

Impact of a dam construction on the intertidal environment and free-living nematodes in the Ba Lai, Mekong Estuaries, Vietnam

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Abstract The impact of high siltation and accumulation of organic and waste material in the intertidal of the dammed Ba Lai River in Vietnam as part of the Mekong estuarine system was investigated by means

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Marine Biology Research Group, Biology Department, Ghent University, Krijgslaan 281, S8, B-9000 Ghent, of marine free-living nematodes. Nutrients content (nitrate, ammonium, total phosphorus, total nitrogen), total suspended solids, total organic carbon, coliform, bacteria *E. coli*, pH, dissolved oxygen, total dissolved solids, methane and hydrogen sulfide concentration, and the nematode communities were characterized in sediment at selected stations along the river above and below the dam. Our results found elevated

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Far Eastern Branch of Russian Academy of Sciences, Federal Scientific Center of the East Asia Terrestrial Biodiversity, Vladivostok, Russia methane concentrations at the upstream side of the dam while hydrogen sulfide concentrations found to be highest in the downstream side of the dam. Furthermore, methane and hydrogen sulfide concentrations were correlated to nematode community characteristics such as trophic composition densities and genera composition. There was a clear difference between the communities above and below the dam. The discontinuous nematode community distribution indicated that the Ba Lai River is impacted by dam construction. Potentially the high deposition and eutrophication could turn the area into a methane-rich area related to predicted impact on nematodes.

Keywords Ba Lai River · Ben Tre · Climate change · Environmental impact · Irrigation · Nematode · Reservoir

Introduction

The Mekong Delta in Vietnam plays a major role as a vast "food basket" for the country and for the global food market, exporting mainly rice, fruits, and seafood worldwide (Dung et al., 2019; Piesse, 2019). However, this delta is facing problems of serious salt intrusion and drought due to reduced freshwater discharge from upstream areas (Park et al., 2021). In order to solve these problems, the construction of dams along the coastal region and estuarine areas has been a priority for the Vietnamese government. In 2002, the first largest dam of the North Ben Tre Irrigation Project in the Mekong delta was built across the mainstream of the Ba Lai estuary (Hoang et al., 2009).

The Ba Lai sluice gate has brought some benefits, such as flood control, freshwater supply, and irrigation, hence favoring the economic development of local communities (Ngo et al., 2017a, 2017b). Its operation however has also resulted in long-term negative effects on the environmental quality. The erection of the Ba Lai dam is considered one of the reasons for the differences between its intertidal and subtidal nematode assemblages and those of the remaining Mekong estuaries (Ngo et al., 2016) (Nguyen et al., 2020) both in terms of densities and species and genera diversity. Furthermore, the dam's construction had an effect on the biochemical components of the estuary, as observed by the increase in total suspended solids, heavy metal concentrations (Hg and Pb), and significant oxygen depletion (Nguyen et al., 2020). The dam's presence may contribute to driving the Ba Lai's ecosystem to a tipping point.

Indeed, soon after the dam began to operate, the quantity of microalgae, the concentration of phosphates and nitrates, and the suspended sediments were higher upstream of the dam and lower near the river mouth area (Veettil et al., 2019). Furthermore, turbidity levels increased since the dam construction due to alluvial silty deposition (Veettil & Ngo, 2018). Also, sediments discharged from upstream could be trapped above the dam's sluice forming a stagnant area (Lin, 2011). Combined with continuous siltation input, this has led to a reduction of the water capacity and subsequently the depletion of oxygen in the bottom layers turning the river's function into a shallow "lake-river" and supporting anaerobic processes that produce a large amount of greenhouse gases such as methane with a warming potential far greater than that of carbon dioxide (Ramaswamy et al., 2001), but also hydrogen sulfide, which is toxic for most eukaryotic life forms (Liikanen et al., 2002).

In aquatic ecosystems, hydrogen sulfide has a major influence on benthic fauna (Bagarinao, 1993), including the dominant group within the meiofauna size class, the nematodes. An early experiment by Rodriguez-Kabana et al. (1965), for example, reported that a decline in the total number of nematodes in flooded rice fields was associated with an increase of hydrogen sulfide. Moreover, Armenteros et al. (2010) found that chemical stressors such as hydrogen sulfide affected the structure and distribution of nematode communities in the Cienfuegos Gulf sediments, Cuba. In contrast to the large amount of studies on the effect of hydrogen sulfide, investigations on the effects of methane in sediment on freeliving nematodes are more limited. However, the presence of methane is often associated with higher sulfide concentrations and therefore its effect is difficult to disentangle. Jensen (1995) studied the effect of methane emissions in soft sediments and discovered that adults from the dominant nematode species Theristus (Penzancia) anoxybioticus were mainly found on the surface of sediments, while juveniles were distributed deeper in sediment. Remarkable is the presence of the mouthless chemosynthetic nematode species Astomonema southwardorum (family Siphanolaimidae) in a large pockmark with active methane seepage in the North Sea at 153–167 m depth (Austen et al., 1993; Dando et al., 1991). Also, Sapir et al. (2014) did a noteworthy observation that methane seeps supported complex ecosystems involving interkingdom interactions between bacteria, nematodes, and parasitic fungi.

In this study, we used nematode communities to investigate the potential impact of a dam on the intertidal benthic environment. It was hypothesized that like subtidal environments of the same river (Nguyen et al., 2020), the intertidal would also be impacted by increased deposition of silt and associated organic waste material resulting in anoxic sediments, the emission of methane, and the production of sulfides which may have effects on the associated free-living nematodes. We used nematodes as an indicator for the ecological effects of the changing biochemical conditions in intertidal sediments. Specifically, we hypothesized that (1) the presence of the dam resulted in different intertidal environments based on the investigated biotic and abiotic variables such as pH, dissolved oxygen(DO), salinity, total dissolved solids (TDS), total suspended solids (TSS), total organic carbon (TOC), nitrate (NO3⁻), ammonium (NH4⁺), total phosphorus (TP), total nitrate (TN), bacteria *E. coli*, total coliform concentrations (coliform), methane (CH₄), and hydrogen sulfide (H₂S) concentrations and (2) intertidal nematode communities differed in relation to changing environmental conditions derived from the presence of the dam.

Methodology

Sampling location

This study was conducted in both wet and dry season in the Ba Lai River, Ben Tre province, located at $106^{\circ}20'0''E-106^{\circ}40'00'' E$ and $10^{\circ}0'0''-10^{\circ}20'0''$ N, within the Mekong Delta in southern Vietnam (Fig. 1). The southern part of Vietnam has a tropical climate with two seasons: wet (May to November) and dry (December to April). Even though the average monthly air temperatures do not fluctuate much in the south, the weather in this region still differs between the two seasons due to the variations in precipitation (Ngo et al., 2017b). From the sea toward the inland, samples were collected in both Ba Lai



Fig. 1 Sampling locations in the Ba Lai River, Vietnam, for both seasons

riverbanks at 8 river transects coded from B1 to B8, within 2 sampling stations per transect for total of 16 intertidal stations (Fig. 1).

Environmental variables

At each station, sediment samples were taken for chemical and physical characteristics, such as nutrient contents (NO_3^- , NH_4^+ , TP, TN), TSS, TOC, coliform, *E. coli*, methane, and hydrogen sulfide. Water salinity, pH, DO, and TDS were measured on-site by a WQC-22A Water Quality Checker (TOA, Japan).

For methane (CH_4) analysis, about 10 mL of the 2-cm top of undisturbed sediment was placed into a 40-mL thread bottle containing 5 mL of 0.1 N NaOH to terminate further bacterial activity. The vial was immediately capped with a Teflon-lined silicone septum and kept cool with dry ice. Once returned to the laboratory, samples were stored at -18 °C until analysis. Methane in sediment samples was analyzed following the method of Leloup et al. (2007). In summary, methane in the headspace was determined by gas chromatography equipped with an Alumina Sulfate PLOT (30 m \times 0.53 mm \times 10 µm, Supelco, USA), a split/splitless injector, and a flame ionization detector. The injector was operated at splitless mode, and temperature of 35 °C. The linear velocity of nitrogen as carrier gas was set at 90 cm/s and the detector was operated at 250 °C, air flow rate of 400 mL/min, and hydrogen flow rate of 40 mL/min. Quantification of methane was carried out using a calibration curve made by injecting various volumes of standard methane gas 15 ppmv (Agilent) with five replicated injections of every sample and standard (Leloup et al., 2007).

For the sulfide (H_2S) analysis, the top 2 cm of undisturbed sediments were carefully placed in a 50-mL PE conical bottom tube with thread cap (ISOLAB). The sediment samples were kept on dry ice during sampling and transportation and stored at – 18 °C prior to the laboratory analysis. Total free H₂S concentrations were measured following the method of Brown et al. (2011). Briefly, the sediment sample stored in the 50-mL plastic tube was mildly defrosted at 4 °C overnight, and then centrifuged at 3000 rpm for 5 min. The supernatant was decanted and the sediment was mixed well with a stainlesssteel spatula. A 10-mL portion of the homogenized sediment was taken and placed into another 50-mL PE tube containing 10 mL of sulfide antioxidant buffer. The mixture was vortexed, and the concentration of sulfide was quickly measured on a potentiometer (Mettler Toledo) equipped with a sulfide ion selective electrode. The concentrations of sulfide in the samples were calculated based on a sulfide calibration curve prepared in the same manner samples (Brown et al., 2011).

Escherichia coli (*E. coli*) and coliform bacteria concentrations in sediments were identified based on the standard most probable numbers (MPN) method (Olson, 1978). The MPN technique estimates microbial population sizes by dilution and incubation of replicated cultures across several serial dilution steps. For each sample, 300 g of surface sediments were collected, stored in sterile plastic bags, and preserved cool. The samples were transported to laboratory and analyzed within 36 h after being sampled.

For the analysis of nutrient concentrations in the sediments, sampling was conducted using a 6-cm inner diameter polycarbonate core pushed into the sediment up to 10-cm deep. The core-housing sediment was extruded, sliced every 1-cm segment and collected to 5-cm depth, and the remaining 5-cm segment for the 5- to 10-cm depth. The portions were preserved at 4 °C during the sampling trip. All sediment and porewater samples were kept frozen at -20 °C until analysis. In the laboratory, porewater was extruded under N2 atmosphere and passed through Whatman GF/C filters. The TOC and TN contents in sediment samples were analyzed by elemental analysis method (Element Analyzer Flash 2000, Thermo Scientific). In the TP analysis, sediment samples were ignited to decompose organic ingredients followed by boiling in 1 N HCl (Andersen, 1976). The TP content as orthophosphate was measured by photometric method using a segmented flow analyzer (SAN++, SKALAR). The NO_3^- and NH_4^+ in porewater samples were also analyzed by on segmented flow analyzer.

Sampling and analyses of nematodes specimens

For analysis of the nematode, communities' triplicate samples were collected at the same 8 intertidal stations at both riverbanks by means of 10 cm^2 transparent plastic cores that were pushed into the sediment to at least 10 cm depth. Samples were preserved in 7% neutralized formaldehyde (pre-heated to 60–70 °C)

before extracting from the sediment by washing the sample over a 1-mm sieve and keeping the fraction retained on a 38-µm sieve. The extracted samples were then separated and collected by flotation technique using Ludox-TM50 (specific gravity of 1.18) and stained with 1% solution of Rose Bengal (Vincx, 1996). All nematode individuals in each sample were counted under a stereomicroscope. About 200 nematodes per sample (if the sample consists of less than 200, all nematodes in that sample) were randomly picked out and processed for making permanent slides for taxonomic identification (De Grisse, 1969). The pictorial key on free-living marine nematodes part III (Warwick et al., 1998), the identification manual for freshwater nematode genera (Zullini, 2021), the free-living nematodes in Vietnam (Nguyen, 2007), the handbook on freshwater nematodes: ecology and taxonomy (Eyualem-Abbe et al., 2006), and the NEMYS database (Bezerra et al., 2020) were referred for the identification. The nematodes were classified into four feeding categories, based on the structure of the buccal cavity according to Wieser (1953): (1A) selective deposit-feeders, (1B) non-selective depositfeeders, (2A) epistratum feeders, and (2B) predators or omnivores (Jensen, 1987).

Data analysis

Nematode communities

Nematode data were presented by the averages and standard deviations of triplicate samples per station per transect (16 station in 8 river transect). The indices of nematode communities, such as density (N), genera richness (S), Shannon–Wiener diversity-H' (Shannon & Weaver, 1949), Hill indices N1 and N2 (Hill, 1973), Hurlbert's index–ES (50) (Hurlbert, 1971), feeding type (1A, 1B, 2A, 2B) (Jensen, 1987; Wieser, 1953), trophic diversity index–TD (Heip et al., 1988), and the Maturity index–MI (Bongers, 1990; Bongers et al., 1991), were tested for significant differences.

The ANOVA design incorporated the factors seasons, stations, and dam sides. The first factor design ("station") compared differences between the stations, consisting of 8 groups. The second design considered the factor "season" including the dry season and the wet season. The third design took into account the presence of the "dam" and was therefore composed of two levels referred as downstream and upstream of the dam.

The Shapiro–Wilk test and Levene's test were used to check for normal distribution and to evaluate the homogeneity of variances (p > 0.05). The data was $\log (x+1)$ transformed if assumptions were not met. Firstly, one-way ANOVA (analysis of variance) with factor "station" was tested to see if there are significant different between station (Table 2). Since the stations within each dam side were not significantly different, which is able to nest in dam (to prevent mixing of the stations from different dam sides) and to keep our focus interest in the effect of 2 factors "dam" and "season," a two-factor ANOVA ("season" and "dam") performed in RStudio (RTeam, 2020) in order to identify significant differences in each univariate variable. When significant differences were found (p < 0.05), a post hoc test (Tukey HSD) was applied for pairwise comparisons between stations. A three-factor PER-MANOVA was replaced if the assumptions were not fulfilled. Because of multiple univariate tests, the multiple obtained p values were corrected with the Benjamini and Hochberg (1995) correction method to control overall experimental type I error and type II error rate.

The structure of the nematode communities was explored by using multivariate analyses. Significant differences between groups in genera composition were analyzed with PERMANOVA based on log(x+1) transformed data. After the PERMANOVA routines, pairwise pseudo t tests were executed to identify which pairs of transects were significantly different from each other. Subsequently, PERMDISP routines were performed to test for homogeneity of multivariate dispersions, indicating location differences through equally dispersed distance to centroids (p > 0.05). A cluster analysis (using Bray Curtis similarity) was performed with all the replicates in order to explore group patterns. The MDS analysis (multidimensional scaling analysis) was used to produce 2D graphs basing on the Bray-Curtis similarity index in order to visualize the pattern. The SIMPER analysis (SIMilarity PERcentages) was conducted to assess similarities and dissimilarities across taxa. This analysis examined the contribution of each taxon to the average Bray-Curtis dissimilarity between groups of samples and also determined their contribution to the similarity within each group (Clarke & Warwick, 2001).

Analyses of environmental variables

Data of methane, hydrogen sulfide, and relevant environmental variables were analyzed by a Draftsman plot to check for skewness and homogeneity. A principal component analysis (PCA) was performed to identify the present environmental gradient in the pattern.

Significant differences of environmental variables between stations, dam sides, and seasons were tested with a three-factor PERMANOVA (3 factors: "season," "station," and "dam"). The multivariate environmental data (H₂S and CH₄, pH, DO, salinity, TSS, TDS, *E. coli*, coliform, NO_3^- , NH_4^+ , TP, and TN) was first normalized and PERMANOVA on Euclidean distances.

A DistLM (distance-based linear model) analysis was conducted on the environmental variables with correlations lower than 0.9, in order to identify environmental factors associated with the variability in the structure of the nematode communities. The DistLM model was performed with the software PRIMER v.6 add on PERMANOVA using a stepwise selection procedure, adjusted R² as selection criteria and visualized by the dbRDA (distance-based redundancy analysis) plots (Anderson et al., 2008).

The software STATISTICA 7.0 was also used to perform non-parametric Spearman rank correlation coefficients (p < 0.05) to identify the correlations between methane, hydrogen sulfide, and relevant sed-iment environmental variables and the univariate data of nematode communities.

Results

Environmental characteristics of the intertidal area of the Ba Lai River

All data on environmental variables are shown in Table 1. Methane was mainly found in the upstream portion of the study, especially at the station B4 with values up to $8422.98 \pm 14,215.95$ (mg/kg) in the dry season, but also at station B8 with concentrations up to $12858.3 \pm 13,639.6$ mg/ kg in the wet season. Hydrogen sulfide exhibited a high concentration at station B3 in the dry season with $316.15 \pm 328.26 \mu$ M. For the other stations, hydrogen sulfide concentrations were ranging from 4.2 ± 3.4 to $214.1 \pm 157.2 \mu$ M (Fig. 2). However, there were no statistically significant differences among stations, dam sides, seasons, and their interaction.

The concentrations of organic material and nutrients in the sediment including TOC, NH_4^+ , NO_3^- , TN, and TP showed in general higher values between the stations B3 and B6, especially in dry season (Table 1). Significant differences were found between stations for TOC, NH_4^+ , TN, and TP. The concentration of NO_3^- was found significantly different based on the interaction between station and season.

Bacteria E. coli were most numerous at station B5 with concentrations of 1110 ± 1485.6 (CFU/g) in the wet season. In the other upstream stations, high E. coli were also found in the wet season (from 246.7 ± 376.5 to 668.3 ± 673.2 CFU/g). Also, coliform bacteria showed the highest values at the station B5 in the wet season with $49,033.3 \pm 47,980.2$ (CFU/g). The coliform concentrations in the remaining stations were also high especially at B7 $(11,400 \pm 9824.5 \text{ CFU/g})$, $(13,233.3 \pm 18,852.1)$ **B**8 CFU/g), and B3 $(3866.7 \pm 2706.2 \text{ CFU/g})$. No significant difference was found in the case of bacteria E. coli. However, coliform bacteria numbers were found to be significantly different between stations (p=0.001). Post hoc comparison identified differences between the 3 downstream stations (i.e., B1, B2, and B3), and between the upstream station B5 with the two adjacent stations B4 and B6.

Variables measured in the water column such as pH, dissolved oxygen (DO), and salinity are presented in Fig. 3. The pH was slightly alkaline; the lowest value was 7.2 ± 0.54 at station B6 in the wet season and the highest value was 8.5 ± 0.14 at B3 in the dry season. No significant difference was found for pH between stations, dam side, and seasons. Salinity ranged from 0.058 ± 0.002 % at station B8 in the wet season to 8.27 ± 0.55 % at the mouth station B1 in the dry season. Salinity was obviously higher in stations located downstream of the dam (i.e., from B1 to B3) where it ranged between 1.2 ± 0.2 and 8.3 ± 0.6 % compared to the salinity in the upstream stations (i.e., B 4 to B8). DO was quite low in both dry and the wet season for all stations ranging from 3.7 ± 0.1 mg/L in the dry season to 6.6 ± 0.2 mg/L

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Table 1 Table c	of environmental varia	ables of Ba Lai Ri	ver in the wet (W) and	the dry season (D)				
Environmental B1			B2		B3		B4	
Variautes W		0	M		M	0	M	D
Hq	7.7 ± 0.1	8.4±0	7.6±0	7.9 ± 0.1	7.3 ± 0.1	8.5 ± 0.1	7.6±0.1	7.9 ± 0.1
DO (mg/l)	4.4 ± 0.2	4.6 ± 0.5	4.4 ± 0.1	4.1 ± 0.4	4.1 ± 0.1	3.8 ± 0.6	3.9 ± 0.1	4.1 ± 0.2
TSS (mg/l)	99.5 ± 83.6	78.7 ± 14.9	170.7 ± 6.5	58.3 ± 25.7	237 ± 61.6	35.3 ± 12.9	72.7 ± 0.6	74.9 ± 11.7
TDS (mg/l)	2270 ± 294.6	8046.7 ± 506.4	968 ± 39.3	6603.3 ± 397.2	808 ± 78.6	3226.7 ± 222.3	322 ± 6.6	512.3 ± 17.2
Sal (%0)	8 ± 0.6	8.3 ± 0.6	2.1 ± 0.1	6.7 ± 0.4	1.2 ± 0.2	3.3 ± 0.2	0.4 ± 0	0.5 ± 0
NH4 (mg/kg)	15.4 ± 3.1	40.8 ± 16.2	24.9 ± 9.9	154.3 ± 39.5	10.4 ± 1.9	106 ± 26.5	2.1 ± 0.7	27.9 ± 9.8
NO ₃ (mg/kg)	11.2 ± 5.3	25.1 ± 16.8	25.6 ± 13.6	16.4 ± 4.8	10.7 ± 4.7	21.4 ± 5	18.7 ± 4.9	21.7 ± 13.7
TOC (mg/kg)	$13,600 \pm 3555.3$	$12,279.7 \pm 1533.5$	$20,266.7 \pm 3330.7$	$20,029.3 \pm 1119.4$	$14,400 \pm 1743.6$	$28,286.3\pm5252.9$	$21,066.7 \pm 3330.7$	$17,810.3\pm2011.7$
TP (mg/kg)	1072.7 ± 154.1	1289.7 ± 146.9	1147.7 ± 214.7	1372.7 ± 136.1	2113.7 ± 253	2256.3 ± 153.6	1580.3 ± 257.9	1830 ± 354.9
TN (mg/kg)	809.3 ± 91	882 ± 75.2	987 ± 158.5	1081 ± 150.4	1355.7 ± 200.2	1463 ± 102.6	1264 ± 178.7	1302 ± 155
E.Coli (CFU/g)	6.7 ± 11.5	0 ± 0	38.3 ± 44.8	0 ± 0	8.3 ± 7.6	0 ± 0	0 ± 0	0 ± 0
Coliform (CFU/g)	1866.7 ± 702.4	46.7 ± 37.9	2623.3 ± 2470.1	720 ± 594.3	3866.7 ± 2706.2	683.3 ± 440.7	1163.3 ± 1261	23.3 ± 40.4
CH4 (mg/kg)	13.9 ± 3.7	44.3 ± 53	18.3 ± 5.6	10.8 ± 10.1	35.6 ± 29.1	2317 ± 3180.6	1234.9 ± 1475.8	$8423 \pm 14,216$
$H_2S(\mu M)$	75.8 ± 31	30.5 ± 46	121.5 ± 17	4.2 ± 3.4	11 ± 13.9	316.2 ± 328.3	118.6 ± 56.3	214.1 ± 157.2
Environmental B5			B6		B7		B8	
variables	w	D	W	D	м	D	м	D
Hd	7.6 ± 0.2	7.8 ± 0.2	7.2 ± 0.5	7.8 ± 0.3	7.5 ± 0.3	7.9 ± 0.1	7.2 ± 0.1	7.9 ± 0.1
DO (mg/l)	3.7 ± 0.4	3.8 ± 0.2	3.8 ± 0.1	3.9 ± 0.4	3.5 ± 0.2	3.9 ± 0.1	6.6 ± 0.2	3.3 ± 0.1
TSS (mg/l)	160.7 ± 30.9	96.7 ± 19.5	154.3 ± 25	457 ± 286.5	131.3 ± 12.7	266 ± 77.9	113.3 ± 18	38 ± 6.6
TDS (mg/l)	252.7 ± 147.3	333.7 ± 70.3	97 ± 9.8	208 ± 19.3	50.3 ± 2.1	144 ± 5.6	56.7 ± 4	86.7 ± 2.3
Sal (%c)	0.3 ± 0	0.3 ± 0.1	0.1 ± 0	0.2 ± 0	0.1 ± 0	0.1 ± 0	0.1 ± 0	0.1 ± 0
NH4 (mg/kg)	4.4 ± 1.2	72.7 ± 20.4	3.6 ± 1.3	188 ± 57.2	21.1 ± 0.3	79.3 ± 15.4	7.3 ± 2	107 ± 29.5
NO ₃ (mg/kg)	7.3 ± 2.7	13.5 ± 3	9.9 ± 4.3	15.8 ± 1.9	17.5 ± 2.4	18.5 ± 3.6	7.4 ± 0.6	13.9 ± 3.3
TOC (mg/kg)	$20,666.7 \pm 7943.1$	$18,970.3 \pm 1489$	$17,066.7 \pm 3629.5$	$15,185\pm850$	$21,966.7 \pm 2466.4$	$21,966.7 \pm 2466.4$	$17,466.7 \pm 3716.6$	$17,466.7 \pm 3716.6$
TP (mg/kg)	1380.7 ± 1005.9	1933.3 ± 343.5	1881.7 ± 155.4	2047.7 ± 181.6	1101.7 ± 127.4	1352.3 ± 154.7	920.3 ± 100.7	1224.3 ± 121.5
TN (mg/kg)	1121.7 ± 125.3	1389.7 ± 144.2	958 ± 62.4	1256.3 ± 59.7	986.7 ± 197.4	975.3 ± 140.8	1009.7 ± 117.6	1097 ± 92.6
E.Coli (CFU/g)	1110 ± 1485.6	10 ± 17.3	246.7 ± 376.5	0 ± 0	353.3 ± 440.5	0 ± 0	668.3 ± 673.2	0 ± 0
Coliform (CFU/g)	$49,033.3\pm47,980.2$	60 ± 60	883.3 ± 1400.1	6.7 ± 11.5	$11,400 \pm 9824.5$	26.7 ± 46.2	$13,233.3 \pm 18,852.1$	26.7 ± 30.6
$CH_4 (mg/kg)$	1210.1 ± 1944.8	5272.9 ± 8995	831.3 ± 663	129 ± 77.7	193.1 ± 290.9	1741.2 ± 2372	$12,858.3 \pm 13,639.6$	2525.9 ± 2160.6
H_2S (μM)	44.4 ± 35.3	112.5 ± 103.3	71.9 ± 22.8	99.6 ± 117.8	96.9 ± 26.1	13.7 ± 14.9	83.9 ± 26	98.2 ± 91.7

at B8 also in the wet season. Significant differences were found between stations (p = 0.001), between B1, B2, and B3, and between B8 and B4, B5, and B7.

Total dissolved solids (TDS) and total suspended solids (TSS) concentrations were in general higher in the downstream than in the upstream stations. TDS ranged from 50.3 ± 2.1 (mg/L) at station B7 in the wet season to 8046 ± 506.4 (mg/L) at B1 in the dry season. TSS ranged between 35.3 ± 12.9 (mg/L) at station B3 in the dry season and 457 ± 286.5 (mg/L) at B6 in the dry season. Significant differences were found for TDS and TSS for the factor station but not for dam sides and season.

In order to visualize the most prominent environmental gradient, a PCA was performed. The first two axes PC1 explain 45.1% and PC2 contributed to 29.9% of the variation. Along PC1 axis, coliform (0.77) and *E. coli* (0.59) were important variables that differentiated sampling stations (Fig. 4). Stations before (downstream) and after (upstream) the dam were mainly separated at different sides of PC2 axis. Along this axis, the variables CH₄ (0.83), H₂S (0.29), TDS (-0.36), and Sal (-0.18) correlated well (Fig. 4).

The characteristics of the intertidal nematode communities

Averages and standard deviations of nematode densities per station in both dry and wet season are shown in Fig. 5. Average nematode densities in the wet season ranged from 63.7 ± 29.7 inds/10 cm² at B8 to 870.9 ± 575.9 inds/10 cm² at B1. The densities varied between from 95.5 ± 69.8 inds/10 cm² and 3269.4 ± 3928.4 inds/10 cm² in the dry season. The nematode densities from the downstream stations were more abundant compared to those situated in the stations upstream from the dam for both the dry and wet season. In some stations such as B4, B5, and B6, the nematode densities in the dry season were on average higher in comparison to the wet season. In contrast, nematode abundance was higher in the wet season at the stations B2, B3, and B8, with considerably higher densities of more than $3000 \text{ inds}/10 \text{ cm}^2$ at stations B2 and B3.

ANOVA analyses found significant differences in densities. And, there was also a significant interaction effect of the factors dam and season on the nematode density and the number of nematode genera (Table 2). The nematode communities consisted of taxa belonging to the two main classes: Chromadorea and Enoplia. In the dry season, 137 genera were found belonging to 49 families of 11 orders. In the wet season, 102 genera distributed over 43 families belonging to 11 orders were identified. Both seasons were similar in terms of order composition including Araeolaimida, Chromadorida, Desmodorida, Desmoscolecida, Dorylaimida, Enoplida, Monhysterida, Mononchida, Plectida, Rhabditida, and Triplonchida.

The relative abundance of Chromadorea was in total 74.29% in the wet season and it reduced to 66.2% in the dry season. In contrast, the relative abundance of class Enoplia was lower in the wet season (25.71%) compared to that in the dry season (33.8%).

The orders Monhysterida and Araeolaimida exhibited the highest densities and accounted for 51.34% and 25.23% of the total density in the wet season. The same orders in addition to the Chromadorida contributed to highest densities in the dry season representing 26.61%, 24.03%, and 23.38% of the total community, respectively.

The families Xyalidae, Linhomoeidae, and Axonolaimidae were the most dominant families, respectively, accounting for 35.37%, 13.38%, and 12.49% of the total number of individuals in the wet season. The most abundant families in the dry season were Xyalidae, Chromadoridae, Comesomatidae, and Axonolaimidae, respectively, contributing to 22.4%, 13.38%, 13.31%, and 13.28% of the total densities.

Ptycholaimellus, Parodontophora, Terschellingia, Desmodora, Metadesmolaimus, and Daptonema were the most abundant genera, each accounting for more than 5% of the total density over all stations and both seasons. Ptycholaimellus and Parodontophora were in general most abundant representing more than 10% of all specimens. The dominant genera which individually contributed to more than 5% of the total density in the wet season were Daptonema (15.93%), Parodontophora (12.58%),Theristus (8.27%), *Terschellingia* (7.06%),and Sphaerotheristus (5.85%). The dominant genera in the dry season were Ptycholaimellus (16.94%), Parodontophora (9.07%), Metadesmolaimus (8.24%), Desmodora (8.12%), Terschellingia (6.86%), and Hopperia (5.2%). In both seasons, Daptonema, Hopperia, and Parodontophora were the most abundant genera in the downstream part of the dam, while the genus Theristus was only abundant in the upstream part of the dam and



Fig. 2 Averages and standard deviations of methane (a) and hydrogen sulfide (b) concentrations in the dry and the wet seasons in the Ba Lai River, Vietnam

the genus *Desmodora* was a dominant genus in the downstream part of the dam in the dry season.

The richness was higher in those stations located in the downstream part of the dam. The number of genera was also on average higher in the wet season for all stations, except for B1 and B8 where it remained the same for both seasons (Fig. 6). ANOVA analyses found significant differences between both dam sides (Table 2). Two-way ANOVA also indicated an interaction effect of both factors dam and season on the genera richness.

The Hurlbert and Shannon–Wiener index were also on average higher in those stations located in the downstream side of the dam. These diversity indices were on average higher in the wet seasons compared to the dry season in most stations downstream and in stations B7 and B8, while in stations B4, B5, and B6, they remained unchanged between seasons (Fig. 7a, b). Significant differences were found between stations for ES (50). There was a significant interaction effect of dam and season for the Hurlbert and Shannon Wiener index (Table 2).

The Hill's indices were higher in the wet season for all stations compared to those in the dry season, except for station B5. These indices were also considerably higher in the downstream stations than those situated in the upstream stations of the dam (Fig. 8). ANOVA analyses showed a significant interaction effect of both factors dam sides and season for the Hill's indices N1 and N2 (Table 2).

The values of the maturity index (MI) index varied between 2 and 3 and were slightly higher in some



Fig. 3 Averages and standard deviations of pH, DO (mg/L), and salinity (%) in the wet season (W) and the dry season (D) in the Ba Lai River, Vietnam

Fig. 4 Principal component analysis based on sedimentary abiotic factors measured in 8 transects and 2 seasons. Circle of the graph represents the vector unit. The percentage of explained variance by two first PCs: 75% (PC1=45.1%; PC2=29.9%) (Note: wbd, wet below dam; wad, wet above dam; dbd, dry below dam; dad, dry above dam)



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PC1

stations in the dry season. PERMANOVA analysis indicated a significant difference in the MI index between the two seasons (Table 2).

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The nematode communities were presented by all feeding type groups including selective deposit-feeders (1A), non-selective deposit-feeders (1B), epistratum feeders (2A), and predators/omnivores (2B). Group 1B was the most dominant in all stations in both seasons, except for station B1 in the dry season where 2A was more abundant one (Fig. 9a). Whereas the group 2B was rare in the downstream stations of the dam, and slightly increased in abundances at the upstream side, group 1A fluctuated strongly between stations and seasons. ANOVA analyses found significant differences between stations in terms of the feeding groups 2A and 2B and Tukey HSD (for factor dam*season) resulted in the significant difference of feeding type 2A between below dam stations and those in the above dam stations (Table 2). There was also interaction effect of both factors dam and season on feeding groups 1A, 1B, and 2A but not for 2B (Table 2).

The trophic diversity index (TD) ranged from about 0.36 to 0.66 and was on average highest at station B5 in both seasons (Fig. 9b). TDs were on average higher in the dry season in those stations situated in the downstream side, while they were higher in the wet season in the upstream stations of the dam. However, there was no significant difference for TD in the case of factors station, season, and dam.

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Fig. 5 Averages and standard deviations of nematode densities in the wet (W) and the dry seasons (D) in the Ba Lai River, Vietnam

	Table 2	ANOVA/PERMANOVA a	nd pairwise	comparison of	f nematode	communities for	or characteristics
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Characteristics	Factors	<i>p</i> -values	Pairwise comparison (p values in bracket)
Density (N)	station	0.006	B1 & B7 (0.03), B2 & B6 (0.03), B2 & B7 (0.015), B2 & B8 (0.03), B3 & B7(0.022)
	dam*season	0.006	bd:d & ad:d (0.006), bd:w & ad:d (0.006), ad:w & bd:d (0.006), bd:w & ad:w (0.014)
Genera richness (S)	station	0.006	B4 & B1(0.006), B5 & B1(0.006), B6 & B1(0.006), B7 & B1(0.006), B8 & B1(0.006), B4 & B2(0.017), B5 & B2(0.015), B6 & B2(0.014), B7 & B2(0.015), B8 & B2(0.015), B4 & B3(0.015), B5 & B3(0.015), B6 & B3(0.022), B7 & B3(0.015), B8 & B3(0.017)
	dam*season	0.006	bd:d & ad:d (0.006), bd:w & ad:d (0.006), ad:w & bd:d (0.006), bd:w & ad:w (0.006)
Hurlbert -ES(50)	station	0.014	
	dam*season	0.006	bd:d & ad:d (0.017), bd:w & ad:d (0.006), bd:w & ad:w (0.004)
Shannon-Weiner (H')	dam*season	0.006	bd:w & ad:d (0.006), bd:w & ad:w (0.02)
Hill's (N1)	dam*season	0.006	bd:w & ad:d (0.006), bd:w & ad:w (0.022)
Hill's (N2)	dam*season	0.023	bd:w & ad:d (0.017)
Maturity index (MI)	season	0.035*	
%1A	dam*season	0.006	bd:w & ad:w (0.014)
%1B	dam*season	0.014	ad:w & bd:d (0.016)
%2A	station	0.017	B5 & B1 (<0.016)
	dam*season	0.0016	bd:d & ad:d (0.006), bd:w & ad:d (0.006), ad:w & bd:d (0.006)
%2B	station	0.006	B2 & B6 (0.04), B2 & B7 (0.04), B2 & B8 (0.022), B3 & B6 (0.03), B3 & B7 (0.03), B3 & B8 (0.022), B4 & B6 (0.022), B4 & B7 (0.022), B4 & B8 (0.015)
Genus composition	station	0.006*	B1 & B4(0.017), B1 & B5(0.015), B1 & B6(0.015), B1 & B7(0.004), B1 & B8(0.004), B2 & B4(0.015), B2 & B5(0.017), B2 & B6(0.004), B2 & B7(0.015), B2 & B8(0.015), B3 & B4(0.004), B3 & B5(0.022), B3 & B6(0.015), B3 & B7(0.02), B3 & B8(0.015)
	dam*season	0.017*	bd:w & ad:w (0.006), bd:d & ad:d (0.006)

*indicates p values generated from non-parametric analyses). bd dammed downstream, ad dammed upstream, w wet season, d dry season

PERMANOVA analyses based on the relative abundance of the genera composition showed significant differences for the interaction effect of both factors dam sides and season (Table 2), as illustrated by the separation of stations in the MDS (Fig. 10).

The result of the SIMPER analysis indicated the genera responsible for more than 50% of similarity and dissimilarity between dam sides and between seasons (Table 3). Four genera *Halalaimus*, *Parodontophora*, *Hopperia*, and *Linhystera* were the major taxa contributing to the dissimilarity in terms of genus composition between the two dam sites, while

the difference between the dry and the wet seasons was mainly explained by *Theristus*, *Parodontophora*, *Terschellingia*, *Metalinhomoeus*, *Daptonema*, and *Sphaerotheristus* in both sides.

Interaction between nematode communities and environmental variables

Correlations were calculated between nematode community descriptors and environmental variables for both seasons separately (Tables 4 and 5). In the wet season, methane was positively correlated with **Fig. 6** Averages and standard deviations of genera richness in the wet (W) and the dry (D) seasons in the Ba Lai River, Vietnam



the density of the non-selective deposit-feeders (1B), while hydrogen sulfide (H_2S) showed positive correlations with the feeding groups non-selective deposit-feeders (1B) and epistratum feeders (2A) as well as with total density. The 1B feeding type nematodes also showed significant correlations with bacteria *E. coli* and other environmental variables such as NO_3^- , NH_4^+ , TN, TP, and TOC as well as pH, DO, TSS, and salinity. The other feeding groups (1A, 2A, 2B) were also positively correlated with a number of environmental characteristics in addition to methane and H_2S (Table 4). Especially, NO_3^- , salinity, DO, and pH showed a significant correlation with most univariate nematode descriptors except density. Furthermore, the environmental variable

CH₄ showed significant positive correlation with DO (r=0.7, p<0.001) and *E. coli* (r=0.47, p=0.02) and H₂S showed a significant correlation with TSS (r=0.55, p=0.006).

In the dry season, only density was positively correlated with H₂S. In contrast, methane did not show any significant correlation with the nematode communities. Density also showed a positive correlation with coliform and TOC, while bacteria *E. coli* showed a negative correlation with MI, but positive correlations with both epistratum feeders (2A) and TD. Genera richness showed a positive correlation with pH, DO, TDS, salinity, and NO₃⁻. The other biodiversity-indices (ES(50), H', N1, N2) were positively correlated with NO₃⁻. In this season, CH₄ showed a



Fig. 7 Averages and standard deviations of indices: a Hurlbert (ES(50)), b Shannon-Weiner (H') in the wet (W) and the dry (D) seasons in the Ba Lai River, Vietnam





negative significant correlation with TDS (r=-0.44, p=0.03) and salinity (r=-0.48, p=0.018), while H₂S showed a positive correlation with TN (r=0.54, p=0.007) and TP (r=0.435, p=0.03). Furthermore, both CH₄ and H₂S have a significant correlation in the dry season (r=0.64, p=0.001).

Cumulative variability of the environmental variables explained 48.35% of the total nematode community variability in the DistLM (Fig. 11). Axis 1 explains 22.1% of the variation and axis 2 explains 6.38% of the variation. The DistLM plot indicates a significant effect of CH₄ (p=0.0007), bacteria *E. coli* (p=0.014), pH (p=0.004), TDS (p=0.0001), NH₄⁺ (p=0.009), NO₃⁻ (p=0.039), and salinity (p=0.0001) on the nematode community composition based on the relative density.

Discussion

Concentration of methane and hydrogen sulfide gases in the environmental situation of dam impact

The accumulation of methane and hydrogen sulfide gases in the area appears to be a consequence of the dam. The methane concentrations showed low values in the downstream portion of the study, where salinity was high. In the dry season, methane showed a significant negative correlation with salinity. However, in the stations upstream of the dam, the highest concentrations of methane were observed at station B4 and station B8. The higher concentrations at B4 are similar with the findings for the subtidal of the same area as Nguyen et al. (2020) found high methane



Fig. 9 Proportion of feeding groups (a) and trophic diversity index (b) in the wet (W) and the dry (D) season in the Ba Lai River, Vietnam



Fig. 10 MDS illustrates for nematode communities in part of downstream stations (bd) and upstream stations (ad) in both seasons in the Ba Lai River, Vietnam (Note: wbd, wet below dam; wad, wet above dam; dbd, dry below dam; dad, dry above dam)

concentration at subtidal stations close to both sides of the dam. When the river brings organic material such as dead plants and animal carcasses from upstream, it stops at the dam barrier, sinks down, and decomposes. Lin (2011) also noted that decaying plant and animal matters settle to the non-oxygenated bottom of a stagnant reservoir where the decomposition process eventually releases dissolved methane as a consequence of methanogenesis processes at the river bottom. Indeed, when oxygen is depleted, anaerobic processes such as methane production can prevail (Bartlett & Harriss, 1993; Oremland, 1988). However, methanogens can also tolerate oxygen to quite some extent and redox-active organic carbon compounds may affect the rates and pathways of CH_{4} production due to different methanotrophic microbial communities among the various environments (Conrad, 2020). This could explain why dissolved oxygen was low in water and the strong correlation with methane in the wet season. Because of this ability of methanogenic microbial processes, the upstream portion of the Ba Lai River works like a shallow

"lake-river" that could become an artificial wetlands of methane emission.

Furthermore, the environmental quality in terms of DO and TSS in some stations was lower than the National Technical Regulations on surface water quality B2 standards (Ministry of Environment and Natural Resources, 2015). The nutrients NH_4^+ , NO_3^- , TP, TN, and TOC had values exceeding regulatory limits, which indicate organic pollution. According to Istvánovics (2009), the construction of a dam led to increased eutrophication, especially nitrate and ammonium, which are more mobile in sediments than orthophosphate, since the latter is chemisorbed by clay minerals, iron (III) oxy-hydroxides, and carbonates. Consequently, this stimulates sedimentation processes in the riverbed. Additionally, the presence of E. coli indicates accurate bacterial contamination by fecal matter in the river (Dufour, 1977). The concentrations of coliform and E. coli in the study area were much higher than those provided by the Vietnamese Standard for Natural Water (Ministry of Environment and Natural Resources, 2015). Overall, the results

Table 3	The percentage of similari	ity/dissimilarity of e	ach group/between	groups and the	proportion c	contribution of	f most important
genera re	sponsible for that similarit	y/dissimilarity based	on $\log(x+1) - trained respectively.$	ansformed nema	tode data		

Groups	Similarity/ dissimilarity (%)	Таха	%	Таха	%
Dammed downstream (bd)	47.85	Parodontophora	8.17	Hopperia	4.08
		Daptonema	6.14	Sabatieria	3.4
		Halalaimus	5.49	Paracomesoma	3.39
		Terschellingia	5.24	Desmodora	3.27
		Linhystera	4.85	Monhystrella	2.81
		Sphaerotheristus	4.16		
Dammed upstream (ad)	44.36	Theristus	14.23	Sphaerotheristus	9.26
		Terschellingia	11.48	Mesodorylaimus	7.62
		Parodontophora	9.43	Daptonema	7.37
bd & ad	70.87	Halalaimus	3.33	Ptycholaimellus	2.02
		Parodontophora	3.08	Amphimonhystrella	1.9
		Hopperia	3.06	Comesoma	1.89
		Linhystera	2.9	Metalinhomoeus	1.84
		Paracomesoma	2.69	Sphaerolaimus	1.82
		Desmodora	2.66	Viscosia	1.73
		Sabatieria	2.58	Sphaerotheristus	1.71
		Daptonema	2.54	Leptolaimus	1.67
		Theristus	2.36	Chromadorita	1.67
		Terschellingia	2.36	Anoplostoma	1.61
		Eumorpholaimus	2.1	Mesodorylaimus	1.57
		Dichromadora	2.06	·	
wet	53.10	Theristus	11.45	Terschellingia	7.4
		Parodontophora	8.94	Mesodorylaimus	5.65
		Sphaerotheristus	7.8	Rhabdolaimus	4.42
		Daptonema	7.5		
dry	37.40	Terschellingia	13.18	Sphaerotheristus	7.99
		Theristus	10.64	Mesodorylaimus	6.5
		Parodontophora	9.31	Daptonema	6.38
Wet & dry	60.03	Theristus	4.08	Monhystrella	2.35
		Parodontophora	3.9	Desmodora	2.26
		Terschellingia	3.71	Dichromadora	2.14
		Metalinhomoeus	3.47	Halalaimus	1.91
		Daptonema	3.41	Udonchus	1.89
		Sphaerotheristus	3.09	Paraplectonema	1.77
		Mesodorylaimus	2.87	Achromadora	1.7
		Metadesmolaimus	2.78	Prismatolaimus	1.67
		Mononchulus	2.38	Viscosia	1.67
		Rhabdolaimus	2.38	Chromadorita	1.65

indicate that the Ba Lai River is not only organically polluted, but it is also in a state of eutrophication. Moreover, input sources of organic carbon from adjacent agricultural activities, such as livestock, poultry, aquacultures, and domestic wastewater from farms and gardens in the basin, are stimulating methanogenesis in surface sediment, and in the deeper sediment layer by diffusion of dissolved labile organic

Table 4 Cc	orrelatio	ons (r and	<i>p</i> value) betv	veen enviror	nmental varia	ables and 1	nematode (communit	y characteris	tics in the w	et season (N	=73)			
		Hq	DO	SST	TDS	Sal	NH_4	NO_3	TOC	TP	ΛL	E.coli	Coliform	CH_4	H_2S
S	r	0.66	0.574	0.384	0.473	0.575	0.331	0.793	0.564	0.558	0.604	0.103	0.146	0.05	0.229
	d	0.000	0.000	0.001	0.000	0.000	0.004	0.000	0.000	0.000	0.000	0.388	0.217	0.672	0.051
Z	r	0.051	-0.007	-0.023	0.475	0.053	0.332	0.129	0.396	0.289	0.309	-0.06	-0.009	- 0.066	0.497
	d	0.669	0.956	0.845	0.000	0.654	0.004	0.276	0.001	0.013	0.008	0.612	0.941	0.578	0.000
ES(50)	r	0.940	06.0	0.268	0.175	0.874	0.297	06.0	0.301	0.302	0.326	0.063	0.071	0.058	0.208
	d	0.00	0.00	0.022	0.139	0.00	0.011	0.00	0.010	0.009	0.005	0.597	0.550	0.628	0.077
$H^{*}(\log_{2})$	r	0.984	0.997	0.069	- 0.009	0.98	0.18	0.795	-0.046	-0.032	-0.03	0.018	-0.006	-0.013	0.086
	d	0.00	0.00	0.560	0.939	0.00	0.127	0.000	0.697	0.785	0.800	0.88	0.963	0.915	0.468
NI	r	0.924	0.895	0.227	0.134	0.876	0.249	0.886	0.23	0.229	0.247	0.043	0.058	0.023	0.165
	d	0.00	0.00	0.053	0.258	0.00	0.033	0.00	0.050	0.051	0.035	0.719	0.623	0.844	0.162
N2	r	0.965	0.957	0.15	0.041	0.937	0.211	0.852	0.093	0.099	0.108	0.03	0.032	-0.001	0.119
	d	0.00	0.00	0.205	0.731	0.00	0.074	0.00	0.435	0.401	0.361	0.785	0.789	0.991	0.314
Nœ	r	0.98	0.991	0.079	-0.012	0.975	0.179	0.8	-0.035	-0.020	-0.02	0.023	0.002	-0.019	0.081
	d	0.00	0.00	0.508	0.918	0.00	0.132	0.000	0.771	0.867	0.869	0.845	0.984	0.876	0.494
III	r	0.98	0.997	0.054	-0.023	0.979	0.177	0.776	-0.072	-0.057	-0.056	0.013	-0.015	-0.016	0.079
	d	0.00	0.00	0.650	0.848	0.00	0.135	0.000	0.545	0.634	0.637	0.912	0.899	0.893	0.507
1A	r	0.695	0.642	0.43	0.035	0.595	0.341	0.71	0.355	0.385	0.399	- 0.064	0.053	0.009	0.184
	d	0.000	0.000	0.000	0.768	0.000	0.003	0.000	0.002	0.001	0.000	0.593	0.655	0.941	0.119
1B	r	0.455	0.36	0.345	0.075	0.27	0.314	0.505	0.674	0.629	0.721	0.272	0.215	0.273	0.321
	d	0.000	0.002	0.003	0.531	0.021	0.007	0.000	0.000	0.000	0.000	0.020	0.067	0.019	0.006
2A	r	0.648	0.594	0.194	0.604	0.641	0.326	0.699	0.307	0.308	0.307	-0.006	-0.025	-0.017	0.279
	d	0.000	0.000	0.101	0.000	0.000	0.005	0.000	0.008	0.008	0.008	0.960	0.831	0.888	0.017
2B	r	0.789	0.775	0.201	-0.077	0.712	0.281	0.657	0.147	0.143	0.137	0.08	0.019	0.04	0.123
	d	0.000	0.000	0.088	0.517	0.000	0.016	0.000	0.215	0.229	0.248	0.499	0.873	0.736	0.299
TD	r	0.964	0.99	0.011	-0.046	0.979	0.143	0.742	-0.138	-0.12	-0.124	0.004	-0.028	-0.031	0.049
	d	0.00	0.00	0.926	0.696	0.00	0.229	0.000	0.244	0.310	0.297	0.976	0.816	0.794	0.679

Table 5 C	orrelati	on (r and p	value) betv	ween enviroi	nmental var	iables and n	lematode co	mmunity ch	naracteristic	s in the dry	season (N=	=24)			
		Hd	DO	TSS	TDS	Sal	NH_4	NO_3	TOC	TP	TN	E.coli	Coliform	CH_4	H_2S
S	r	0.525	0.426	-0.148	0.771	0.772	- 0.126	0.518	- 0.252	-0.16	-0.258	-0.233	0.159	-0.13	-0.193
	d	0.008	0.038	0.491	0.000	0.000	0.559	0.009	0.235	0.455	0.224	0.272	0.458	0.544	0.366
Z	r	0.397	-0.233	-0.258	0.368	0.366	0.157	-0.005	0.547	0.038	0.100	-0.097	0.792	-0.161	0.462
	d	0.055	0.274	0.224	0.077	0.079	0.463	0.980	0.006	0.861	0.642	0.652	0.000	0.453	0.023
ES(50)	r	0.272	0.247	-0.011	0.389	0.389	-0.078	0.493	-0.027	0.218	0.002	-0.198	0.135	-0.009	-0.045
	d	0.199	0.244	0.959	0.060	0.060	0.717	0.014	0.899	0.307	0.994	0.353	0.531	0.967	0.835
$H^{*}(\log_{2})$	r	0.08	0.007	0.056	0.166	0.166	- 0.095	0.482	0.079	0.229	0.035	-0.194	0.18	0.004	0.005
	d	0.709	0.973	0.795	0.437	0.437	0.658	0.017	0.711	0.281	0.870	0.365	0.399	0.985	0.998
N1	r	0.169	0.065	0.005	0.232	0.233	- 0.099	0.549	0.079	0.209	-0.036	-0.181	0.123	-0.022	-0.046
	d	0.429	0.763	0.981	0.275	0.273	0.645	0.005	0.715	0.327	0.867	0.399	0.567	0.917	0.829
N2	r	-0.044	-0.091	0.008	0.051	0.051	-0.124	0.444	0.127	0.223	0.003	-0.139	0.116	-0.008	-0.06
	d	0.840	0.673	0.970	0.814	0.812	0.563	0.030	0.554	0.294	066.0	0.518	0.589	0.969	0.780
N ₈₀	r	-0.223	-0.158	0.024	-0.044	-0.043	-0.165	0.353	0.09	0.182	0.008	-0.084	0.062	0.009	-0.083
	d	0.296	0.460	0.912	0.837	0.840	0.440	0.091	0.676	0.395	0.972	0.697	0.775	0.965	0.701
IM	r	0.005	-0.105	0.226	-0.176	-0.175	-0.37	-0.006	0.034	-0.061	-0.399	-0.451	-0.268	-0.213	-0.074
	d	0.980	0.626	0.287	0.409	0.414	0.075	0.976	0.874	0.776	0.053	0.027	0.205	0.316	0.731
1A	r	-0.119	-0.471	0.277	-0.326	-0.326	0.059	-0.100	0.068	0.079	0.119	-0.239	0.026	-0.022	0.007
	d	0.580	0.020	0.190	0.120	0.120	0.784	0.642	0.752	0.715	0.578	0.261	0.902	0.918	0.975
1B	r	-0.342	0.119	-0.157	-0.286	-0.288	0.206	-0.042	-0.029	0.189	0.455	0.415	-0.063	0.343	-0.016
	d	0.102	0.580	0.465	0.176	0.172	0.334	0.847	0.891	0.375	0.025	0.044	0.771	0.100	0.940
2A	r	0.533	0.245	- 0.099	0.698	0.699	-0.276	0.173	-0.065	-0.157	-0.405	-0.215	0.242	-0.254	0.097
	d	0.007	0.247	0.644	0.000	0.000	0.192	0.420	0.763	0.463	0.049	0.313	0.255	0.230	0.651
2B	r	-0.18	-0.095	0.131	-0.326	-0.324	0.045	-0.11	0.087	-0.155	- 0.244	- 0.089	-0.339	-0.144	-0.148
	d	0.399	0.660	0.540	0.120	0.122	0.837	0.609	0.686	0.470	0.251	0.678	0.105	0.502	0.491
CIL	r	0.125	0.365	-0.259	0.160	0.159	0.021	-0.147	- 0.097	-0.029	0.201	0.504	-0.059	0.213	0.021
	d	0.559	0.079	0.222	0.454	0.458	0.921	0.493	0.651	0.891	0.347	0.012	0.782	0.318	0.923

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Fig. 11 The DistLM plot of nematode communities and environmental variables in the wet and the dry season in the Ba Lai River, Vietnam (*Note*: wbd, wet below dam; wad, wet above dam; dbd, dry below dam; dad, dry above dam)

matter (Schulz & Conrad, 1995). Additionally, under eutrophic conditions, the allochthonous input contributed to high autochthonous production increasing measures of organic matter (TOC, TN, TP) and accumulation of allochthonous organic matter in the littoral zone contributed to the increase in methane concentrations in the sediment (D'Ambrosio & Harrison, 2021; Furlanetto et al., 2012).

Similar to methane, hydrogen sulfide was also high at almost all stations upstream from the dam. However, the highest concentration was found in the dry season at station B3, which is downstream of the dam. This gas expressed a negative correlation with TSS in the dry season while it showed a significantly positive correlation with both TP and TN in the wet season. According to Nguyen et al. (2020), the average H_2S values in the subtidal area of the dammed Ba Lai River were higher than that in an adjacent dam-free Ham Luong River in the Mekong estuarine system due to high accumulation of organic matter. Meanwhile, Mitterer et al. (2001) stated that the high organic load in an oxygen-depleted environment could lead to anoxic conditions which in turn lead to the reduction of sulfate followed by the generation of methane. The authors assumed that the co-production of these reduced gases was due to the presence of noncompetitive substrates for the two types of microbes. Such is the case of the Ba Lai River, where methane and hydrogen sulfide showed significant correlation in the dry season and sulfate reduction to sulfide and methanogenesis were mutually exclusive microbial reactions (Mitterer et al., 2001). Our results are also further supported by Fedorov et al. (2019), who found a close relationship between the concentrations of methane and hydrogen sulfide, as well as the abundance of sulfite-reducing bacteria *clostridia* in the bottom sediments.

Intertidal nematode communities in the dam impacted river

Our results found that the intertidal nematode communities in the Ba Lai River typically display high density and diversity. However, the communities were strongly influenced and shaped by the effect of the dam compared to the other factors (i.e., station or season). Indeed, the dam represents a barrier for it divides the river ecosystem into a downstream and an upstream section also reflected in the subtidal nematode community structure (Nguyen et al., 2020). Most indices of intertidal nematode assemblages, such as density, genera richness, and bio-indices (ES(50), H', N1, N2), showed a significant dam*season interaction, and pairwise comparison revealed that most downstream stations were significantly different from upstream stations in terms of the abovementioned variables. While epistratum feeders' group (2A) was the most abundant group in the river mouth station B1 in the dry season, predators/omnivores (2B) was exceedingly rare, especially downstream. According to Nguyen et al. (2020), the differences in environmental conditions related to dam in the Ba Lai River were like a driver of change for subtidal nematode community's characteristics.

The presence of the dam was reflected also in the composition of nematode communities which were separated in two significantly different groups. Downstream, the marine nematodes Parodontophora, Daptonema, Halalaimus, and Terschellingia contributed to within group similarity, while upstream communities were mostly Theristus, Terschellingia, Parodontophora, Sphaerotheristus, and Mesodorylaimus. The communities on the upstream side also contained freshwater genera such as Mesodorylaimus, Rhabdolaimus, Metalinhomoeus, Mononchulus, Mononchulus, Mylonchulus, Udonchus, Monhystrella, Prismatolaimus, Metadesmolaimus, and Ironus. According to McLusky (1971), the intrusion of freshwater and marine species in the estuarine area depends on the daily flow of freshwater and the tidal regime. However, in the case of Ba Lai River, the distribution of these species depends on the dam sluice operation schedules throughout the dry and wet seasons (Ngo et al., 2017a).

Noticeably, there was no difference in the nematode order composition between intertidal region (this study) and the subtidal area (Nguyen et al., 2020) of the Ba Lai River, with 11 nematode members including Araeolaimida, Chromadorida, Desmodorida, Desmoscolecida, Dorylaimida, Enoplida, Monhysterida, Mononchida, Plectida, Rhabditida, and Triplonchida. *Parodontophora* and *Theristus* were the two major genera responsible for the differences in subtidal community composition in terms of abundances between the downstream and upstream part of Ba Lai estuary (Nguyen et al., 2020). However, in the intertidal area, *Parodontophora*, *Halalaimus*, *Hopperia*, and *Linhystera* were the main contributors to the dissimilarities between the dam sides. In particular, Parodontophora communities were found to have wide distribution in the Mekong estuarine system (Ngo et al., 2016). Seasonally, *Theristus, Terschellingia, Metalinhomoeus, Daptonema*, and *Sphaerotheristus* were most responsible for the dissimilarity in nematode assemblages between the dry and wet seasons. Moreover, genus *Desmodora* was reported to have a high contribution to group similarity (61.1%) at the river mouth in the Mekong estuarine system (Ngo et al., 2016) but not in the Ba Lai River where they only were dominant in the downstream stations during the dry season.

How are the intertidal nematode communities associated with methane, hydrogen sulfide, and relevant environmental variables in the presence of a dam?

The dam has greatest influence on intertidal nematode communities in the Ba Lai River by dividing the river into two environmentally distinct habitats resulting in downstream and upstream nematode communities. Environmental variables demonstrated that the water quality was organically polluted and the DistLM analysis indicated that CH₄, E. coli, pH, TDS, NH₄⁺, NO₃⁻, and salinity values were most closely related with variations in nematode communities. CH_4 was positively correlated to feeding type 1B in the wet season and this was the most dominant feeding group, including the genera Parodontophora, Metadesmolaimus, Daptonema, Theristus, Pseudolella, Sphaerotheristus, Sabatieria, and Metalinhomoeus. According to Adão et al. (2009) and Danovaro and Gambi (2002), the proportions of feeding guilds in nematode assemblages is associated with food availability and quality and feeding group 1B is positively correlated with the bacteria E. coli. Indeed, CH₄ concentration showed a positive correlation with DO and bacteria E. coli in the wet season but a negative correlation with TDS and salinity in the dry season. These correlation patterns suggest that methane concentration increases in organically polluted conditions in the upstream area that is often effected by waste water discharge and runoff from the domestic and agricultural sources in the basin (Ngo et al., 2017a; Vettil and Ngo, 2018). In fact, human settlements and rice cultivation may have major roles in causing water impairment (Nguyen et al., 2021). Hamzah and Hattasrul (2008) found that a high amount of sediment load, like in the case of the Ba Lai river, is often related to high concentrations of pathogenic bacteria because they can be attached with the sediment particles (Murdoch & Cheo, 1996). High abundance of bacteria in sediment were also an important food resource for group 1B (Michiels & Traunspurger, 2004). Our result deduced that CH_4 can have close relationship with the high percentage of feeding type 1B nematodes in the condition of fecal and organic pollution pointing to the indirect impact of the Ba Lai dam on the inhabiting fauna.

Although H₂S did not significantly correlate to bacteria E. coli but it expressed a negative correlation with TSS in the dry season and positive correlations with TP and TN, under the eutrophic condition. The DISTLM performance did not indicate an effect of H₂S on the distribution pattern of nematode communities. However, H₂S showed a significant positive correlation to feeding types 1B and 2A nematodes in the wet season and to community density in the dry season. According to Vanreusel et al. (2010), the strong biochemical gradients such as high H₂S and food limitations may lead to the change in the abundance of assemblages and to the success of nematode genera characterized by specific trophic or lifestyle modes. Although comprising different habitats, this theory might explain the correlation between hydrogen sulfide and both feeding groups non-selective deposit-feeders (1B), epistratum feeders (2A), and community densities. Our results were also supported by the case study on free living nematodes from the Cienfuegos Gulf (Cuba), which showed that chemical stressors such as hydrogen sulfide were important factors affecting the structure and distribution of nematode communities (Armenteros et al., 2010). However, it is in contrast with the experiment of Rodriguez-Kabana et al. (1965) who reported that nematode densities declined when hydrogen sulfide increased in flooded rice fields.

In both dry and wet season, the bio-indices were positively correlated with NO_3^- in the sediment. Like observation in the subtidal nematode communities of the Ba Lai River (Nguyen et al., 2020), the genera richness and the Shannon–Wiener index positively correlated with pH and NO_3^- . Moreover, the MI index showed positive correlations with salinity, DO, pH, and NO_3^- in the wet season, but was negatively correlated with *E. coli* in the dry season. The MI index was also found to have significant positive

correlation with $NO_2^- + NO_3^-$ in whole Mekong estuarine system (Ngo et al., 2016). The densities of nematode assemblages were positively correlated with TDS, NH_4^+ , TOC, TN, and TP in the wet season and with TOC and coliform in the dry season. This is similar to the intertidal nematode communities observed in the Mira estuary, where a high abundance of nematodes was found in the enrichment of organic matter (Adão et al., 2009). In contrast with the subtidal nematode communities in the Ba Lai River study of Nguyen et al. (2020), there was no significant correlation between density and any environmental variables. This is different from lab experiments of Schratzberger and Warwick (1998), who reported that the addition of organic matter resulted in a significant decrease in most of univariate measures. The author also found, in muddy conditions, that the increasing amount of organic matter led to the decrease of diversity caused by declining abundances of dominant nematode species (Schratzberger & Warwick, 1998).

Similar to other previous studies, salinity and NO_3^- were environmental factors associated with many characteristics of the nematode communities such as density, bio-indices, feeding types, trophic diversity, and mature index when they also played important role in shaping the structure of nematode communities (Adão et al., 2009; Attrill, 2002; Ngo et al., 2016; Smol et al., 1994). However, this observation was dissimilar to subtidal nematode communities in the Ba Lai River which did not show any correlation with salinity (Nguyen et al., 2020). The reason may be due to the periodic opening and closing of a dam, which can drastically affect the nematode communities by influencing salinity concentration (Nicholas et al., 1992).

Conclusion

The Ba Lai dam operation limits the water flow circulation scheme, leading to high deposition of sediment rich in organic matter in the upstream area and alters the river ecosystem. Methane and hydrogen sulfide are concentrated in the sediment accumulation areas such as around the sluice gate. Methane concentration was highest in the upstream side of the dam, while hydrogen sulfide highest in the downstream side. Methane and hydrogen sulfide were significantly correlated to the nutrient contents and organic pollution in the river. The results showed that methane and hydrogen sulfide have an effect on the characteristics of nematode communities such as non-selective deposit-feeders (1B), epistratum-feeders (2A), community densities, and dominant genera. Nematode communities were clearly separated into two assemblages in the downstream and upstream communities. Due to the eutrophic conditions and deposition under low water circulation caused by the dam, the ecological processes of the Ba Lai River appear to be converted into a shallow lake and would produce a large amount of greenhouse gases, including methane and hydrogen sulfide.

Author contribution All authors have equally contributed to the conceptualization; methodology, NXQ, NTMY, AV, LL, NVD, BKV; data acquisition: NXQ, NTMY, TTT, NVD, PNH; statistical and data analysis: NXQ, NTMY, AV, LL, TTT; mapping, BKV; investigation, NXQ, NTMY, AV, LL, TTT; mapping, BKV; investigation, NXQ, NTMY, TTT, AV, LL, PNH, BKV, NTHY, NDH, HQB, NHQ, LP; writing original draft preparation, NXQ, NTMY; revision, AV, LL, LP; supervision, AV, NDH; project administration, NXQ. All authors have substantially contributed for reviewing and agree to the published version of the manuscript.

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Declarations

Conflict of interest The authors declare no competing interests.

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