes have been identified to genus level, using transmitted light microscopy (400–1000 $\times$ magnification) following the standard taxonomic keys. At least 150 individuals/sampling were identified, with an average of 183 ± 24 individuals/sampling. To determine the accuracy of identifications, 10% of the identified specimens were checked by a second taxonomist.

The genera identified were grouped into trophic guilds following the classification of Wieser (1953), with changes introduced by Moens and Vincx (1997) and Hamels et al. (2001). Four trophic groups were taken into account: selective deposit feeders (1A), non-selective deposit feeders (1B), epistrate feeders (2A), and predators/omnivores (2B) (Table 1). The genera were assigned colonizer–persister values of 1 through 5 based on Bongers (1990) and Bongers et al. (1995). These classifications were adopted to investigate life history strategies and disturbance responses of the communities.

Table 1: Marine Nematode Trophic Guild Classification

Trophic Guild	Code	Buccal Morphology	Primary Resource	Example Genera
Selective deposit feeders	1A	Small, unarmed or weakly armed	Fine detritus, bacteria	Tobrilus, Tripyla, Theristus
Non-selective deposit feeders	1B	Simple, cylindrical, no teeth	Bacteria, fine organic particles	Sabatieria, Dorylaimopsis, Terschellingia
Epistrate feeders	2A	Equipped with hollow teeth/stylet	Diatoms, algae, epiphyte growth	Chromadora, Euchromadora, Desmodora
Predators / Omnivores	2B	Large, heavily armed, often with teeth	Meiofauna, macrofaunal juveniles	Oncholaimus, Eurystomina, Enoploides

2.4. Diversity and Functional Indices

The taxonomic diversity was calculated as genus richness (S), Shannon diversity index (H'), Pielou's evenness (J'), and Margalef richness index (d). The FD package (Laliberte and Legendre, 2010) in R 4.3.1 was used to measure functional diversity, which was based on a functional trait matrix that included: (1) c-p value (ordinal, 1–5); (2) trophic guild (categorical, 4 levels); (3) adult body size class (ordinal, small/medium/large based on published genus averages); and (4) cuticle ornamentation type (categorical, smooth, annulated, and ornamented). The functional richness (FRic) matrix was calculated using c-p value, trophic guild, body size category in the adult stage, and cuticle ornamentation. Distributions of functional traits were quantified as functional richness (FRic), functional evenness (FEve), functional divergence (FDiv), and Rao's quadratic entropy (Q). The ecological condition of nematode communities was also evaluated by the Maturity Index (MI), Nematode Channel Ratio (NCR), and NemaSPEAR[%]. The NCR was calculated as the proportion of bacterivores relative to bacterivores plus fungivores, while NemaSPEAR [%] represented the proportion of contaminant-sensitive genera within each assemblage.

2.5 Statistical Analyses

Data analysis was carried out in R version 4.3.1 with the following packages: vegan, FD, mvabund and ggplot2. Data on abundance of nematodes were fourth-root transformed before community analyses. Non-metric multidimensional scaling (nMDS) and hierarchical clustering were used for the visualization of Bray–Curtis dissimilarity matrices. The differences in assemblage compositions between disturbance categories were assessed using PERMANOVA with 999 permutations. The Benjamini–Hochberg false discovery rate correction was used for pairwise comparisons. Multivariate dispersion was tested for homogeneity with PERMDISP. Indicator genera related to disturbance categories were determined with the help of indicator species analysis. The relationships between nematode community composition and environment were analysed by canonical correspondence analysis (CCA), and functional trait relationships were analysed by redundancy analysis (RDA).

The relationships between environmental variables and nematode community composition (or functional trait matrices) were analyzed with the help of canonical correspondence analysis (CCA) and redundancy analysis (RDA). Environmental factors were normalised by $\ln(x+1)$, which is expected to normalise the distributions and test multicollinearity as variance inflation factors (VIF); variables with $VIF > 10$ were eliminated sequentially, and one variable with higher ecological interpretability was retained. For ordinal logistic regression, the ordiR2step function was used to select the minimum adequate model of environmental predictors that explained significant variation (permutation test, 999 permutations, adjusted R² criterion, significance threshold $\alpha = 0.05$). Individual bioindicator indices vs. environmental variables: Pairwise Spearman rank correlations were calculated using the cor.test function with Holm multiple comparisons correction.

3. Results

3.1 Environmental Characterization of Study Sites

There were significant differences among disturbance categories (HD, ID, and LD) in the environmental variables (Table 2). The levels of the metals were significantly different between the sites ($p < 0.001$). The mean concentrations of Pb in the samples were $187 \pm 42 \mu\text{g/g}$ (dw) at HD sites, $67 \pm 15 \mu\text{g/g}$ (dw) at ID sites, and $8 \pm 3 \mu\text{g/g}$ (dw) at LD sites. Cu concentrations were 134 ± 28 , 48 ± 11 , and $6 \pm 2 \mu\text{g/g}$ dw at HD, ID, and LD sites, respectively. Zn concentrations were highest at HD sites ($412 \pm 89 \mu\text{g/g}$ dw), followed by ID sites ($143 \pm 32 \mu\text{g/g}$ dw) and LD sites ($21 \pm 7 \mu\text{g/g}$ dw). Cadmium concentrations were comparable, at 3.8 ± 0.9 , 1.2 ± 0.4 , and $0.2 \pm 0.1 \mu\text{g/g}$ dw at the HD, ID, and LD sites, respectively.

The organic content of the sediments also varied widely by disturbance category. Total organic carbon (TOC) was highest at HD sites ($5.8 \pm 1.2\%$), followed by ID ($2.9 \pm 0.6\%$) and LD sites ($1.4 \pm 0.3\%$) ($p < 0.001$). Total nitrogen

concentrations were $0.71 \pm 0.14\%$, $0.39 \pm 0.08\%$, and $0.19 \pm 0.04\%$ at HD, ID, and LD sites, respectively. The C: N ratio increased from HD sites (8.2 ± 1.1) to ID sites (9.8 ± 1.4) and LD sites (14.7 ± 2.3).

The difference was also large between the categories for sediment grain size parameters. Median grain size was lowest at HD sites ($48 \pm 12 \mu\text{m}$), intermediate at ID sites ($132 \pm 31 \mu\text{m}$), and highest at LD sites ($287 \pm 54 \mu\text{m}$). Sorting coefficient values were 2.8 ± 0.4 , 1.9 ± 0.3 , and 1.2 ± 0.2 at HD, ID, and LD sites, respectively ($p < 0.001$). The number of nematodes per unit area was significantly different between disturbance categories ($p < 0.001$). The highest density was recorded at LD sites ($2934 \pm 512 \text{ ind}/10 \text{ cm}^2$), followed by ID sites ($1847 \pm 387 \text{ ind}/10 \text{ cm}^2$), whereas HD sites showed the lowest density ($892 \pm 214 \text{ ind}/10 \text{ cm}^2$).

Table 2: Environmental Parameters Across Disturbance Categories

Parameter	High Disturbances (HD)	Intermediate Disturbances (ID)	Low Disturbances (LD)	p-value
Pb ($\mu\text{g/g dw}$)	$187 \pm 42^{**}$	$67 \pm 15^*$	8 ± 3	< 0.001
Cu ($\mu\text{g/g dw}$)	$134 \pm 28^{**}$	$48 \pm 11^*$	6 ± 2	< 0.001
Zn ($\mu\text{g/g dw}$)	$412 \pm 89^{**}$	$143 \pm 32^*$	21 ± 7	< 0.001
Cd ($\mu\text{g/g dw}$)	$3.8 \pm 0.9^{**}$	$1.2 \pm 0.4^*$	0.2 ± 0.1	< 0.001
TOC (%)	$5.8 \pm 1.2^{**}$	$2.9 \pm 0.6^*$	1.4 ± 0.3	< 0.001
Total nitrogen (%)	$0.71 \pm 0.14^{**}$	0.39 ± 0.08	0.19 ± 0.04	< 0.001
C: N ratio	8.2 ± 1.1	9.8 ± 1.4	$14.7 \pm 2.3^{**}$	< 0.001
Median grain size	48 ± 12	132 ± 31	$287 \pm 54^{**}$	
Sorting coefficient (ϕ)	2.8 ± 0.4	1.9 ± 0.3	$1.2 \pm 0.2^{**}$	< 0.001
Total nematode density ($\text{ind}/10\text{cm}^2$)	892 ± 214	$1,847 \pm 387$	$2,934 \pm 512^{**}$	< 0.001

3.2 Total Nematode Abundance and Assemblage Composition

We identified 87 nematode genera from 19 families across all stations. Chromadoridae, Cyatholaimidae, Xyalidae and Comesomatidae were the most species-rich families (14 genera, 11 genera, 9 genera and 8 genera, respectively). The nematode assemblage composition was different between the disturbance categories. Sabatieria (24%), Dorylaimopsis (16) and Terschellingia (12) were more abundant on the HD sites. The genera Chromadora, Prochromadora, Hypodontolaimus, Oncholaimus and Eurystomina were overrepresented in LD sites.

3.3 Taxonomic Diversity Indices

All taxonomic diversity indices that were measured were significantly different between the disturbance categories (Table 3). The number of genera (genus richness) was highest at the LD sites (44 ± 7) and significantly lower at the ID sites (31 ± 6 , $p < 0.001$) and the HD sites (18 ± 4 , $p < 0.001$). Shannon diversity values were highest at LD sites (3.41 ± 0.22), intermediate at ID sites (2.74 ± 0.28), and lowest at HD sites (1.82 ± 0.31) ($p < 0.001$). Pielou's evenness decreased from 0.88 ± 0.05 at LD sites to 0.79 ± 0.07 at ID sites and 0.61 ± 0.09 at HD sites. Margalef's richness index showed a similar pattern, with values of 8.27 ± 1.14 , 5.84 ± 0.93 , and 3.12 ± 0.71 at LD, ID, and HD sites, respectively.

PERMANOVA analysis indicated that there were significant differences in community composition among the disturbance categories (pseudo-F = 24.7, $R^2 = 0.67$, $p < 0.001$). The pairwise comparison indicated that HD, ID, and LD assemblages were significantly different (FDR-adjusted $p = 0.01$). The results of PERMDISP analysis indicated larger dispersion of the HD samples (distance to centroid = 0.38) than the LD samples (0.22).

Table 3: Nematode Diversity and Ecological Indices

Diversity Index	High Disturbances (HD)	Intermediate Disturbances (ID)	Low Disturbances (LD)	p-value
Genus Richness (S)	18 ± 4	31 ± 6	44 ± 7	< 0.001
Shannon Diversity (H' , $\log_2$)	1.82 ± 0.31	2.74 ± 0.28	3.41 ± 0.22	< 0.001
Pielou's Evenness (J')	0.61 ± 0.09	0.79 ± 0.07	0.88 ± 0.05	< 0.001
Margalef's Index (d)	3.12 ± 0.71	5.84 ± 0.93	8.27 ± 1.14	< 0.001
Maturity Index (MI)	1.74 ± 0.18	2.53 ± 0.24	3.42 ± 0.31	< 0.001
Nematode Channel Ratio (NCR)	0.87 ± 0.06	0.68 ± 0.08	0.44 ± 0.09	< 0.001
NemaSPEAR[%]	72.4 ± 8.3	38.7 ± 6.1	12.3 ± 4.2	< 0.001

3.4 Trophic Guild and c-p Class Composition

There was significant variation in trophic food web structure among disturbance classes. Bacterivores (1B) were relatively abundant at the HD sites ($68 \pm 9\%$) compared to the LD sites ($38 \pm 6\%$). The proportion of selective

deposit feeders (1A) was greater at ID sites. There was a reduction in the percentage of epistrate feeders at the HD sites compared to the LD sites ($27 \pm 5\%$ versus $11 \pm 4\%$). The abundance of the predator–omnivores (2B) was significantly lower along the disturbance gradient, from $18 \pm 4\%$ at the LD sites to $4 \pm 2\%$ at the HD sites (Kruskal–Wallis $H = 13.8$, $p < 0.001$).

There were also differences in the c–p composition across disturbance categories. The c–p 1 and c–p 2 taxa at HD sites accounted for $74 \pm 8\%$ of the total abundance, while the c–p 4 and c–p 5 taxa accounted for $6 \pm 3\%$. At LD sites, c–p 4–5 taxa represented $31 \pm 6\%$, while c–p 1–2 taxa represented $28 \pm 5\%$. The Maturity Index decreased significantly from LD (3.42 ± 0.31) to HD sites (1.74 ± 0.18) ($p < 0.001$). MI showed significant negative correlations with Pb ($r = -0.84$), Cu ($r = -0.79$), and Zn ($r = -0.76$) ($p < 0.001$). NCR values increased from LD (0.44 ± 0.09) to HD sites (0.87 ± 0.06) and showed a significant correlation with TOC ($r = 0.81$, $p < 0.001$).

3.5 Functional Trait Diversity

Diversity indices for functionality varied between disturbance types. Functional richness within HD plots constituted 23% of functional trait space in LD plots. Richness in terms of genera in HD plots comprised 41% of the richness of genera in LD plots.

The Rao quadratic entropy index value reduced from 0.74 ± 0.09 in LD plots to 0.31 ± 0.06 in HD plots, confirming convergence of the progressive traits at the bacterivore, small-bodied, c–p 1–2 region of the functional space, defining a stress-tolerant assemblage of the world (19). Functional evenness values in LD, ID, and HD plots were 0.41 ± 0.06 , 0.54 ± 0.08 , and 0.38 ± 0.07 , respectively. The functional divergence value decreased from 0.71 ± 0.06 in LD plots to 0.43 ± 0.08 in HD plots.

Table 4: Environmental Drivers of Community Structure

Environmental Variable	Axis 1 Loading	Axis 2 Loading	Variance Explained (%)	VIF	p-value
Pb concentration	-0.84	0.22	18.2	3.1	< 0.001
TOC content	-0.79	0.31	14.7	4.2	< 0.001
Cu concentration	-0.76	0.18	11.4	3.8	< 0.001
Median grain size	0.71	-0.29	8.9	5.1	< 0.001
Zn concentration	-0.68	0.24	7.4	4.9	0.001
Dissolved oxygen	0.63	0.41	5.8	2.7	0.002
Sediment nitrogen	-0.58	0.19	4.7	3.4	0.008
Residual (unexplained)	-	-	41.7	-	-

3.6 Canonical Ordination and Environmental Drivers

The Bray–Curtis nMDS ordination was a two-dimensional solution with stress of 0.12. Disturbances of HD, ID, and LD resulted in distinct separation of nematode assemblages in ordination space.

The forward selection CCA model found seven significant environmental factors that accounted for 58.3% of the total constrained variation in nematode community structure. Pb concentration explained 18.2% of variation, TOC (14.7%), Cu (11.4%), median grain size (8.9%), Zn (7.4%), DO (5.8%), and sediment N (4.7%).

The first two CCA axes explained 41.6% of constrained inertia. *Sabatieria* and *Dorylaimopsis* had greater association with high concentrations of Pb and TOC, while *Oncholaimus* and *Chromadora* had the opposite ordination.

4. Discussion

The observed changes in nematode assemblages along the disturbance gradient were consistent with established ecological responses of benthic communities to pollution stress. Highly disturbed sites had lower levels of richness, diversity, and evenness and higher levels of opportunistic taxa, suggesting community simplification and loss of disturbance-sensitive taxa (20). Similar changes have been described for marine nematode communities subject to organic enrichment and contaminant stress, where organic enrichment leads to assemblage simplification and increased dominance of opportunistic nematodes (21, 22). This is further corroborated by the sharp drop in the number of predator–omnivore species at very disturbed sites. Increased c–p values are often correlated with predaceous nematodes and omnivorous nematodes, which are typical components of relatively stable environments; thus their decrease may suggest breakage of higher trophic members of meiofaunal food chains (23). These changes could have ecological significance as the demise of high trophic level organisms may affect trophic dynamics, energy flow and benthic ecosystem function. Thus, the results indicate that in addition to changes in taxonomic richness, there is the possibility of complementary information from trophic composition. Total nematode density was lower at highly disturbed sites than at intermediate-disturbance sites, suggesting that abundance alone may not consistently reflect pollution intensity. A moderate level of organic enrichment can lead to higher abundances of opportunistic groups and higher food availability, while more extreme contamination can lead to lower density overall with time. Therefore, community composition, diversity indices, life-history strategies and functional traits can be a more useful way to gauge ecological condition than abundance over a gradient of disturbance heterogeneity.

A particularly important finding was the stronger response of functional diversity to increasing disturbance. A major contribution of this study is the demonstration that functional trait-based approaches provide enhanced

sensitivity for detecting ecological changes compared with taxonomic metrics alone (24). While genus richness decreased along the gradient, the changes in functional richness and Rao's quadratic entropy were even greater, suggesting that ecological strategies and functional trait space were disproportionately affected (25). Functional trait space contraction was linked to assemblages with tolerant, small-bodied, low c–p and bacterivorous taxa at highly disturbed sites. This trend indicates that environmental filtering exists, with pollution and related environmental characteristics promoting a more restricted set of ecological strategies and a decrease in sensitive life-history and trophic traits (26). The functional convergence could have implications for the resilience of the ecosystem as the loss of functionally distinct taxa decreases the ecosystem's ability to respond to increased environmental stress.

These findings support the value of combining functional and taxonomic approaches in nematode-based biomonitoring. Taxonomic indices describe shifts in richness, diversity, and community composition, while functional indices describe shifts in ecosystem strategies, resource use, environmental tolerance, and potential shifts in ecosystem function. Functional approaches may thus detect aspects of ecological degradation not quantified by conventional taxonomic measures. However, the present spatial design does not assert that functional metrics always lead to earlier detection of disturbance over time. For such early-warning capability to be demonstrated, one would need to take repeated temporal observations. However, increased functional responses were seen across the current disturbance gradient, suggesting that functional metrics may also offer additional and perhaps sensitive data to support coastal ecological assessment.

Heavy-metal contamination and organic enrichment were also shown to be closely related to changes in nematode community structure in multivariate analysis. The strongest individual association was for lead, followed by total organic carbon and copper, consistent with previous findings that gradients of contaminants can significantly restructure benthic meiofaunal communities (27). However, these relationships do not imply specific causal effects of individual contaminants. Coastal areas are often impacted by several pressures such as increased levels of metals, organic pollution, changes in sediment characteristics, and low oxygen levels, all of which frequently occur together in industrial or urbanized environments. The patterns of biological response observed are thus more likely to be reactions to interacting environmental gradients, and not to a single pollutant.

Environmental variables accounted for a significant amount of community variation, but there was still some unexplained variation. This residual variation may be due to unmeasured environmental factors, interactions among organisms, spatial variation, or the random dynamics of communities (28). Sediment characteristics are particularly important because fine depositional environments may form accumulations of organic matter and contaminants, which may be correlated with habitat structure and pollution intensity (29). Therefore, caution should be used when interpreting the present associations when defining community changes due to individual contaminants. Future studies that consider wider habitat factors, repeat samples across seasons and years, and/or have longer time series may be better able to separate contaminant effects from sedimentary and natural environmental variability (30).

From a biomonitoring perspective, the results indicate that an integrated nematode-based assessment can provide multiple and complementary indicators of ecological condition. Monitoring could involve using existing indices and metrics of nematode ecology, including the Maturity Index, trophic composition, conventional diversity indices, and other validated indices and metrics, in combination with more targeted functional trait analyses at sites with evidence of ecological degradation (31). Further multivariate analyses and physicochemical measurements might then be applied to the study of the environmental pressures that were associated with observed biological changes (32, 36). A tiered design might allow for both routine ecological screening and diagnostic assessment to be performed, but would need to be assessed through further testing in additional coastal systems for cost-effectiveness and transferability. Nematodes can be useful for benthic monitoring, as a relatively small sample of sediment can yield a lot of ecological information, but widespread use of nematodes requires a standardized sampling protocol, taxonomic methodologies, traits database, and region-specific calibration (33, 34, 35).

More broadly, recent sustainability-focused studies emphasize the use of digital monitoring, cleaner energy systems, carbon-reduction technologies, reusable catalytic materials, and environmental-performance frameworks to support sustainable resource management.^{35–41}

Several limitations should therefore be recognized. The study represents a spatial assessment conducted within a single biogeographic region and doesn't account for seasonal or interannual variability. Genus-level identification, although commonly used in nematode biomonitoring, may also obscure species-specific differences in contaminant sensitivity and ecological function. The species identification achieved by morphological tools might be enhanced by molecular methods, e.g., DNA metabarcoding, but only if sufficiently comprehensive reference databases are available. Overall, the study demonstrates that including taxonomic diversity, trophozoology, c–p life-history strategies, functional traits, and multivariate environmental relationships together gives a more comprehensive characterization of nematode responses to coastal disturbance.

5. Conclusion

This study provides empirical evidence that free-living marine nematodes are sensitive and reliable indicators of changes in the coastal ecosystem along anthropogenic disturbance gradients. The combination of taxonomic diversity, trophic guild composition, colonizer–persister classification, and functional trait metrics enabled a broad picture of community response to pollution. Under high contamination levels, nematode assemblages were reduced and dominated by tolerant bacterivorous and colonizer species compared to assemblages found at reference sites that contained diverse, functionally complex communities.

Functional trait metrics were more sensitive to disturbance than taxonomic indicators, with the functional trait

space being reduced to 23% of reference conditions while genus richness was reduced by 41%. These results show the need for functional approaches to be integrated into marine biomonitoring systems. Multivariate analyses revealed that heavy metals and organic enrichment were important factors in nematode community restructuring, highlighting the importance of integrated assessments for understanding complex coastal stressors.

A two-tier monitoring framework combining routine ecological indices (MI, NCR, and NemaSPEAR) with targeted functional trait analysis and multivariate assessment of impacted sites provides a practical approach for coastal ecosystem monitoring. The nematode-based approach proposed in this framework is sensitive and cost-effective, and is a useful tool for assisting pollution assessment and sustainable management of coastal marine ecosystems.

Use of AI tools declaration

The authors declare they have not used Artificial Intelligence (AI) tools in the creation of this article.

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Conflict of interest

The authors declare that there are no conflicts of interest regarding the publication of this paper.

Ethical Statement

This study did not involve human participants, vertebrate animals, or protected species, and therefore ethical approval was not required.

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