Intensive Coastal Shrimp Aquaculture Reduces Zooplankton Abundance and Diversity in the Aquaculture Ponds

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Abstract

In shrimp aquaculture systems, zooplankton represent a potential food source for larvae and juveniles due to its high nutritional value and size-suitability. Although many studies investigated zooplankton community in various aquaculture systems, little knowledge exists on how this varies among different culture systems. Here, we investigated how zooplankton abundance, diversity and density differ among three shrimp culture systems, namely extensive, semi-intensive and intensive. In total, 28 zooplankton species of 7 different groups were recorded. Copepods, rotifers and decapods were dominant groups, accounting for more than 80% of total zooplankton density. *Brachionus plicatilis* was the most abundant species. A key finding was that species richness and diversity were higher in extensive and semi-intensive culture than in intensive culture. Zooplankton density was highest (10^6 ind/m^3) in extensive systems which was 3 and 8 times higher than in intensive and semi-intensive systems, respectively. Density of zooplankton was lowest (4886 ind/m^3 in May) in the early stage of culture but notably higher in the later stage (8.9×10^5 ind/m^3 in June and 5.9×10^5 ind/m^3 in July). This is probably because the zooplankton community in the culture systems experienced a high predation pressure by cultured organisms during the early stage but were gradually less preyed upon over time. The obtained findings suggest that zooplankton assemblages in the ponds appeared to be an important food source for cultured organisms, especially during the early stage. It would be beneficial to establish an abundant assemblage of zooplankton in shrimp culture system prior to stocking.

Introduction

The aquaculture industry has been the fastest growing food production sector in many countries during the past decades given its potential economic and social values (Anand et al. 2019, FAO 2018). Aquaculture systems have been developed from small scale, low technological extensive to intensive rearing systems. Extensive culture systems completely rely on natural productivity from the biodiversity available in the culture systems, while in semi-intensive and intensive culture systems, additional inputs like feeding, environmental management, disease prevention and treatment are required (Hena and Hishamuddin 2014, Reis et al. 2020). According to FAO (2007), in semi-intensive and intensive systems, feed accounts for 40–80% of operational cost. In contrast, extensive systems require minimal inputs and depend mainly on natural diets for its production (FAO 2007).

Natural diets in aquaculture ponds like phytoplankton and zooplankton are rich in protein, vitamins, minerals, and others essential elements for the development of aquaculture species. They are important for various cultured species especially during the early stage of the culture (reviewed in Abualreesh 2021; Hena and Hishamuddin 2014). The availability and biomass of zooplankton in aquaculture systems can have significant contribution to its production. Thus, it is of crucial importance to have a better understanding of zooplankton community in these systems. Although many studies have examined the zooplankton community in various aquaculture systems (e.g. Coman et al. 2003; Gronning et al. 2019; Shil et al. 2013; Ghosh et al. 2011; Neto et al. 2009; Hena and Hishamuddin 2014), little is known about

how they differ among systems of difference levels of culture intensity such as extensive, semi-intensive and intensive.

In this study, extensive ponds were stocked with black tiger shrimp (*Penaeus monodon*) post larvae while semi-intensive and intensive ponds were stocked with white leg shrimp post larvae (*Litopenaeus vannamei*). Both species have been reported as effective zooplankton predators, especially during the early stage (Ling Lee Chen and Yung Chen 1992; Coman et al. 2006). The shrimp post larvae are still small and zooplankton are more attractive to them. At the later stage, when shrimp become larger, zooplankton may become too small to feed on and shrimp generally prefer to feed on larger epibenthic preys such as *Acetes spp.*, (Coman et al. 2003). Consequently, zooplankton contribution to shrimp diet, ultimately shrimp biomass is little (Coman et al. 2003).

Given the low stocking density in the extensive system, we assume the predation pressure imposed by shrimp larvae on the zooplankton community in this system is lower than in the semi-intensive and intensive system. Besides the difference in the stocking density, water in semi-intensive and intensive systems were exchanged on a regular basis while no water exchange was done in the extensive system (see Materials and Methods for more details). Water exchange frequency and level of disinfection caused by the use of chloride gradually increased from extensive to intensive. A higher water exchange frequency together with a stronger disinfecting water treatment in the intensive system may result in a stronger dilution effect on the zooplankton community in this system. Taking those differences together, we predict a higher zooplankton abundance, diversity, and density in the extensive system than in the other two systems, while being more notably when compared only to the intensive system.

On the other hand, regarding the changes of the zooplankton community over time, we hypothesize that the zooplankton community in semi-intensive and intensive systems will experience a higher predation stress at the early stage of the culture period than at the later stage. Therefore, zooplankton abundance and density may be lower at the beginning of the culture period compared to the later stage.

In this study, we investigated the abundance, species diversity, composition, and density of zooplankton in three commercial shrimp culture systems, namely Extensive, Semi-Intensive and Intensive during the culture period. In addition to examining the effect of the culture system on the zooplankton community, we also investigated how the zooplankton community in three culture systems changed over time during the culture period.

Materials And Methods

We studied how the zooplankton community differs among the three culture systems at different sampling moments. The differences among these culture systems are mainly characterized by stocking species, density and water exchange strategy.

Study site, zooplankton collection and species identification

The study was conducted from May to July 2020. Samples were collected from six shrimp ponds located in Tam Giang Lagoon, Thua Thien Hue in the central part of Vietnam. The studied ponds cover three different types of culture intensities practiced in the area, including extensive, semi-intensive and intensive with two ponds per level of culture intensity (hereafter called culture systems). Environmental characteristics of the ponds are typical for aquaculture ponds in the central part and southern Vietnam (e.g. Nguyen et al. 2011). The ponds were 1.2-1.5m deep. Bottom and bases of intensive ponds were coated with black polyethylene to prevent water loss. The area of the ponds was 2,500–7, 000m^2 for the extensive culture, 3,500m^2 for the semi-intensive culture and 2,000 m^2 for the intensive culture. Extensive ponds were stocked with black tiger shrimp (*Penaeus monodon*) post larvae at 50 ind/m^2. Semi-intensive and intensive ponds were stocked with white leg shrimp (*Litopenaeus vannamei*) post larvae at 50 ind/m^2 and 250 ind/m^2, respectively. During the study, no water exchange was done for extensive ponds. The ponds were only filled once a month to replenish the water loss. For semi-intensive ponds, 50% of the water was exchanged once every month. The water in intensive ponds were exchanged more frequently at one time per week during the first month, and every day thereafter from the second month of the culture period through siphoning and refilling.

In total, 18 zooplankton samples were collected from six shrimp ponds. All samples were collected using a conical plankton net (mesh size 90 µm, length 100cm, mouth/opening diameter 37cm) that was towed on surface by hand and collected at a distance of 20m long and at 5$m.s^{-1}$ speed. All samples were kept in 500mL bottles, preserved in 5% formalin (pH neutralized) and transferred to the laboratory at Hue University of Agriculture and Forestry for species identification and biomass determination. At the laboratory, all samples were cleaned with fresh water to remove dust. The samples were then filtered through a zooplankton sieve (250 µm) to obtain the macro-zooplankton which were then counted. Zooplankton smaller than 250 µm were retained on a 20 µm sieve and subsequently were brought into the suspension of filtered seawater added up to the volume of 50 mL. After thoroughly mixing, an 1 ml aliquot was used for counting under an MPC-1 binocular microscope. Individuals were morphologically identified to species level if possible, following the protocol developed by Grosjean et al. (2004), Balcer et al. (1984), Goswami (2004), Kelso et al. (2012).

The density of zooplankton species at each station was expressed as a count per cubic meter. To determine zooplankton density, the amount of water filtered by the plankton net during each sampling was estimated using the formula: Volume filtered water = Pulled distance × Opening area of net.

As a measure of biodiversity patterns, similarities in the species composition between stations were estimated by the Bray-Curtis index and stations were clustered based on this index. The species richness was calculated by the Margalef's index (d) (Margalef 1958):

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

Where S: Number of species, N: the total number of individuals.

The species diversity was estimated by Shannon Wiener index (H') (Shannon 1948) and Simpson index D (Odum 1971) :

Shannon Index (H'):

$$H' = -\sum_{i=1}^{S} p_i ln p_i$$

Where S: Number of species, pi is the frequency of the i^{th} species.

Simpson index (D):

$$D = \frac{1}{\sum_{i=1}^{s} p_i}$$

The species evenness was estimated by Pielous's index (Pielou 1966)

$$J' = \frac{H\prime}{H\prime_{max}}$$

Where: H': the number derived from the Shannon diversity index,

H'max: the maximum value of $H^{'}$.

$$H\prime_{max} = -\sum_{i-1}^{S} \frac{1}{S} ln \frac{1}{S} = lnS$$

Water quality

Water quality of the ponds were monitored at every sampling. Salinity and temperature were recorded using a hand-portable refractometer (PCE 0100, China) and thermometer (Netsuken, Japan) respectively.

Data analyses

All the analyses were conducted using R version 4.1.3 (R Core Team 2019) with the following packages: 'lme4' (v.1.1–21, Bates et al. 2015), 'dplyr' (v.1.0.8, Wickham et al. 2022), 'car' (v.3.0–12, Fox and Weisberg 2019), 'emmeans' (v.1.7.3, Lenth et al. 2022), 'multcomp (v.1.4–18, Hothorn et al. 2008) and 'rcompanion' (v.2.4.15, Mangiafico 2022). GraphPad Prism v.5 was used for making plots. PRIMER v.6 was used for analyzing of SIMPER (Similarity Percentages), and MDS (Multi-Dimensional Scaling), based on the matrix of the similarity in the species composition (Bray-Curtis 1957), species richness index D, species diversity index H and species evenness index J'.

Zooplankton density and number of species was log-transformed before being analyzed. The effect of intensity levels and sampling times was analyzed using Aligned Ranks Anova (Wobbrock at al. 2011)

with intensity level and sampling time as fixed- independent variables. We also compared the density of three major zooplankton groups including copepods, rotifers and decapod larvae among three different culture systems (Extensive, Semi-Intensive, Intensive) using Aligned Ranks Anova (Wobbrock at al. 2011).

Results

Water quality

Water temperature of the ponds during the study period was in range of 31–34^{o}C, with a mean of 32 ± 1.1^{o}C. There was no significant difference in temperature among the studied ponds during the study. For salinity, it was highest in Intensive ponds (mean: 33.8 ± 1.9‰), followed by Extensive ponds (mean: 19.8 ± 3.1‰), and lowest in semi-intensive ponds (mean: 16.3 ± 1.0‰). Salinity fluctuated less in semi-intensive ponds but was highly fluctuating in extensive and intensive ponds.

Zooplankton abundance and diversity

In total, 28 zooplankton species that belong to 7 groups (amphipoda, copepoda, hydromedusa, mysidacea, ostracoda, rotifera, and sergestidae) were observed from three sampling times from 3 types of shrimp culture systems. Among these groups, copepods were the most diverse group with 21 species recorded. The other 6 groups only accounted for 7 species. Among the 28 species, 5 species were unique to the extensive culture (*Canuella sp, Laophontella sp*, *Oithona rigida, Sarsia sp*, and *Mesopodopsis orientalis*) 7 species were unique to the semi-intensive culture (*Amphiascus inermis, Eudactylopus sp, Labidocera pavo, Paracalanus crassirostris, Oithona simplex, Oithona brevicornis*, and *Euterpina acutifrons*), and 4 species were unique to intensive culture (*Photis sp, Amphiascus sp, copepodite*, and *Subeucalanus subcrassus*). In terms of density, rotifers, copepods, and decapods were the three major groups, accounting for more than 80% of total zooplankton individuals.

Species richness index (d) was highest in semi-intensive culture, followed by extensive culture and lowest in intensive culture (Fig. 1a). Diversity index, represented from the Shannon index and Simpson index, was lower in the intensive system compared to extensive and semi-intensive systems (Fig. 1b & 1d). Similarly, the evenness index (Pielou) in extensive and semi-intensive systems was higher than in the intensive system (Fig. 1c).

The similarity index (Bray-Curtis) showed that the zooplankton population in the studied ponds is highly similar (45–50%, except pond intensive 1), especially in May with a similarity index of > 60% (Fig. 2). Among the ponds in three culture systems, semi-intensive and extensive ponds often had a high similarity index (65–70%). Within each culture system, similarity index between the ponds was in the range from 55–70% (Fig. 2).

Cumulative dominance indicated that the structure of zooplankton populations in all the ponds among the three culture systems was not balanced, with the total density of three major species accounting for >

80% of the total density in each population (Fig. 3). In particular, total density of *Brachionus plicatilis* and decapod larvae accounted for > 65% of total density in every pond.

Zooplankton composition and density in three different culture systems

There was no difference in the number of zooplankton species among the three culture intensities and between sampling times (Table 1, Fig. 4a). In terms of density, effects of culture system and sampling time are significantly (Table 1, Fig. 4b). Density was 3–8 times higher in extensive ponds (10^6 ind/m^3) compared to intensive (2.9×10^5 ind/m^3) and semi-intensive ponds (1.3×10^5 ind/m^3). Among three sampling times, density was lowest in May (4886 ind/m^3) but there was no difference between June (8.9×10^5 ind/m^3) and July (5.9×10^5 ind/m^3) ($P < 0.001$, Table 1, Fig. 4b). In intensive and extensive cultures, density was significantly higher in June and July compared to in May. For semi-intensive culture, there was no difference in density of zooplankton between three sampling moments ($P_{sampling\ time \times culture\ intensity} = 0.033$, Table 1).

Table 1
Results of Aligned Ranks ANOA test testing the effect of culture intensity and sampling time on zooplankton density and number of species

Aligned Ranks Anova	DF	SS	MS	P value
Number of species				
Sampling time (month)	2	0.11	0.06	0.290
Culture system	2	0.14	0.07	0.215
Interaction	4	0.15	0.04	0.482
Density				
Sampling time (month)	2	16.95	8.48	**< 0.001**
Culture system	2	2.48	1.24	*0.053*
Interaction	4	5.54	1.39	**0.033**

Density of major zooplankton groups

Both sampling time and culture system significantly affect copepod density. Highest copepods density was observed in June (28,093 ind/m^3), and lowest in May (589 ind/ m^3) ($P < 0.001$, Table 2, Fig. 5a). Among the three culture systems, copepod density was lowest in intensive culture (144 ind/m^3) ($P < 0.001$, Table 2, Fig. 5a), two magnitude orders lower than in extensive culture (27,299 ind/m^3) (Fig. 5a).

There was no significant difference between extensive and semi-intensive culture. For rotifers, there was no effect of sampling time on density (*P* = 0.229, Table 2). Rotifer was more abundant in intensive culture (364,037 ind/m^3) than in semi-intensive culture (15,125) (*P = 0.038*, Table 2, Fig. 5b). Decapod larvae density was lowest in May compared to June and July with no difference between the latter two sampling times (*P < 0.001*, Table 2, Fig. 5c). Among three culture intensities, decapod density was lowest in intensive culture and highest in extensive culture (*P < 0.001*, Table 2, Fig. 5c).

Table 2
Results of Aligned Ranks ANOVA test testing the effect of culture intensity and sampling time on density of copepods, rotifers and decapod larvae

Aligned Ranks Anova	DF	SS	MS	P value
Copepods				
Sampling time (month)	2	10.259	5.129	**< 0.001**
Culture intensity	2	26.981	13.49	**< 0.001**
Rotifers				
Sampling time (month)	2	2.63	1.31	0.229
Culture intensity	2	6.73	3.36	**0.038**
Decapods				
Sampling time (month)	2	30.615	15.308	**< 0.001**
Culture intensity	2	13.974	6.987	**< 0.001**

Discussion

Zooplankton composition and density

A total of 28 zooplankton species under 7 groups were recorded from six ponds of three culture systems. The most diverse group was copepods, which accounted for 75% of the total number of observed species (21/28) during the study. The other 6 groups accounted for 25%. This finding is consistent with previous studies on zooplankton diversity in shrimp ponds. For example, Shil et al (2013) found that the zooplankton community in semi-intensive shrimp *(Macrobrachium rosenbergii*) farm was dominated by copepods (54% of total species). Porchas-Cornejo et al (2013) reported that copepods were the most abundant groups and accounted for > 65% of zooplankton species in all studied ponds stocked with *Litopenaeus vannamei*. Similar results were also reported in Islam et al (2007), Ghosh et al (2011), and Preston et al (2003). Interestingly, some other studies found rotifers as the most diverse group (Alam et al. 1989; Ali et al. 1985; Mathias 1991). In terms of density, rotifers, copepods, and decapod larvae were the three dominant groups, which in total, accounted for > 80% of zooplankton density. This indicates that zooplankton abundance in shrimp ponds was mainly characterized by biomass of a few major groups.

A key novel finding was that species richness and diversity were higher in extensive and semi-intensive culture than in intensive culture. The high predation pressure in intensive system compared to that in the other two systems may pose a stronger controlling effect on the zooplankton community, not only on biomass but also species composition and diversity. In a review, the impact of top-down control (predation) on the zooplankton community is generally stronger in ecosystems with low species diversity than in those with higher species diversity (Daewel et al. 2014). In addition, the use of chloride in treating the incoming water before each water exchange could have significantly lowered zooplankton abundance and diversity in the refilling water in intensive culture.

Effect of culture system and sampling time on number of zooplankton species and density

During the study, no differences in the number of zooplankton species were found among three culture systems, regardless of sampling times. Copepods, rotifers and decapods are the dominant groups, in terms of species, in all culture systems. In previous studies, these groups together with cladoceras were found to be dominant in semi-intensive shrimp farms (Ghosh et al. 2011; Shil et al. 2013; Hena and Hishamuddin, 2014). Our results not only confirm this pattern of zooplankton in semi-intensive culture farms but also extend it to extensive and intensive culture systems.

Concerning density, with higher nutritional levels due to the use of fertilizer, artificial feeds and shrimp waste, semi-intensive and intensive culture systems are expected to support higher biomass of phytoplankton, subsequently higher density or biomass of zooplankton. However, density was highest in the extensive system, which was 4–8 times higher than in intensive and semi-intensive ponds. This result, while unexpected, could be partly explained by the differences in water exchange practice and the associated water treatment among the three culture systems. In extensive ponds, no water exchange was done during the study. The ponds were only refilled once per month to replenish for the water loss. In contrast, in semi-intensive ponds, 50% of the water was exchange once per month. The water in intensive ponds were daily replaced through siphoning and refilling. Furthermore, the incoming water in semi-intensive and extensive was disinfected by the use of chloride. These water exchange strategies could have reduced zooplankton abundance and diversity in semi-intensive and especially in intensive ponds. In a previous study of zooplankton in shrimp ponds in southern Vietnam, Grønning et al (2019) reported a rapid decline in abundance and biomass of copepods and other zooplankton caused by a sudden dilution of culture water. Another possible explanation is the difference in grazing pressure of shrimp larvae on zooplankton among the three culture systems. The stocking density in the intensive system is 5 times higher than in the extensive system. Finally, high nutrient level, low oxygen level especially on the bottom of the pond and high turbidity could be important factors in limiting biomass or even removing nutrient-sensitive species in intensive culture system.

In all three culture systems, density was lowest at the beginning of the culture period (in May) with an average of 4886 ind/m^3 and highest in June (8.9×10^5 ind/m3), with 2 magnitudes higher than in May. Density slightly declined in July (5.9×10^5 ind/m^3. In line with this observation, Hena and Hishamuddin

(2014) noted that zooplankton density was low at the start of the culture and increased during the culture period with a moderate decline in the last 3 weeks of the culture season. Similar results were also reported in other studies (Cardozo et al. 2007; Hena and Hishamuddin, 2014; Preston et al. 2003). The increase in zooplankton density during the culture season can be explained by the combination of zooplankton being released from the predation of culture organisms and the recruitment through reproduction over time. During the early stage of the culture period, the observed low density of zooplankton might be the result of being controlled by the predation effect of culture organisms and the limited availability of primary production of phytoplankton (Coman et al. 2003; Hena and Hishamuddin, 2014). For example, zooplankton density was reported to sharply decrease at the early stage of the culture after the introduction of *P. japonicus* post larvae into the culture system and then increased over time (Coman et al. 2003).

Effect of culture system and sampling time on abundance of the dominant zooplankton groups

To further explore the effect of culture system and sampling time, density of the dominant groups including copepods, rotifers and decapods were further analyzed. Density of copepods and especially decapods was higher in extensive and semi-intensive systems than in intensive culture with a maximum difference of two magnitude orders. This pattern was maintained throughout the culture period. The low stocking density in extensive and semi-intensive systems compared to the intensive system could be a reason, especially during the early stage of culture when culture animals are still small and have a high grazing effect on zooplankton.

Regarding the effect of culture time, density of copepods and decapods, it was low at the early stage but strongly increased during culture period, highest in June and slightly reduced in the later stage of the culture period. Similar to this, Grønning et al (2019) observed a low copepod density at the start of the production season. Density then steadily increased and reached a peak in the mid of the production season before slightly decreasing at the end of the production season. In the current study, during the peak, the density of copepods and decapods were two orders of magnitude higher than in the early stage. It is possible that the high grazing pressure during the early stage of culture organisms was reduced over time, resulting in high production of copepods and decapods in the later stage of the culture period. Interestingly, rotifer density was unlikely to be affected by culture systems as well as culture time. This indicates that the observed variation pattern of total zooplankton density among three culture systems was mainly driven by the variation of copepods and decapods but not rotifers. One possible explanation for this pattern is the prey size-selection behavior of the culture organisms. The small size of rotifers (mainly < 200 µm) compared to the large size of copepods (200–950 µm) (Grønning et al. 2019) and decapods (> 500 µm: Nguyen et al, unpublished data) makes them unlikely or less likely to be consumed by cultured organisms. Copepods are typically preferred by shrimp and most frequent (52–79% of samples) observed in the stomach of *Litopenaeus vannamei* (Porchas-Cornejo et al. 2013).

Conclusion

A better understanding of zooplankton community in aquaculture systems is essential for improving management practice and farming production. This study provided essential data for assessing the state of zooplankton community in different common aquaculture systems and at different culture stages. Our results suggest that at the beginning of the culture period with the introduction of shrimp larvae, zooplankton abundance in all three systems was low but gradually increased over time, possibly caused by the change of predation pressure over time. The observed findings suggest that zooplankton, particularly copepods and decapod larvae, contributed significantly to the nutrition of farmed shrimp, especially in the early stage of development. Thus, it would be beneficial to establish an abundant assemblage of zooplankton in shrimp culture system prior to stocking.

Declarations

The authors have no competing interests to declare that are relevant to the content of this article

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Author contribution

Anh Tuan Nguyen: Conceptualization, Methodology, Investigation, revision & editing. Ngoc Nguyen Tran: Conceptualization, Methodology, Investigation, Project administration. Tam T. Tran: Data analysis, writing – original draft. Duc Thanh Nguyen: Investigation, Trinh Si – Hai Truong: Methodology, Investigation.

Informed Consent Statement: Not applicable

Data Availability Statement: The datasets generated during and/or analysed during the current study are available from the corresponding author on reasonable request.

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Figures

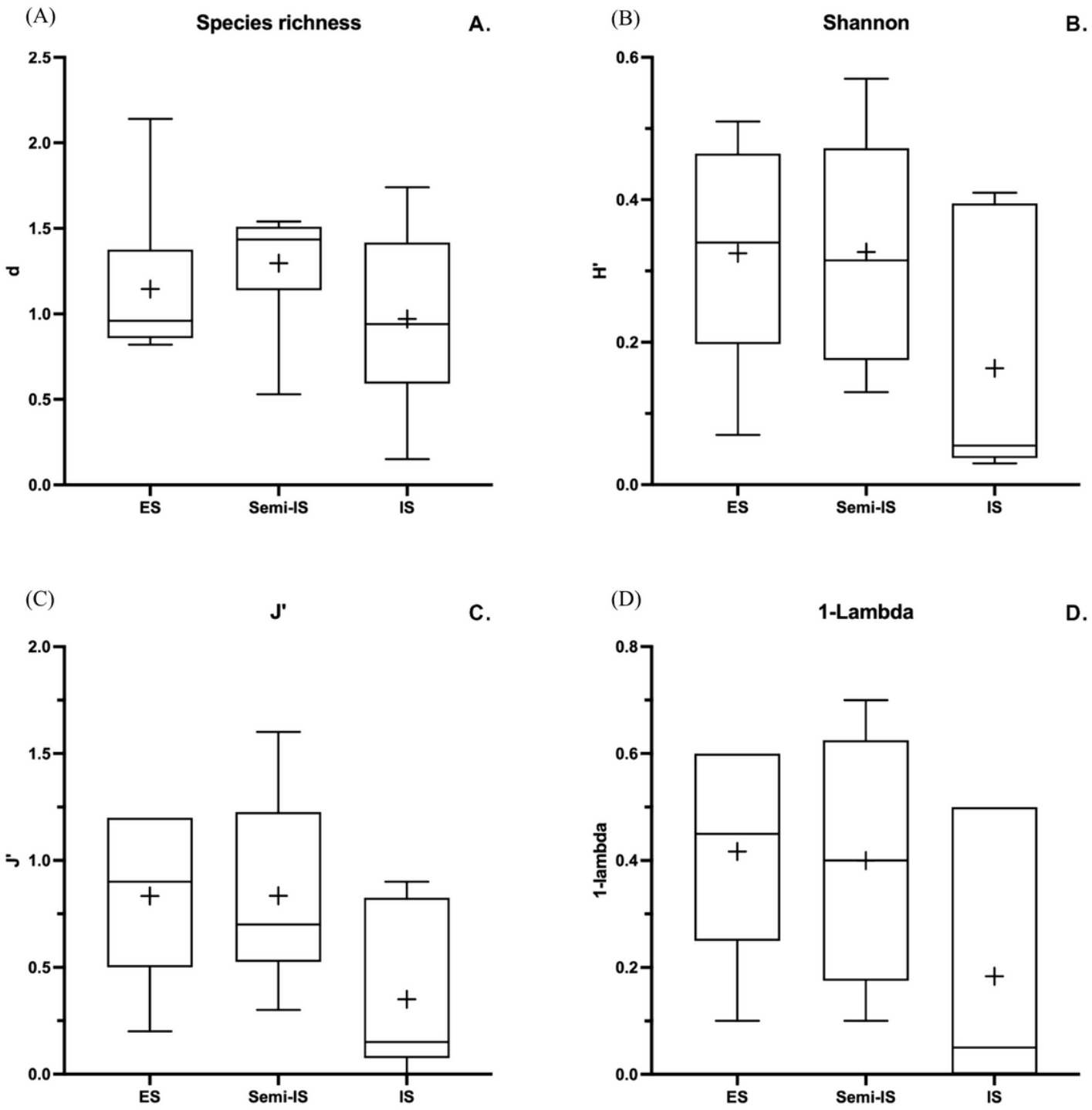

Figure 1

Biodiversity indices of zooplankton in three culture systems (ES: Extensive, Semi-IS: Semi-Intensive, IS: Intensive)

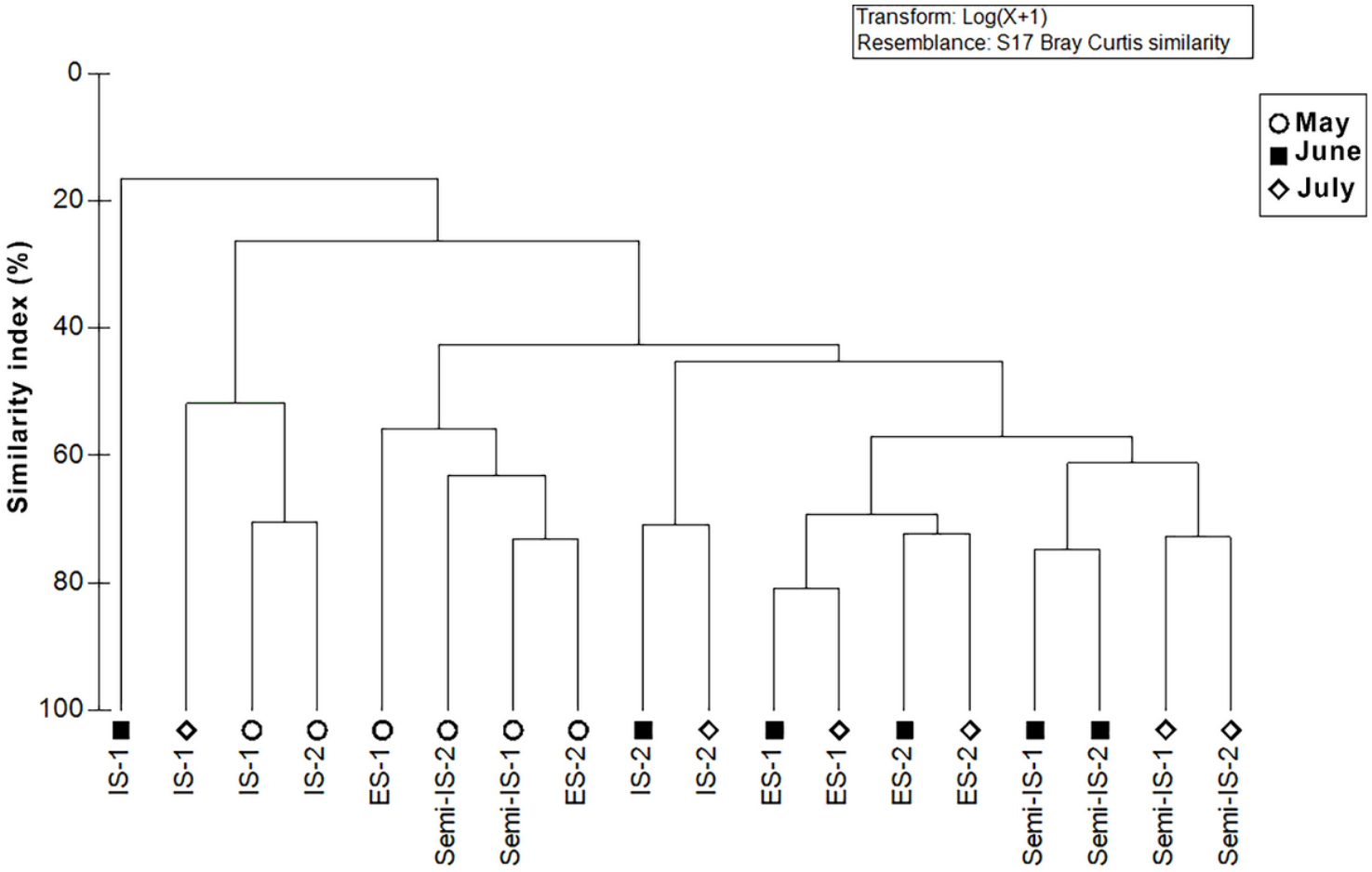

Figure 2

Bray-Curtis index on zooplankton population structure among different ponds in three culture systems (ES: Extensive, Semi-IS: Semi-Intensive, IS: Intensive) from May to July

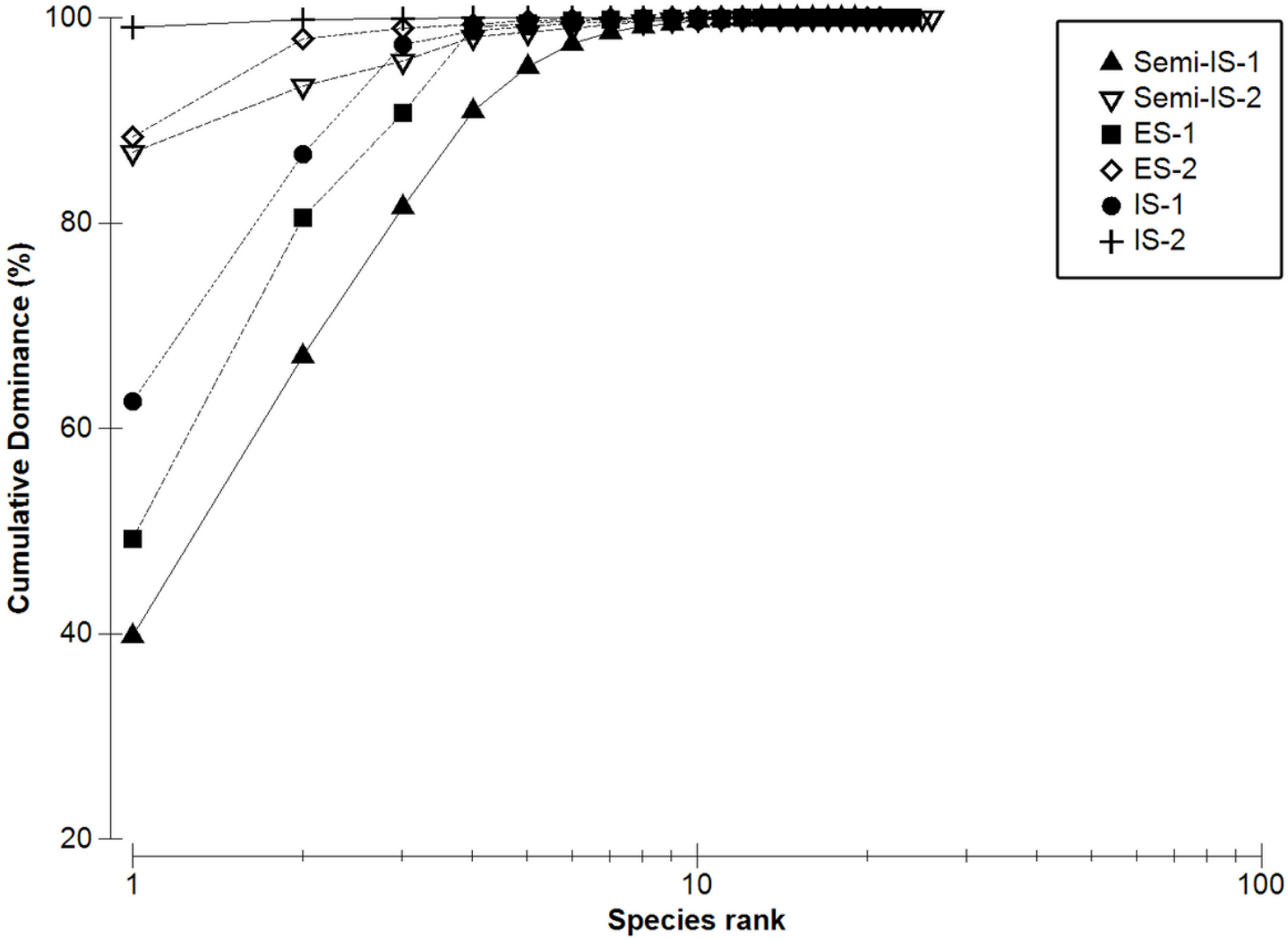

Figure 3

Cumulative dominance of zooplankton populations in six studied ponds

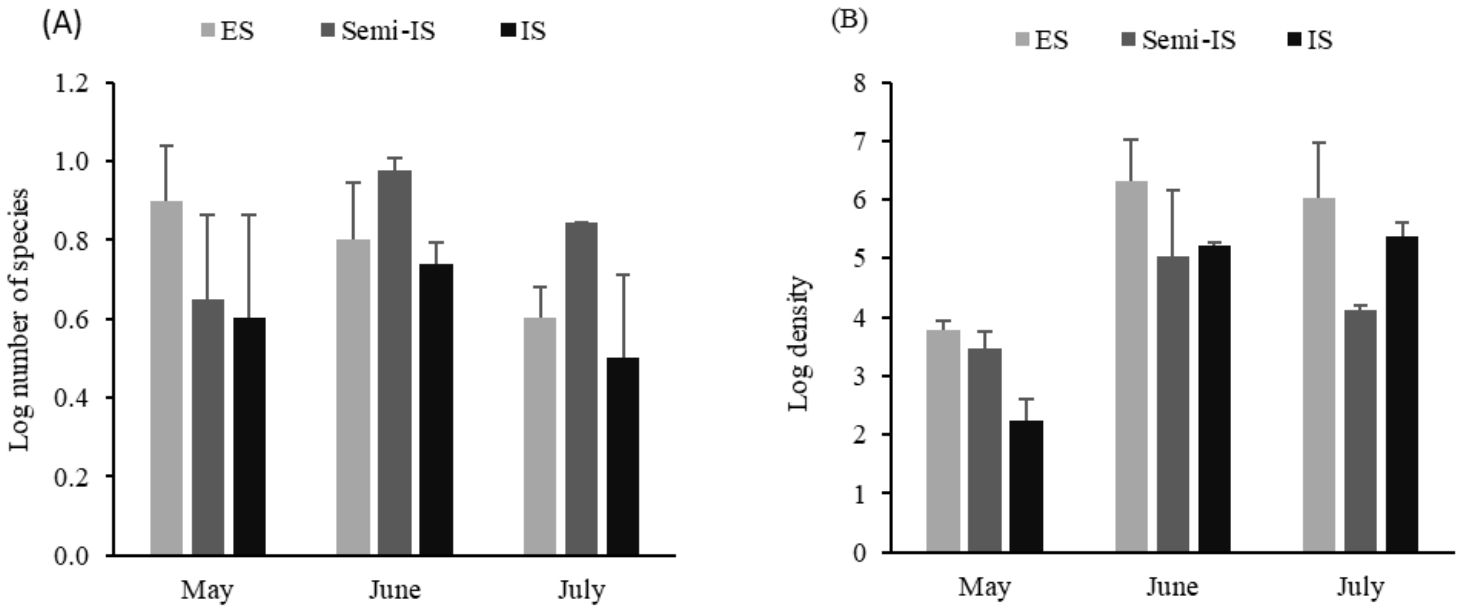

Figure 4

a: Log-number of species of zooplankton found at different sampling times (month) in three different culture systems; b: Log-density of all zooplankton species at different sampling times (month) in three different culture systems (ES: Extensive, Semi-IS: Semi-Intensive, IS: Intensive). Means are given with their standard errors

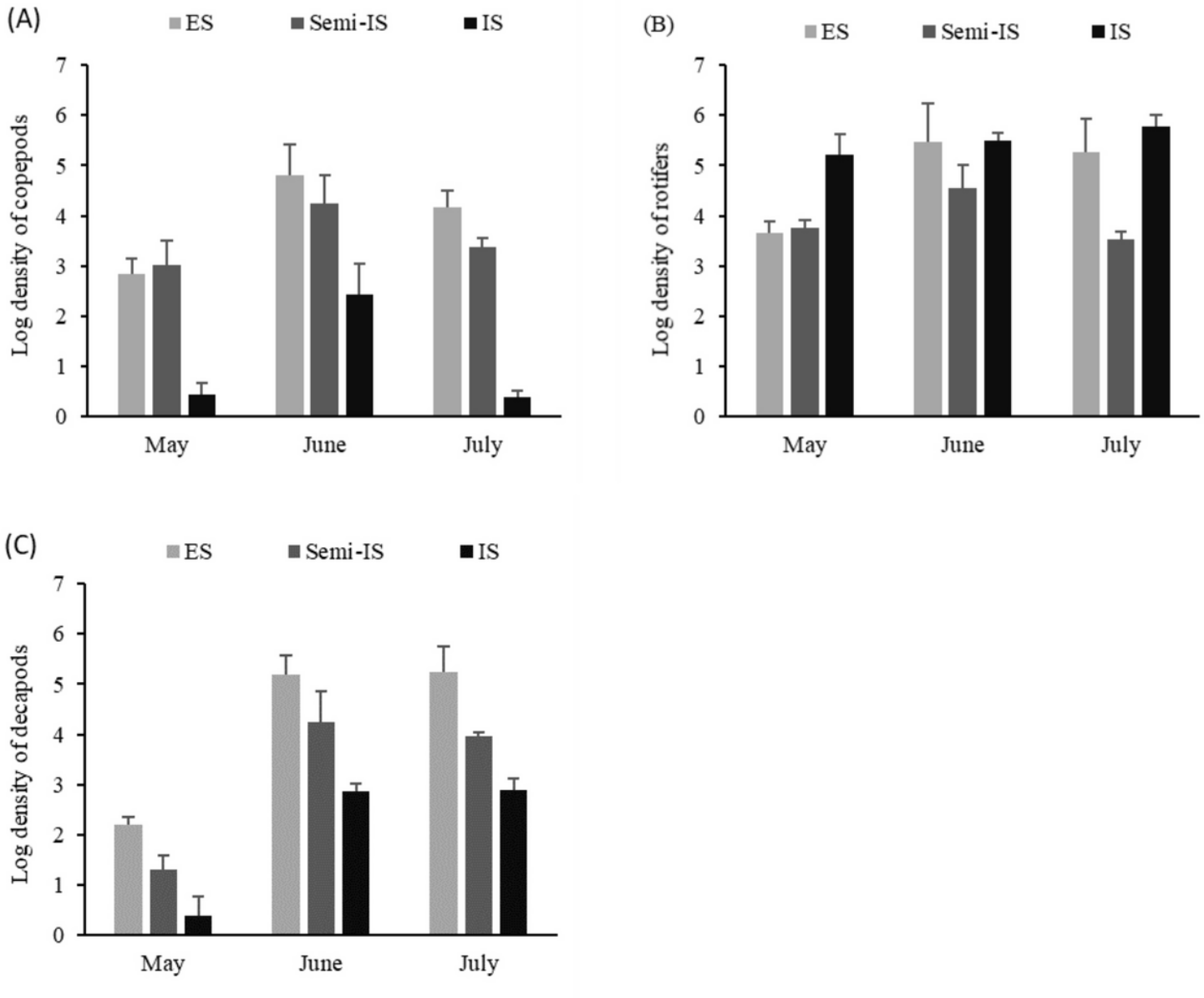

Figure 5

a: Log-density of copepods, b: Log-density of rotifers, c: Log-density of decapods at different sampling times (month) in three different culture systems (ES: Extensive, Semi-IS: Semi-Intensive, IS: Intensive). Means are given with their standard errors