

Applying zooplanktonic biological indices for assessment of water quality in the watersheds of Ray river, Baria-Vung Tau province

Tran Ngoc Diem My*, La Duong Song Nhi



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ABSTRACT

The Ray River is an important river system in Ba Ria-Vung Tau Province, Vietnam, supplying water for domestic, agricultural, and industrial purposes, thereby facilitating the province's economic development. The research was executed in 2024 across 9 watersheds, comprising 5 sites on the Ray River and 4 sites on the Ray River reservoir, throughout both the rainy and dry seasons. The findings documented 111 species of zooplankton, categorized into 50 genera, 13 orders, and 5 primary groups: Protozoa, Rotatoria, Cladocera, Copepoda, and Ostracoda. The research noted variations in zooplankton communities throughout the two seasons, suggesting saline intrusion at specific locations along the Ray River during the dry season. The Shannon-Wiener biodiversity score varied between 1.95 and 3.43, whereas the Pielou Evenness index consistently exceeded 0.8. The biological indicators of zooplankton communities indicate that most aquatic systems exhibited generally clean water quality, with only two locations demonstrating moderate pollution levels (α -mesosaprobe) during the dry season. The findings indicate that the aquatic environment in the Ray River system, encompassing both the river and reservoir, satisfies the water supply criteria for supplying domestic water and irrigation water in Ba Ria-Vung Tau Province. The study suggests utilizing zooplankton as an indicator for precise water quality scenarios, owing to their sensitivity to environmental fluctuations and brief life cycles. This would also offer a cost-effective approach for environmental monitoring and management of the river system.

Key words: zooplankton, water quality assessment, biological indices, Ray River, Ba Ria – Vung Tau province

1 INTRODUCTION

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2 The Ray River originates in Xuan Loc District (Dong
3 Nai Province) and traverses multiple regions of Ba
4 Ria-Vung Tau Province, spanning around 90 km and
5 constituting a significant transportation canal. The
6 Ray River Basin and Ray River Reservoir are vital
7 water sources in the Ba Ria-Vung Tau region, serv-
8 ing crucial functions in supplying water for home,
9 agricultural, and industrial purposes, while also safe-
10 guarding the surrounding natural environment. The
11 Ray River supports biodiversity through a diverse ar-
12 ray of plants and fauna. The regions next to the river
13 encompass vital natural ecosystems that contribute to
14 climate regulation and mitigate soil erosion.
15 The Ray River Reservoir, with a capacity of over 240
16 million m³, was established to store and manage water
17 for industrial, residential, and flood mitigation pur-
18 poses¹. The reservoir's water is processed to pro-
19 vide clean and safe drinking water to hundreds of
20 thousands of families in Ba Ria-Vung Tau, guaran-
21 teeing daily access to potable water. Agriculture in
22 this region is predominantly dependent on water from
23 the Ray River. Irrigation systems extract water from

24 rivers and reservoirs to irrigate rice fields, industrial
25 crops (including rubber, pepper, and cashew), and
26 fruit trees. In the dry season, the Ray River supplies
27 essential water to maintain agricultural productivity.
28 The Ray River Reservoir supplies water to significant
29 industrial zones in the region, including Phu My In-
30 dustrial Zone and My Xuan Industrial Zone. The
31 reservoir's water fulfills the operating requirements of
32 companies and businesses in the region, hence aid-
33 ing the economic advancement of Ba Ria-Vung Tau
34 Province.
35 Nonetheless, human activities, including water ex-
36 traction, industrial waste disposal, and routine prac-
37 tices, are causing the Ray River and its reservoir to
38 confront issues like water contamination and deterio-
39 rating ecological quality. Ensuring clean water and
40 safeguarding the aquatic ecosystem in the river and
41 reservoir is crucial for conserving biodiversity and
42 regulating the local climate.
43 Zooplankton are small organisms that drift in aquatic
44 environments; they possess the ability to swim but
45 are unable to navigate against the current. They are
46 extensively distributed over freshwater, brackish, and

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47 marine ecosystems, including both stagnant and flow-
 48 ing waters. Their body dimensions exhibit consid-
 49 erable variation, spanning from tens of micrometers
 50 (protozoa) to several millimeters (larger zooplank-
 51 ton). Zooplankton serves as an essential connection
 52 between primary producers and higher trophic lev-
 53 els in aquatic environments². Their distribution is
 54 contingent upon various parameters, like flow con-
 55 ditions, salinity, and nutrient availability³. Multiple
 56 studies underscore zooplankton as effective bioindi-
 57 cators, particularly in aquatic environments contam-
 58 inated by organic waste and heavy metals. Zooplank-
 59 ton can swiftly respond to environmental alterations,
 60 effectively indicating ecosystem health, with minimal
 61 monitoring expenses^{4,5}.

62 To comprehensively and precisely evaluate the con-
 63 taminants in water sources, it is essential to utilize
 64 biological markers in conjunction with physical and
 65 chemical indicators. Physical and chemical indicators
 66 provide data at the moment of measurement, but they
 67 rapidly fluctuate over time, thereby failing to accu-
 68 rately represent the true features of the water environ-
 69 ment. The organism's body will preserve these char-
 70 acteristics, which affect it at multiple levels. Presently,
 71 monitoring programs regard it as a constraint that
 72 nearly all periodic environmental assessments pre-
 73 dominantly emphasize physical and chemical indica-
 74 tors of water.

75 It is imperative to preserve biodiversity and regulate
 76 the local climate by ensuring the quality of water and
 77 the protection of the aquatic ecosystem in the Ray
 78 River system. This necessitates consistent environ-
 79 mental surveillance to promptly identify anomalous
 80 occurrences. The research aimed to evaluate the fea-
 81 sibility of employing zooplankton bioindicators for
 82 water quality evaluation in the Ray River basin sys-
 83 tem. This study was undertaken to establish a scien-
 84 tific foundation for zooplankton population structure
 85 and their potential as bioindicators within the river
 86 system.

87 **RESEARCH METHODS**

88 Samples were collected at 9 sites, including 5 sites on
 89 the Ray River Basin (SR1, SR2, SR3, SR4, SR5) and
 90 4 sites on the Ray River Reservoir (HSRay1, HSRay2,
 91 HSRay3, HSRay4), during two seasons: dry season
 92 (March 2024) and the rainy season (September 2024)
 93 (Figure 1).

94 Sampling locations in the Ray River Basin:
 95 SR1, SR2, SR3, SR4, and SR5: These points must be
 96 designated along the Ray River as it traverses distinct
 97 regions, commencing with Xuan Loc District in Đồng

Nai Province and proceeding through numerous sites
 in Ba Ria-Vung Tau Province.

98
 99
 100 Sampling locations at Ray River Reservoir: HSRay1,
 101 HSRay2, HSRay3, HSRay4: These points must be
 102 situated on the Ray River Reservoir, a vital water
 103 source for nearby residences, agriculture, and busi-
 104 ness. Qualitative and quantitative zooplankton sam-
 105 ples were obtained utilizing a Juday net featuring a
 106 mesh size of 45 μm, a mouth diameter of 0.4 m, and a
 107 net length of 0.9 m. The net was towed seven times at
 108 a velocity of 0.3 m/s, and the samples were transferred
 109 to 100-ml vials and preserved with 5% formalin. The
 110 identification of zooplankton species was based on
 111 the subsequent sources: The Rotifer Fauna of Wis-
 112 consin⁶; The Plankton of South Viet Nam: Fresh Wa-
 113 ter and Marine Plankton⁷; Rotatoria: Die Rädertiere
 114 Mitteleuropas by Max Voigt⁸; Fauna of Vietnam⁹;
 115 Freshwater Biology¹⁰; Free-Living Freshwater Pro-
 116 tozoa¹¹; Copepoda-Calanoida-Diaptomidae¹²; Zoo-
 117 plankton quantification was performed by enumer-
 118 ating organisms using a Sedgwick-Rafter counting
 119 chamber.

120 Biodiversity indicators, including the Shannon-
 121 Wiener diversity index (H') and the Pielou Evenness
 122 index (J')¹³, were employed to assess the variability,
 123 similarity, and stability of zooplankton populations
 124 at each sample location in relation to pollution levels.
 125 Table 1 juxtaposes the outcomes of the biological
 126 index analysis with evaluation scales derived from
 127 Staub et al. (1970) and Pielou (1966)^{14,15} (Table 2).

128 All water and zooplankton samples are analyzed at
 129 the Department of Ecology and Evolutionary Biol-
 130 ogy, Biology-Biotechnology Faculty, University of
 131 Sciences, Vietnam National University.

132 The research findings were computed and analyzed
 133 utilizing Excel 2013 and the Primer 6 software, em-
 134 ploying a 95% confidence interval to determine bi-
 135 ological indices. The t-test and ANOVA variance anal-
 136 ysis were performed using SPSS with the LSD method
 137 at a 95% confidence interval.

138 **RESULTS AND DISCUSSION**

139 **Zooplankton composition**

140 The examination of species composition in the Ray
 141 River system over two seasons identified 111 taxa
 142 across 50 genera, 13 families, and 5 groups: the Ro-
 143 tatoria group represented the highest proportion with
 144 45 species, constituting 40.54%, followed by the Pro-
 145 tozoa group with 34 species (30.63%), the Copepoda
 146 group with 17 species (15.32%), the Cladocera group
 147 with 12 species (10.81%), and the Ostracoda group,
 148 which had the lowest representation with 3 species,

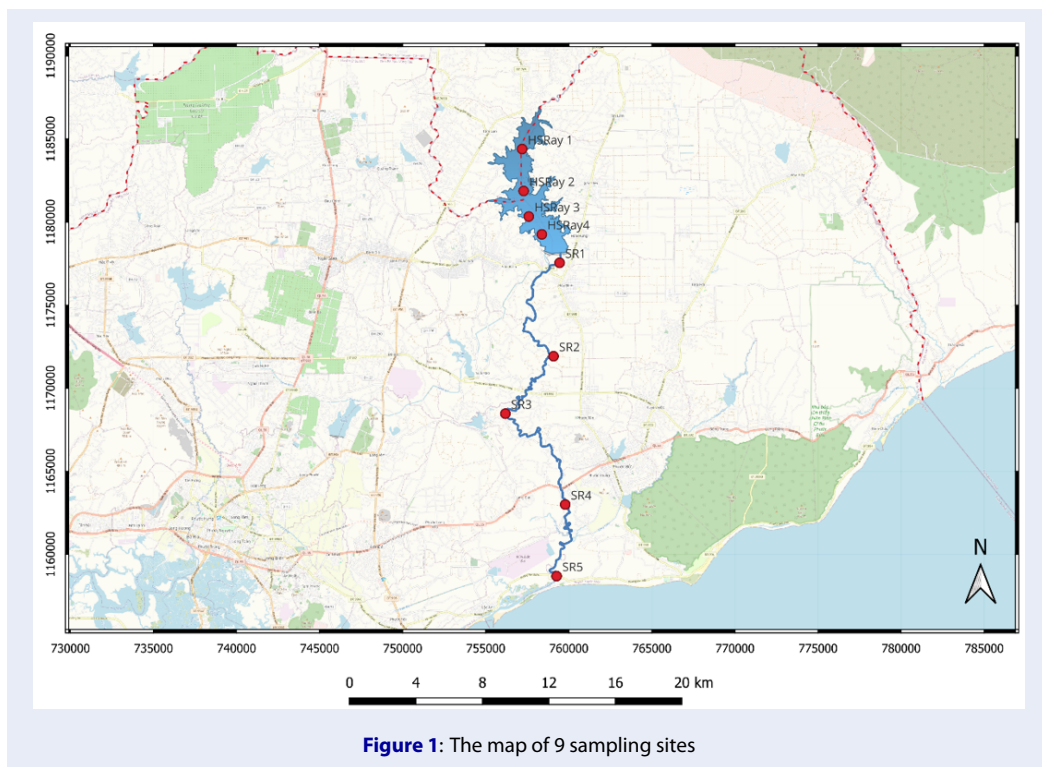


Figure 1: The map of 9 sampling sites

Table 1: Water quality assessment based on the Shannon-Wiener diversity index (H')¹⁶

Shannon-Wiener diversity index (H')	Water quality
< 1	High pollution (Polysaprobic)
1 - 2	Heaviest pollution (α -polysaprobic)
> 2 - 3	Moderate pollution (β -mesosaprobic)
> 3 - 4,5	Slight pollution (Oligosaprobic)
> 4-5	Pure water

Table 2: Scoring scale for assessing the sustainability of the zooplankton community corresponding to the level of pollution.¹⁴

Peilou index J'	Sustainable – Pollution scales
$J' > 0,8$	sustainable communities – oligosaprobic.
$0,6 < J' < 0,8$	less sustainable communities – Mesosaprobic / moderate pollution
$0,4 < J' < 0,6$	very less sustainable communities - Mesosaprobic α / heaviest pollution
$J' < 0,4$	unsustainable communities – Polysaprobic / high pollution

149 accounting for 2.7% (Figure 2). Of the 111 doc-
 150 umented species, 83 species from five groups were
 151 identified during the rainy season, with a notable pre-
 152 dominance in the Rotatoria group (44 species). Dur-
 153 ing the dry season, 74 species across 5 groups were
 154 documented, with the Protozoa group exhibiting the
 155 highest diversity at 28 species. The study identi-
 156 fied 46 species present in both seasons and detected
 157 Nauplius larvae from the Copepoda group at every
 158 sampling location in both seasons. The zooplankton
 159 findings demonstrated a notable increase relative to
 160 Nguyen Van Khoi's study¹⁷, which discovered merely
 161 36 species and revealed little richness, since numerous
 162 species were prevalent in coastal regions but absent in
 163 the river. The lack of common freshwater zooplank-
 164 ton species from the classes Cladocera, Chaetognata,
 165 Pteropoda, and Heteropoda significantly diminished
 166 the species mix. The Copepoda group comprised
 167 73.53% of the total zooplankton species.
 168 During the rainy season, there were more species at
 169 sample sites than during the dry season (Figure 3).
 170 This was likely because the water levels rose quickly
 171 and more nutrients from land ran off, which helped
 172 zooplankton grow ($p = 0.004$). In the rainy season,
 173 the Rotatoria group comprised more than 50% of the
 174 total species seen. During the dry season, elevated
 175 salinity promoted the survival and proliferation of the
 176 Protozoa and Copepoda groups. The Protozoa group
 177 exhibited an increase in species from 17 during the
 178 rainy season to 27 in the dry season, whereas the
 179 Copepoda group rose from 10 species to 13, includ-
 180 ing some species typical of brackish water habitats.
 181 This suggests that seasonal variables and salinity levels
 182 impact zooplankton at the sampling locations year-
 183 round. The results of this study align with those of
 184 research such as Zakaria (2007)¹⁸, Nguyen Thi Kim
 185 Lien (2013)¹⁹, and Nguyen Manh Hung (2003)²⁰, all
 186 of which observed that zooplankton exhibited sea-
 187 sonal variation impacted by salinity variables.
 188 The zooplankton groups during the rainy season in-
 189 dicated a substantial disparity in species composition,
 190 ranked as follows: Rotatoria > Copepoda > Protozoa
 191 = Cladocera > Ostracoda ($p = 0.000$). During the dry
 192 season, a statistically significant variation in species
 193 composition was observed among the categories, clas-
 194 sified as Protozoa = Rotatoria > Copepoda > Clado-
 195 cera > Ostracoda ($p = 0.0035$). The t-test indicated
 196 significant variations in species composition between
 197 the two seasons for the Rotatoria group ($p = 0.001$),
 198 Protozoa group ($p = 0.002$), and Copepoda group (p
 199 = 0.01). The other groups exhibited no notable differ-
 200 ences. The locations in the Ray River basin exhibited a
 201 significant variation in species composition between

the two seasons ($p = 0.008$), whereas the sites in the
 Ray River reservoir displayed no significant variations
 in species composition.

Density of zooplankton individuals

At the sampling sites, the density of zooplankton
 ranged from 12,100 to 117,625 individuals/m³. The
 individual density among groups also showed signifi-
 cant differences, ranked as Rotatoria = Copepoda >
 Protozoa > Cladocera = Ostracoda ($p=0.005$). No-
 tably, the individual density in the dry season was
 higher than in the rainy season ($p=0.008$).

During the dry season, the total density of individuals
 ranged from 16,906 to 117,625 individuals/m³, with
 the highest density observed at site SR5 and the low-
 est at site SR2. In the rainy season, the total density
 ranged from 12,100 to 43,196 individuals/m³, with
 the highest density at site HSRay 2 and the lowest at
 SR1. At the sites in the Ray River basin, there was a
 recorded difference in the total number of zooplank-
 ton individuals between the dry and rainy seasons,
 with individual counts increasing by 1 to 4 times com-
 pared to the rainy season ($p<0.05$). This phenomenon
 is explained by the intrusion of saline water at sites
 SR3, SR4, and SR5, along with low water flow, which
 led to the dominance of certain species in these areas.
 Meanwhile, the total density of individuals in the Ray
 River reservoir showed no significant differences be-
 tween the rainy and dry seasons.

The dominant species that thrive at the sampling
 points during the rainy season include Keratella trop-
 ica (SR1), Pompholyx complanata (SR2), Thermocy-
 clops hyalinus (SR3), Anuraeopsis fissa (SR4), Mi-
 crocyclops varicans (SR5), AT Nauplius (HSRay1,
 HSRay3, HSRay4), and Trichocerca pusilla (HSRay2).
 In the dry season, the dominant species in the wa-
 ter bodies include Nauplius larvae (SR1, SR3, SR4,
 SR5, HSRay1, HSRay2, HSRay4) and Keratella trop-
 ica (SR2, HSRay3). The density of these species in the
 water bodies accounts for 20–80% of the total indi-
 viduals. Most of the dominant plankton species are
 widely distributed ecologically; they are quite com-
 mon in natural water bodies and thrive while com-
 peting with other species. Research indicates that el-
 ements like pH and nutrient levels play a crucial role
 in the variations observed within zooplankton species
 groups. Species like Lepadella sp., Mesocyclops sp.,
 Polyarthra sp., and Brachionus sp. serve as bioindi-
 cators for nutrient-rich environments²¹. Species
 such as Phacus caudata, Brachionus spp., Keratella
 cochlaeris, Moina spp., Daphnia spp., Bosmina spp.,
 Cyclops spp., Mesocyclops spp., Chironomus larvae,

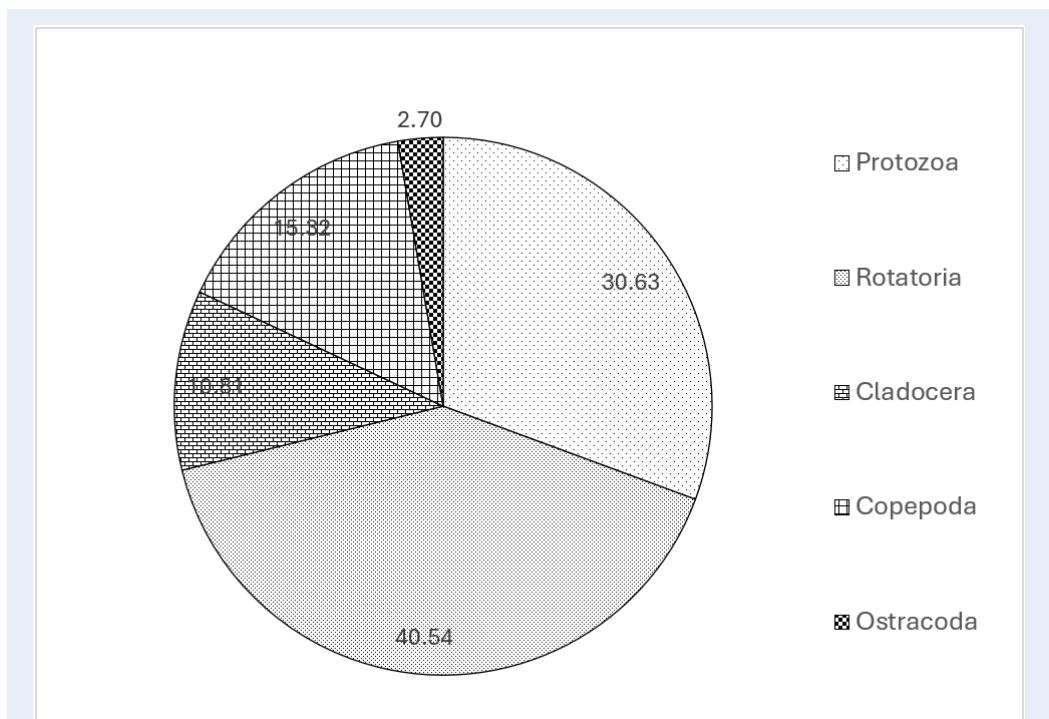


Figure 2: Structure of the zooplankton community in the sampling sites.

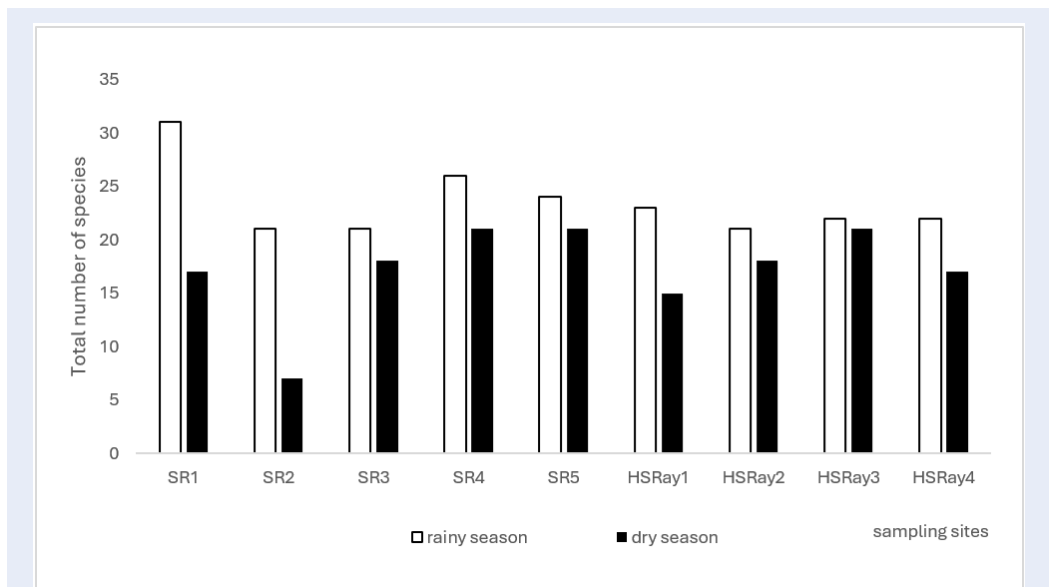


Figure 3: Total number of zooplankton species at the sampling sites during 2 seasons

Oxytricha, Eristalis tenax, and others have been documented as resilient to polluted water sources⁵. If the Rotatoria group is the dominant species in the water bodies during the rainy season, the Copepoda group shows a notable increase and dominance during the dry season, particularly in the flowing water bodies in the Ray River system. In the dry season, the increase in salinity causes the density of Copepoda plankton to be notably high because this group thrives in saline environments²².

Different water body types have also shown variations in the structure of zooplankton communities. In the Protozoa group, the species composition in flowing water is higher than in standing water ($p = 0.028$), but the individual density in standing water is higher than in flowing water ($p = 0.008$). Meanwhile, in the Cladocera group, both species composition and density in flowing water bodies are higher than in standing water bodies ($p = 0.004$). During the rainy season, the presence of Rotatoria is high in all water bodies in the Ray River system, ranging from 40 to 90%; however, during the dry season, the Copepoda group dominates in terms of individual numbers, particularly in the flowing water bodies of the Ray River basin (Figure 5).

Notably, the study found that species of brackish water lived along the Ray River (SR3, SR4, SR5) during the dry season. These species included *Acartia bispinosa*, *Acartia clausi*, *Acartia tsuensis*, *Clausocalanus furcatus*, *Paracalanus crassirostris*, and *Hemicyclops japonicus*. This is supported by data on salinity directly measured in the water bodies. Salinity in the dry season varies from 0.1 to 13.6‰, with significant measurements at SR3 (11.2‰), SR4 (13‰), and SR5 (13.6‰) (unpublished data). The findings correspond with earlier research identifying *Acartia clausi*, *Acartia centrura*, *Acartia danae*, *Acartiella sinensis*, *Paracalanus crassirostris*, and *Hemicyclops japonicus* as zooplankton species from brackish and saline aquatic habitats^{23,24}. This indicates a saline intrusion into the water bodies during the dry season, which is also an important point to consider in water resource management, as the Ray River system supplies water for agriculture and domestic use in the province.

The Shannon-Weiner diversity index (H') at the sampling locations varies from 1.95 to 3.43 (Figure 6). The H' index exhibits greater variability during the dry season compared to the rainy season. During the rainy season, environmental circumstances are more conducive to the growth and development of zooplankton, leading to increased species composition and a correspondingly elevated H' index relative to the dry season. Analysis of the H' index in conjunction

with the water quality evaluation in Table 1 indicates that water quality during both seasons varies from relatively clean to moderately polluted (α - meso-aprobe). No sampling point was evaluated as significantly contaminated according to the biological indicators of zooplankton. The Pielou Evenness index (J') varies from 0 to 1, with values nearing 1 signifying a more stable zooplankton community. The findings indicate that the J' index values span from 0.8 to 0.99 (Figure 7). The Pielou index indicates that all sampling locations in the Ray River system include stable zooplankton ecosystems with a minor degree of contamination.

The study measured COD content at sampling points, which ranged from 18 to 35 mg/L. This falls within the classification from A2 (suitable for domestic water supply with necessary treatment) to B1 (appropriate for irrigation) (unpublished data). The COD data indicates that the water quality in Ray River continues to meet the established usage standards. It is essential to integrate additional indicators to achieve more precise assessment outcomes. The research conducted by Nguyen Duong DT²⁵ determined that the water quality was satisfactory. Certain indicators, such as pH, ranged from 6.42 to 6.75, while the dissolved oxygen (DO) concentration exceeded 10 mg/L, surpassing the minimum threshold of 5 mg/L²². Parameters including turbidity, total suspended solids (TSS), chemical oxygen demand (COD), and heavy metal concentrations were all within low and safe thresholds. Following preliminary treatment, the majority of water quality parameters conformed to the permissible thresholds of the Standard of Vietnam for surface water (QCVN 08-MT:2015/BTNMT) requirements (A2 level), signifying that the water is appropriate for household consumption. The water quality of the Ray River is presently evaluated as satisfactory and stable, fulfilling the criteria for household and industrial use. In the study conducted by Phan TTT²⁶, the water quality findings revealed a trend of pollution, especially in the aquatic systems of the Ray River Reservoir and the segment of the Ray River near the sea¹⁷. This highlights the importance of evaluating water quality through diverse indicators, especially biological ones, which improve accuracy by demonstrating the adaptation of aquatic organisms, regardless of sampling time or place.

The Ray River system serves as a crucial water source, necessitating prompt recognition of rapid changes for the implementation of timely management and remedial actions. The physicochemical indicators measured solely represent the environmental circumstances at the time of sampling and do not provide a

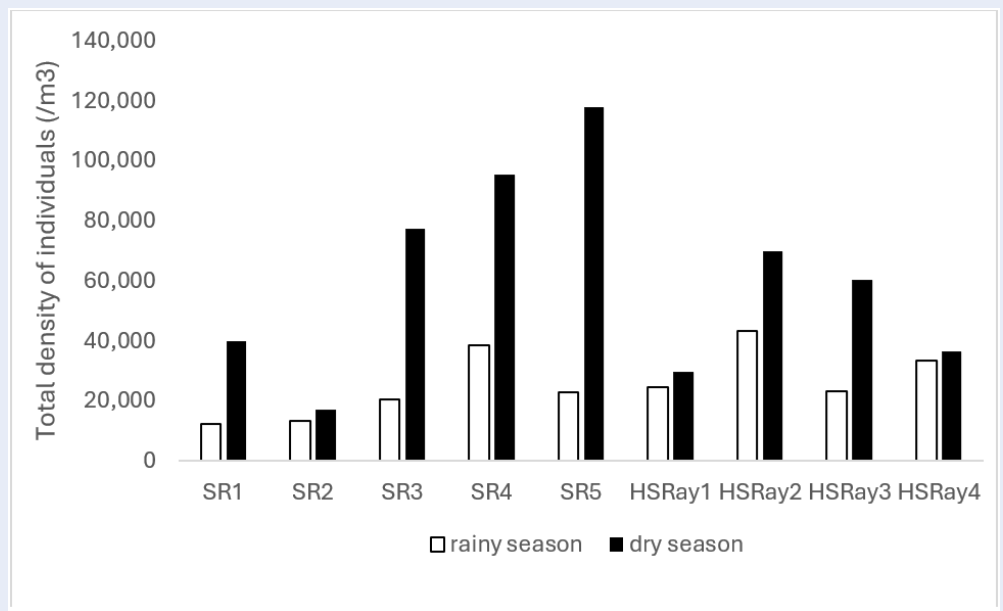


Figure 4: Total density of individuals at sampling sites during two seasons

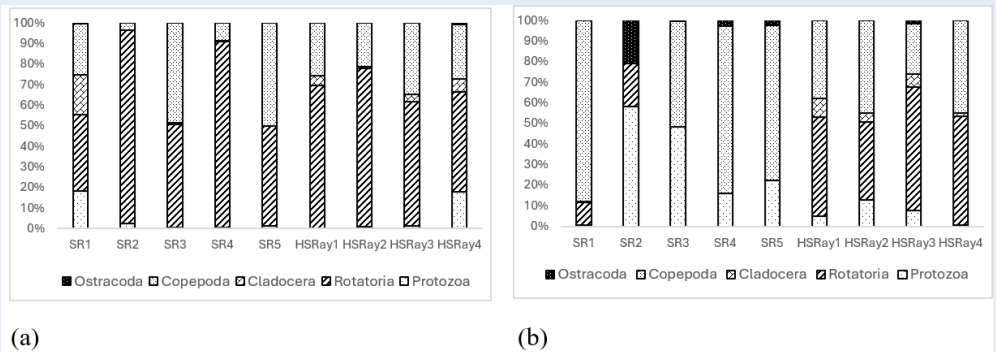


Figure 5: Variation in the structure of zooplankton communities at sampling sites based on the number of individuals (a: rainy season; b: dry season)

359 reliable evaluation of water quality. To improve pre-
 360 cision, many measurements are required, which may
 361 incur significant expenses. The results show that the
 362 biological indices of zooplankton can be effectively
 363 used as a tool for monitoring water quality in the Ray
 364 River. Environmental variables will influence species
 365 inhabiting aquatic settings, yielding a more precise
 366 depiction of water quality. Consequently, a compre-
 367 hensive evaluation of water quality in the examined
 368 region necessitates the integration of physicochemi-
 369 cal analysis and biological markers.

CONCLUSION

The study recorded 111 species among five zooplankton categories, with the Rotatoria group representing the predominant portion at 40.56%. Zooplankton density demonstrates more pronounced changes in the dry season than in the wet season. The analysis of the biological indicators H' and J' indicates that the water quality of samples from both seasons shows that the aquatic environment in the Ray River system is currently stable, meeting the standards for domestic water supply, as well as for agricultural and industrial applications. The Ray River functions as the primary water source for the water treatment facility in Long Phước commune, Ba Ria - Vung Tau province,

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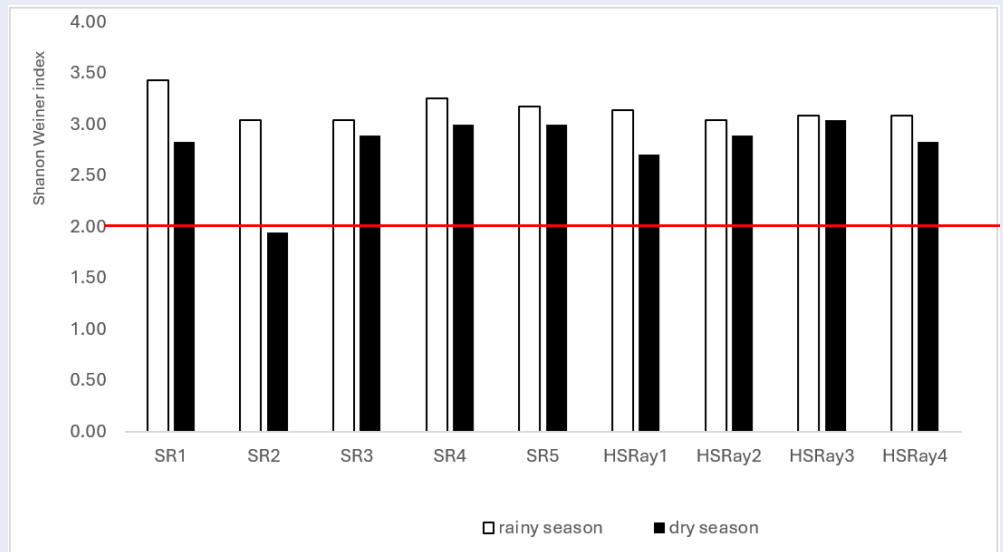


Figure 6: Shannon Weiner index at sampling sites during two seasons

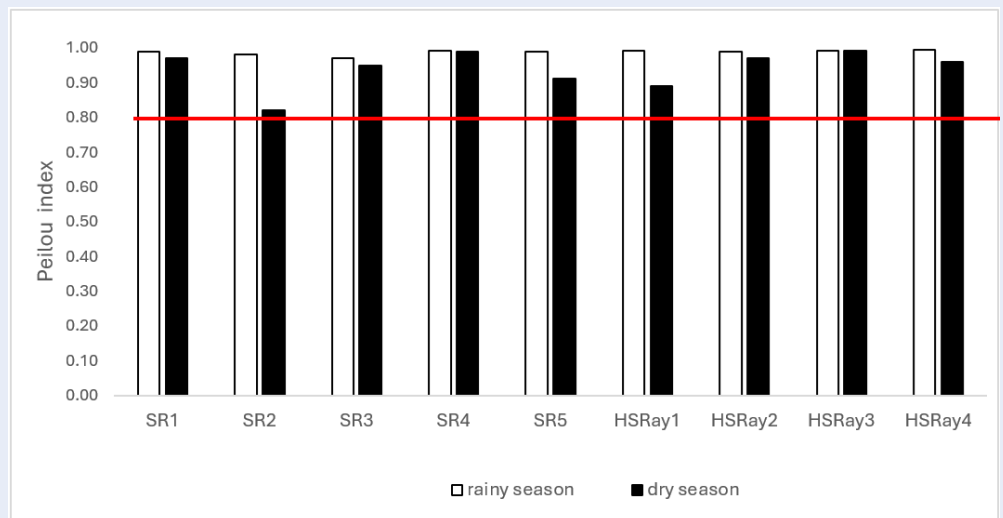


Figure 7: Peilou index at sampling sites during two seasons

384 demonstrating the dependability of this water supply
 385 for residential use. The results show that the biological
 386 indices of zooplankton can be effectively used as a
 387 tool for monitoring water quality in the Ray River.

388 However, climate change and economic development
 389 may jeopardize the stability of the water source in
 390 the future. Saline intrusion, prolonged drought, and
 391 over-extraction of groundwater may reduce the volume
 392 and quality of the Ray River. The study noted
 393 saltwater intrusion during the dry season at sampling
 394 sites along the Ray River, a problem that managers

395 must consider when developing sustainable management
 396 techniques to protect the river system's ecosystem
 397 and alleviate potential future challenges. It is essential
 398 to incorporate additional tools for monitoring
 399 water quality to effectively identify potential rapid
 400 changes in environmental management.

ABBREVIATIONS

XXXX

403 **ACKNOWLEDGMENTS**

404 XXXX

405 **AUTHOR’S CONTRIBUTIONS**

406 XXX

407 **FUNDING**

408 XXXX

409 **AVAILABILITY OF DATA AND**
410 **MATERIALS**

411 Data and materials used and/or analyzed during the
412 current study are available from the corresponding
413 author on reasonable request.

414 **ETHICS APPROVAL AND CONSENT**
415 **TO PARTICIPATE**

416 Not applicable.

417 **CONSENT FOR PUBLICATION**

418 Not applicable.

419 **COMPETING INTERESTS**

420 The authors declare that they have no competing in-
421 terests.

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