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HOW VARIABLE ARE NEMATODE COMMUNITIES RESPONDING TO SEASONAL FACTOR?

Ngo Xuan Quang[1, *], Nguyen Ngoc Chau[2], Ann Vanreusel[3]

[1]*Department of Environmental Management and Technology, Institute of Tropical Biology, VAST, 85 Tran Quoc Toan, Dist. 3, Ho Chi Minh city, Vietnam*

[2]*Department of Nematology, Institute of Ecology and Biological Resources, VAST, 18 Hoang Quoc Viet, Cau Giay, Hanoi, Vietnam*

[3]*Ghent University, Ledeganckstraat 35, B-9000 Gent, Belgium*

[*]Email: ngoxuanq@gmail.com

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ABSTRACT

The temporal variation of nematode communities in 8 mouth stations of the Mekong River system was investigated in order to compare the change between the dry and the wet season. The nematode communities were characterized in terms of densities, diversities, maturity index, trophic structure and age structures. The results showed that there were no particular temporal trends in biotic parameters measured except for the Shannon – Wiener diversity index which shows significantly weakly differences between seasons. Our results showed that the spatial differences are larger than the temporal variation. Sediment composition remained the most important factor explaining the community patterns.

Keywords: nematode communities, seasonal scale, Mekong estuary.

1. INTRODUCTION

Almost all studies on seasonality in estuarine nematodes were done in temperate regions characterized by time series over 4 obvious seasons: spring, summer, autumn and winter [1, 2]. Furthermore, Li and Vincx [3] noted that most time series studies on nematodes have a low temporal resolution. Some studies provide a general description of seasonality in tropical estuarine meiofauna or benthic communities [4, 5] but although many studies were done on estuarine nematodes from (sub) tropical estuaries [4, 6, 7, 8, 9, 10, 11, 12] no study was comparing between dry and rainy seasons except the study by Nozais et al. [13] and Boufahja et al. [14] with very little seasonal information on nematodes. There are also 2 unpublished PhD dissertations of Lai [15] and Nguyen [16] to investigate the nematode communities with some description of the seasonal variation in the Can Gio mangrove forests, South Vietnam.

Hence, in order to understand the ecology of estuarine nematodes from the Mekong and especially to use them in monitoring studies it is crucial to know how much the structure of nematode communities varies between 2 tropical seasons. The main aim of this study is therefore to identify and understand the difference in general structure of the present nematode communities between the wet season and the dry season in the Mekong estuary. We selected one station from the mouth of each estuary representing a north south gradient and sampled them respectively in the dry and wet season of two consecutive years.

2. METHODOLOGY

2.1. Study area, sampling coordinates and map

Nematode samples were collected at the mouth of 8 estuaries (Cua Tieu, Cua Dai, Ba Lai, Ham Luong, Co Chien, Cung Hau, Đinh An and Tran De) in the wet season (September 2008) and dry season (March 2009) (figure 1).

2.2. Abiotic samples and data collection

Some environmental water parameters such as temperature, dissolved oxygen and pH were measured at each station in both seasons. The samples for grain size analysis were collected and analyzed for the dry season only.

2.3. Nematode samples in collection and processes

The nematode samples were collected using cores of 3.6 cm diameter (10 cm^2 surface area) and 10 cm high. The cores were pushed down into the sediment up to 10 cm deep. Per station and sampling event, 3 replicates were taken and collected in plastic bottles. The samples were fixed in 60 ^oC hot of 7 % formalin solution and gently stirred. Samples were decanted and extracted at the laboratory of Marine Biology Section, Ghent University and the Department of Environmental Management and Technology, Institute of Tropical Biology, Vietnam. Samples were sieved and collected through 1000 µm to a 38 µm mesh and extracted by the flotation technique using Ludox-TM50 (specific gravity of 1.18).

To facilitate sorting the nematodes, the samples were stained with 1 % solution of Rose Bengal. Nematodes were identified by using a high magnification microscope Leica (Type III) and Olympus BX41 using different documents [17, 18, 19] and the NEMYS database of the Marine Biology Section, Ghent University, Belgium [20].

2.4. Data analyses

Nematode data were analyzed by using Microsoft Excel, software PRIMER v.6 and STATISTICA 7.0. Diversity of nematode communities was measured using the Shannon-Wiener diversity - H’(log_2) [21] which is calculated from the proportional abundances p_i of each species (abundance of the species (N_i) per total abundances (N_t)): $H' = -\Sigma (p_i*\log (p_i))$ (in which $p_i = N_i/N_t$ = relative abundance of the i^{th} species or genus. The Maturity index – MI [22] was also used. The formula for calculating the Maturity Index is $MI = (\Sigma v_i f_i)/n$ where v_i is the c–p value for the nematode family i, f_i is the frequency of nematode family i, and n is the total number of individual nematodes in the sample.

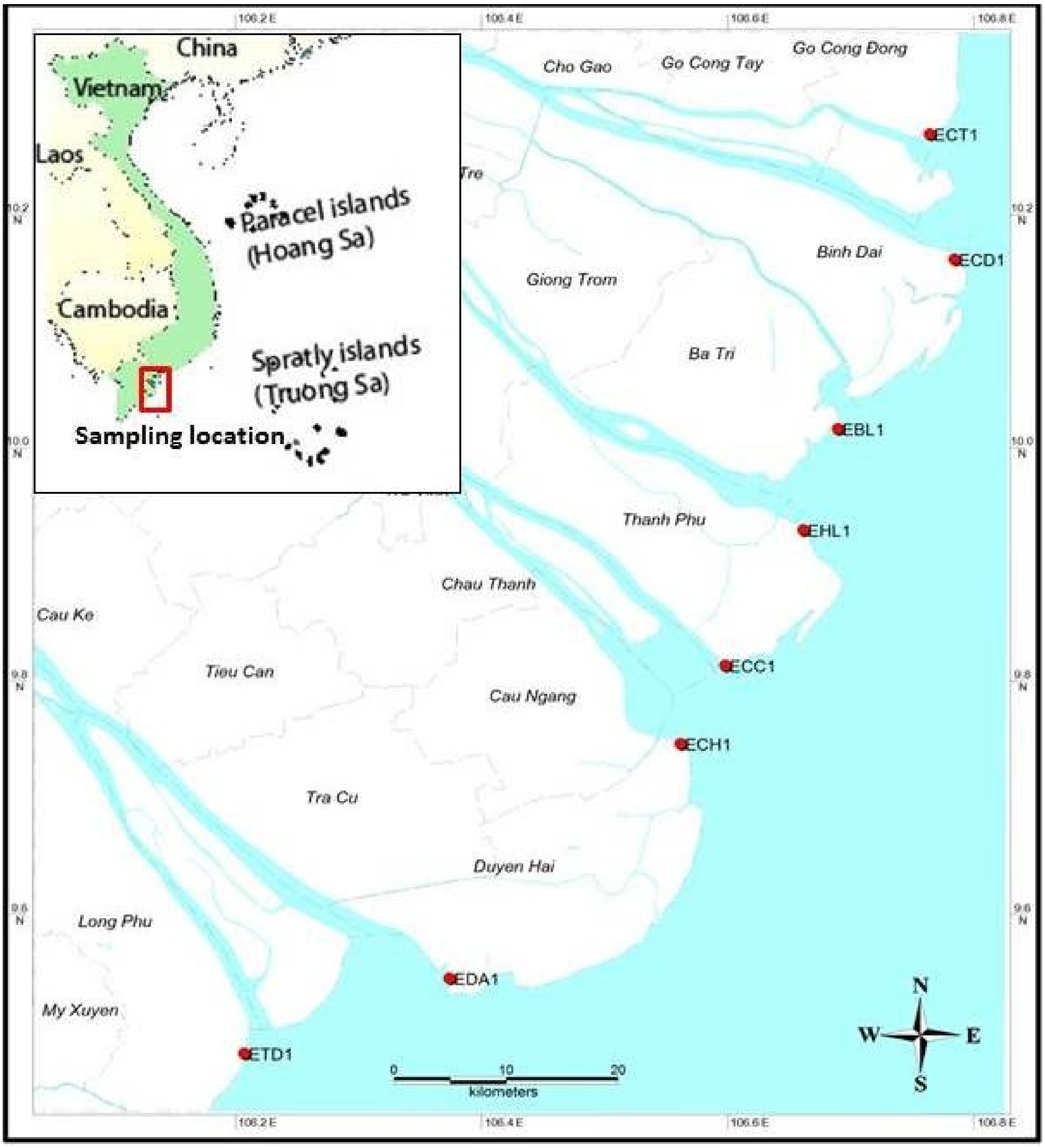

Figure 1. The sampling location at the mouths of 8 Mekong estuaries in the wet and dry seasons.

To detect significant differences between seasons, a two way - ANOVA analysis was applied after checking the homogeneity of variances with the Levene's test ($p > 0.05$). Data were transformed with $\log(x + 1)$ prior to analysis when assumptions were not fulfilled. Then, posthoc test (Tukey HSD) was applied for comparison in order to find significant differences between dry and wet season at each station.

To detect the significant differences between dry season and wet season in the case that 2 way ANOVA analysis could not be applied, (in case of MI, trophic structure and age structure) differences were tested between dry and wet season by applying the 2 way - PERMANOVA technique with factors season and estuary. The Euclidean distance was used as a resemblance measure. The main test was done to check the significant level of the interaction effect before pairwise test to explore which station exhibited significant differences between 2 seasons. The PERMANOVA+ is an add-on package to the PRIMER v.6 software programme.

3. RESULTS

3.1. Environmental variables in water column

Because of the tropical climate, the temperature did not change much between seasons. The temperature in the wet season ranged from 27.3 ^{o}C to 32.1 ^{o}C and in the dry season it did from 29.8 ^{o}C to 35.1 ^{o}C. Water dissolved oxygen in the wet season varied from 4.9 mg/l to 8 mg/l and in the dry season it did from 5.4 mg/l to 9 mg/l. The pH changed slightly between the 2 seasons since they varied from 6.83 to 7.8 in the wet season and from 7.4 to 8.62 in the dry season.

3.2. The grain size of the sediment

The percentage of sediment grain size at the 8 mouth estuarine stations in the dry season was expressed in figure 2. Almost all stations were consisting of 100 percentage of sand except ETD1 and especially ECC1.

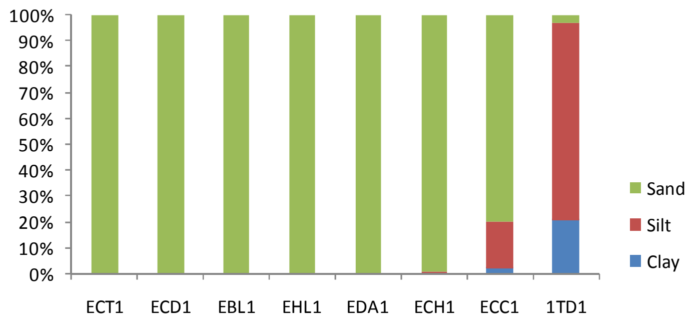

Figure 2. The percentage of sediment size classes at 8 mouth estuarine stations.

3.3. The densities of nematode communities in both seasons

The densities of the nematode communities in the dry season ranged from an average of 207 to 3111 ind./10cm^2, while in the wet season they ranged from an average of 454 to 3137 ind./10cm^2. The highest densities were found in station ETD1, for both seasons. ECT1 showed the lowest densities in the dry season but in the wet season, ECH1 had the lowest densities of all stations.

The densities of the nematode communities did not show a similar seasonal trend at all stations. In some stations, the densities of nematode are higher in the dry season compared to the wet season such as stations ECT1, ECD1, EBL1 and ETD1 while an opposite pattern is present in the remaining stations. In addition, the two way ANOVA analyses confirmed that there were no significant difference between the dry season and wet season ($F_{(1, 32)} = 0.1228$, $p = 0.728$), whereas stations differed significantly ($F_{(7, 32)} = 33.19$, $p < 0.0001$) and also the interaction effect was significant ($F_{(7, 32)} = 3.51$, $p = 0.0066$). The Tukey HSD test confirmed that not any of the pairwise comparison of seasons within the same station was different, whereas several stations especially ETD1 and EBL1 differed from the majority of other stations by their higher densities in both seasons.

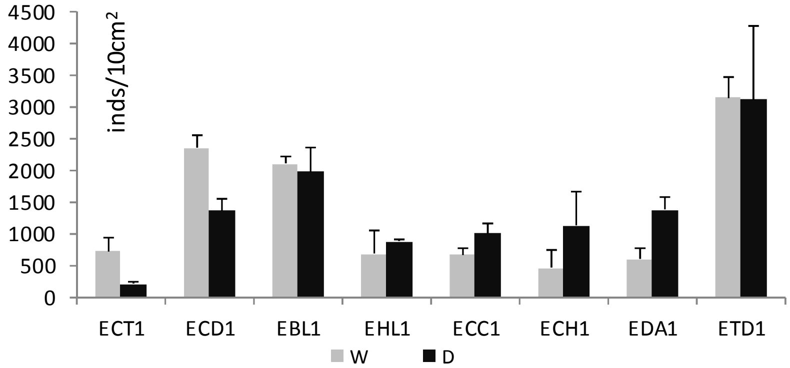

Figure 3. The average ± standard deviation of nematode densities (ind./10cm^2) in 2 seasons at the 8 mouth estuaries (W: wet season, D: dry season).

3.4. The diversity of nematode communities in both seasons

A total of 173 nematode genera were found at the mouth of the 8 estuaries covering both seasons, in which the dry season showed 137 genera and the wet season 134 genera. The Shannon - Wiener index at genus level showed values in the dry season that ranged from 2.445 to 4.18, while in the wet season they ranged between 1.829 and 4.00. Some stations like ECT1, ECD1, EHL1 and ECH1 have rather a large variation between 2 seasons, while the seasonal difference in these stations was not consistent since diversity increased or decreased depending on the station from one season to the other. According to the two way ANOVA analysis there is significant (although weak) difference between seasons ($F_{(1, 32)} = 4.356$, $p = 0.0449$), while also stations differed significantly ($F_{(7, 32)} = 9.419$, $p < 0.0001$) . There is also a significant interaction effect ($F_{(7, 32)} = 8.309$, $p < 0.0001$). Tukey HSD test for interaction effect showed that only 3 stations were significant different between dry and wet season such as ECT1 ($p = 0.02$), ECD1 ($p = 0.036$), EHL1 ($p = 0.03$).

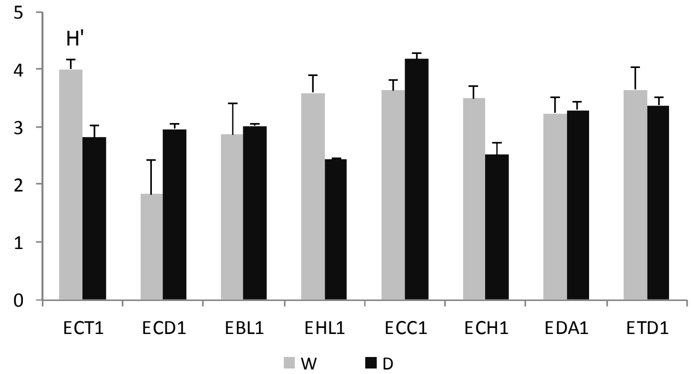

Figure 4. The average ± standard deviation of nematode diversity (Shannon-Wiener index) in dry season (D) and wet season (W).

3.5. The MI of nematode communities at 8 Mekong estuaries in both seasons

The Maturity index (MI) showed averaged values in the wet season ranging from 2.4 to 2.97 and from 2.3 to 3.2 in the dry season. There were some stations that showed a high fluctuation between the 2 seasons such as ECD1, ECH1, EHL1 and EDA1, and again the direction of fluctuation was not consistent in the same direction for all stations.

The PERMANOVA results showed there was a significant difference between stations (p= 0.0001) but there was no significant difference between seasons (p > 0.05). The interaction effect showed significant differences (p = 0.0001). The pair-wise test indicated that seasonal variation only occurred at station ECD1 (p = 0.0068) and EHL1 (p = 0.015).

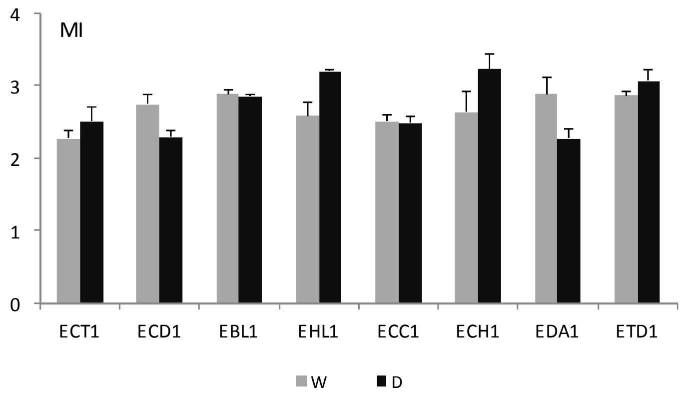

Figure 5. The average ± standard deviation of the Maturity Index of nematode communities following seasonal scale at 8 Mekong estuaries.

3.6. The trophic structure of nematode communities in both seasons

The structure of feeding types at each station did not show much difference between both seasons. The feeding type 2B showed the highest fluctuation at EHL1 and ECH1, where each time the group 2 B increased in the dry season. EDA1 showed an opposite trend for the same feeding type.

The 2 - way PERMANOVA test was also applied to test for significant differences between seasons and stations for the relative abundance of the 4 feeding types (table 1). The results showed that for all feeding types the interaction was significant. Table 1 shows which estuaries show a seasonal difference (pairwise test).

Table 1. The results of the 2 - way PERMANOVA test for percentage of feeding types between dry season and wet season at each station by factor: season, estuary, interaction between seasons and estuaries.

Feeding type	Factor	p- value	Seasonal change in stations (pairwise test)
%1A	Season	0.0013	ECD1 (p = 0.036), EHL1 (p = 0.0349), ECC1 (p = 0.003)
	Estuary	0.0001	
	Interaction	0.0023	

%1B	Season	0.0588	
	Estuary	0.0001	EBL1(p = 0.0076), ECC1(p = 0.0013)
	Interaction	0.0007	
%2A	Season	0.6136	
	Estuary	0.0001	EDA1 (p = 0.0349)
	Interaction	0.0012	
%2B	Season	0.1084	
	Estuary	0.0001	EHL1 (p = 0.0157), ECH1 (p = 0.0265), EDA1 (p = 0.0064)
	Interaction	0.0001	

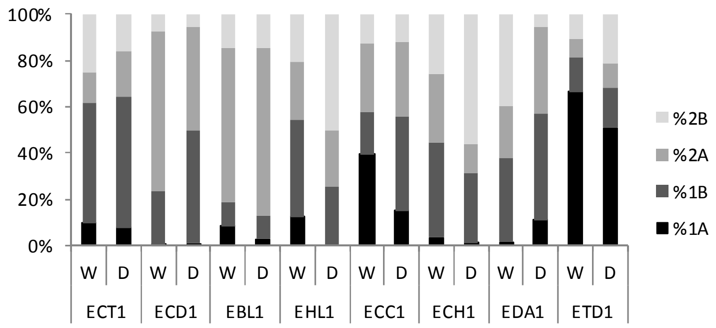

Figure 6. The percentage of feeding types 1A, 1B, 2A, 2B in the dry season (D) and wet season (W) at 8 Mekong estuaries.

3.7. Age structure of nematode communities in the 8 Mekong estuaries

Table 2. The results of the 2 - way PERMANOVA test for percentage of age structure between dry season and wet season at each station.

Ages	Factor	p- value	Seasonal change in stations (pairwise test)
% Juvenile	Season	0.0033	ECT1 (P = 0.0187), ECD1 (p = 0.0441), EBL1 (p = 0.0073), ETD1 (p = 0.0002)
	Estuary	0.0001	
	Interaction	0.0001	
% Female	Season	0.01	EDA1(p = 0.007), ETD1(p = 0.003)
	Estuary	0.001	
	Interaction	0.001	
% Male	Season	0.104	ECT1 (p = 0.0228), EBL1 (p = 0.0026), ETD1 (p = 0.004)
	Estuary	0.001	
	Interaction	0.001	

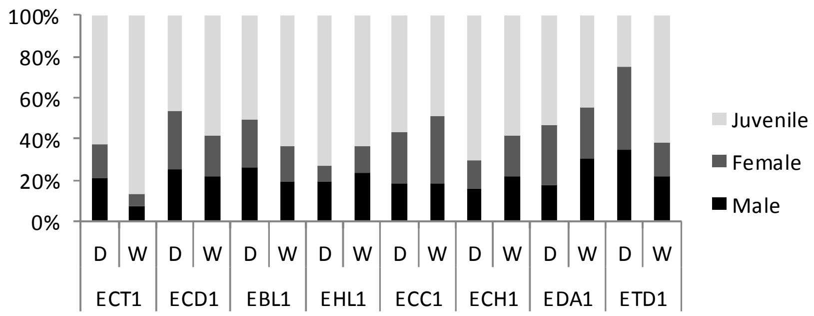

Figure 7. The average ± standard deviation of the percentage females (a) and males (b) and juvenile (c) nematodes in the dry season (D) and wet season (W).

In general, juveniles often appear in a dominant proportion (> 50 %) independent of the season. Station ETD1 is the main exception since juveniles are represented only by 25.2 % in the dry season and 61.9 % in the wet season. The adult individuals show in general an equal balance between males and females.

The 2-way PERMANOVA test was applied on the percentage of age structure between dry season and wet season at each station. The results (table 2) showed that for % juveniles, males and females the interaction between estuaries and seasons was significant. Table 2 shows which estuaries show a seasonal difference (pairwise test).

4. DISCUSSION

Almost all variables characterizing different aspects of the nematode communities at the 8 mouth estuarine stations such as densities, the maturity index, feeding types and age structure only differed in a few stations between the dry and the wet season. This was similar to the results by Hodda and Nicholas [23, 24] studying the temperate Hunter River estuary. They found that the densities and species compositions at the different sampling sites changed during the year but not following a consistent pattern across all sites. This pattern was attributed to the influence of “non-seasonal environmental changes” [9].

Hodda and Nicholas [25] further showed that the most important defect in the stability of k-dominance curves over the year and the changes of curves over different seasons seems mainly due to changes in the density of the most common and most opportunistic species. Furthermore, Hodda [23] also concluded that the variations among nematode communities in 3 Australian estuaries were due to other than seasonal related factors. In our study, results showed that significant differences for different nematode community characteristics was mostly present for the spatial comparison and for the interaction effect but not for the seasonal comparison. Some of the differences between both seasons is even possible due to a spatial patchiness not captured by three replicates from one season only.

According to Heip et al. [26] in a study on the benthic ecology of estuarine intertidal flats using the Westershelde as an example, one important conclusion from the analysis of spatio-temporal variance components by Ysebaert and Herman [27] on macrobenthos, was that a significant amount of variation occurs in the ‘station*year’ interaction factor. This is also the case in our study since a significant difference was mainly present in the case of the interaction effect between seasons and stations. It suggests that communities from different locations change differently with time.

In addition, the trophic structure of nematode communities in the Mekong was also not found different between dry and wet seasons but it was significantly different between stations. This was supported by studies of Hodda [23] and Hourston et al. [9] on tropical Australian estuaries, who suggested that free-living marine nematodes tend to be selective in the food they ingest. The presence of large amounts of a particular type of food at a locality would favour colonisation by nematodes that belong to a particular trophic group. Seasonal and other not food related environmental variables influence weakly the nematode communities according to the authors. However, this is opposite to studies done in temperate areas where densities clearly tend to be higher in the spring/summer [2] with changes in trophic group composition being attributed to coincidental seasonal changes in food resources [8, 26]. Also in subtropical estuaries studied by Ansari and Parulekar et al. [28] the nematode communities showed a substantial change in their dominant feeding type from season to season.

It was clear from our results that there were only weak or no changes between seasons in most stations whereas there are prominent alterations for the spatial scale for many aspects of the nematode communities. For all the measured variables of the nematode communities, only the Shannon – Wiener diversity index showed a weak significant difference between seasons. The dissimilarities that occurred between both seasons were mainly due to the composition and abundance of genera, especially the dominant genera.

These results were also similar to unpublished work of Nguyen [16] who compared nematode communities in the Can Gio mangrove forest between the dry and the wet season. The ANOSIM showed significant difference in seasonal variation but the Global R was only 0.394. The comparison also mentioned that the trophic structure was not significantly different between seasons but the biodiversity indices such as Shannon-Wiener, Margalef, and Hill were. The results of this study also indicated that differences between various stations were larger than between seasons.

Some studies [23, 29] also showed that spatial variation was larger than temporal variation like in the Mekong estuarine system. Alterations on a temporal scale may be caused by temperature or the relative abundances of the different types of food present as the most obvious factors explaining mainly density changes [9, 26]. Therefore, spatial variation was larger than the temporal variation in most stations because the spatial variation was driven by sediment composition which seems still the most important factor for community composition, whereas seasonal variation is driven by temperature, food and salinity which are secondary drivers for nematode community composition.

5. CONCLUSION

Therefore, we conclude that nematode communities were not influenced by seasonal factor in term of densities, maturity index, trophic structure and age structures except for the Shannon – Wiener diversity index. They were mainly controlled by sediment composition. Our results also indicated that spatial variation in these mouth estuarine stations is larger than seasonal factors due to tropical climate.

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TÓM TẮT

NGHIÊN CỨU SỰ BIẾN ĐỘNG CỦA QUẦN XÃ TUYẾN TRÙNG THEO MÙA VỤ

Ngô Xuân Quảng[1, *], Nguyễn Ngọc Châu[2], Ann Vanreusel[3]

[1]Phòng Công nghệ và Quản lý Môi trường, Viện Sinh học nhiệt đới, Viện HLKHCNVN, Số 85, Trần Quốc Toản, Quận 3, Tp. Hồ Chí Minh

[2]Phòng Tuyến trùng học, Viện Sinh thái và Tài nguyên Sinh vật, Viện HLKHCNVN, 18 Hoàng Quốc Việt, Cầu Giấy, Hà Nội

[3]Đại học Tổng hợp Ghent. Số 35, Ledeganckstraat, B-9000 Gent, Vương quốc Bỉ

[*]Email: ngoxuanq@gmail.com

Quần xã tuyến trùng ở 8 cửa sông Mekong đã được nghiên cứu trong mùa mưa và mùa khô nhằm đánh giá sự biến động và phản ứng của chúng đối với yếu tố mùa vụ. Các thông số về quần xã tuyến trùng như mật độ phân bố, chỉ số đa dạng sinh học, chỉ số sinh trưởng, cấu trúc dinh dưỡng và cấu trúc giới tính được đưa vào đánh giá và đối chiếu. Kết quả nghiên cứu cho thấy các thông số của quần xã tuyến trùng ở 8 điểm cửa sông Mekong không khác biệt theo mùa ngoại trừ chỉ số đa dạng sinh học Shannon – Wiener nhưng không đánh kể. Hơn nữa, kết quả nghiên cứu chứng minh rằng sự biến động theo không gian lớn hơn biến động về thời gian. Điều này bị chi phối chính bởi đặc điểm trầm tích.

Từ khóa: quần xã tuyến trùng, mùa vụ, cửa sông Mekong.