



Ecological Effect Evaluation of Artificial Reefs Based on Spatial Heterogeneity of Demersal Nekton Community in Beibu Gulf

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Authors' Contribution

ZL designed the research and wrote the manuscript. CPM, TZZ, YJ, CGB conducted the research and revised the manuscript. All authors read and approved the manuscript.

Key words

Artificial reef, Biodiversity evaluation, Density assessment, Nekton community, Spatial heterogeneity

ABSTRACT

To explore the ecological effects of artificial reef (AR), multidimensional evaluations involving density, biodiversity and spatial heterogeneity of nekton community were studied in Beibu Gulf China. The spatial pattern of nekton community was clearly classified into two groups (A and B) in April 2017. However, the variation of nekton densities between A (48529 ind/km²) and B (30220 ind/km²) was not significant ($P=0.17$). Therefore, the ecological effects evaluation of AR merely based on density assessment is not sensitive enough. Shannon-Wiener diversity (H') and Pielou evenness (J') in group A (2.74 and 0.78) were significantly higher than that in group B (2.3 and 0.7), implying an obvious ecological restoration effect of AR. Furthermore, crustaceans such as *Alpheus brevicristatus*, *Charybdis callianassa* and *Harpisquilla harpax*, showing a distinct preference tropism to AR, were turned out to be the main factors contributing to the spatial heterogeneity of nekton community between A and B, which was closely related to dissolved oxygen (DO), water transparency (Tra) and total inorganic nitrogen (TIN). The findings in this paper are significant for the protection and restoration of crustaceans resources along coastal water, given the rapid establish of AR around the world.

INTRODUCTION

Artificial reef (AR) is thoroughly considered to be an effective method to improve marine ecological environment, protect and restore coastal aquatic living resources (Relini *et al.*, 2007; Dupont, 2008). There have been a large amount of researches concerning the effect evaluation of AR for fishery resources enhancement and ecological restoration (Abelson, 2006; Seaman, 2007). Previous studies have concluded that AR produce up flows

that will transport nutrients from water bottom to upper layers, and increase the nutrients in water bodies, which leads to the multiply of phytoplankton, and eventually affects the abundance, diversity and community structure of zooplankton and nekton by the bottom-up effect (Graham *et al.*, 1992; Kim *et al.*, 2008; Champion *et al.*, 2015). As the superior consumers in the food chain, nekton community can reflect more information than other marine organisms. Therefore, studies based on characteristic of nekton community can be regarded as a comprehensive evaluation of AR ecological effects.

At present, researches on AR effectiveness are mainly focused on species composition and resources variations of fish. Many studies have shown that bait effect, flow field effect and refuge effect produced by AR have improved the breeding, growth and inhabiting condition for fish community (Relini *et al.*, 2002; Scarcella *et al.*, 2015). As reported by Rilov and Benayahu (2000), fish species and individuals in AR areas are generally more abundant

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than those in natural reef areas and its surrounding areas, which highlights the biological attractivity and residency duration of AR. Resources variation of marine organism was once believed to be the most intuitive presentation to the ecological effects of AR. However, the comprehensive ecological effects of AR cannot be merely reflected relying on biological resources dynamic. Therefore, it is urgent to explore other feasible ways to make up the deficiency of the current method mainly relying on resource assessment.

Community is an important structural and functional unit for achieving material cycle and energy transmission in marine ecosystems (Evans *et al.*, 1987). Studies based on community composition, structure feature and dynamic regularity can reflect the status and change trends of the ecosystem (Nicholson and Jennings, 2004; Schmölcke and Ritches, 2010). Therefore, researches based on nekton community are essential to explore the ecological effects of AR. Alpha diversity index as one of the most important indicators of biological community studies, is used to analyze the biodiversity variations among different time or space, and discuss the influence of human activities on biological community diversity and stability (Castillo-Rivera and Zavala-Hurtado, 2002). However, when a species in a nekton community is replaced by another one with similar ecological functions, such changes on community structure can seldom be reflected through α -diversity indexes (Beisel *et al.*, 2003; Pool *et al.*, 2015).

With further understanding of biodiversity, beta diversity has been gradually introduced into the field of AR. Beta diversity can reflect more information on biological community than α -diversity indexes (Anderson *et al.*, 2011). It not only describes species numbers and composition, but also considers the ecological niche relationship between different species. It is primarily used to seek and explain the biological response to environmental heterogeneity (Legendre *et al.*, 2005; Anderson *et al.*, 2013; Zeng *et al.*, 2017). Multivariate statistical analysis methods including hierarchical Cluster analysis (CLUSTER), non-metric multidimensional scaling analysis (NMDS), and redundancy analysis (RDA), are mathematical statistical methods that can reduce the complexity of the data matrix, extract the important information, and intuitively show the interrelationships among the variables. As a result, multivariate statistical analysis applying to biological community and ecosystem studies has gradually become a new research hotspot attracting the attention of scholars in the related fields (Godoy *et al.*, 2002; Brazner and Beals, 2011; Simonsen *et al.*, 2013).

ARs in Beibu Gulf have been established since 2009, the nekton community structure and its habitat are supposed to have been changed ever since. However, there have been no reports referring to the succession of nekton community

and their relationship with physicochemical environmental factors. Therefore, multidimensional evaluation methods are concerned in this research to find out the key biological and abiotic factors responding to the ecological effects of AR, based on the nekton community and physicochemical factors datasets collected in Beibu Gulf AR and its adjacent waters in April and September 2017.

MATERIALS AND METHODS

Study design

The study area was located in the AR and its adjacent area ($21^{\circ}19.419' \sim 21^{\circ}31.419'N$, $108^{\circ}9.054' \sim 108^{\circ}28.138'E$) of Beibu Gulf. Depth of the study area varied from 13.84 to 20.86 m according to acoustic detection. The specifications and models and distribution range of ARs were determined by sonar (Simrad EY60 and AquaScan Sensor, 200KHz) detection and underwater photography (Blue ROV2) before biological samplings, and the results were shown in Figure 1. There were five types of ARs founded in the study area, including three kind of benthonic reefs (height: 4 m, 5 m, 6 m, respectively) and two kind of suspended reefs (consist of 2 m benthonic reefs + 3 m suspended reefs and 2 m benthonic reefs + 4 m suspended reefs, respectively). As fishing was strictly forbidden in the South China Sea from May to September, the time of biological sampling was set in April 2017. Nine sampling sites were selected in the studied area (Fig. 1), 6 of which were located in the artificial reefs and its adjacent area (S1~S6) and 3 in the control area (S7~S9).

Data collection and processing

Nekton in the study area were collected by trawl sampling. The process of trawling lasted for nearly half an hour and the trawling speed was almost 2~3 knot at each sampling site. Width and height of trawl net were 16 m and 0.7 m, respectively. Due to the constraints of seabed topography and sea conditions, there were some differences in biological sampling intensity at each sampling site and the fishing data was unavailable at site S7 in September owing to the broken of sample net. In order to eliminate the influences of differential trawling duration and speed, the densities of nekton resources at each sampling site were standardized at 30 minutes by the mean speed of 3 knots.

Individuals obtained in each site were mainly classified into fish, cephalopods, decapod, crabs and squilloidea and identified to species in the field. Species that were difficult to identify were cryopreserved and brought back to the laboratory for further identification. All specimens were measured and weighed for nekton species less than 50 individuals, and species more than 50 individuals were sampled randomly referring to their size

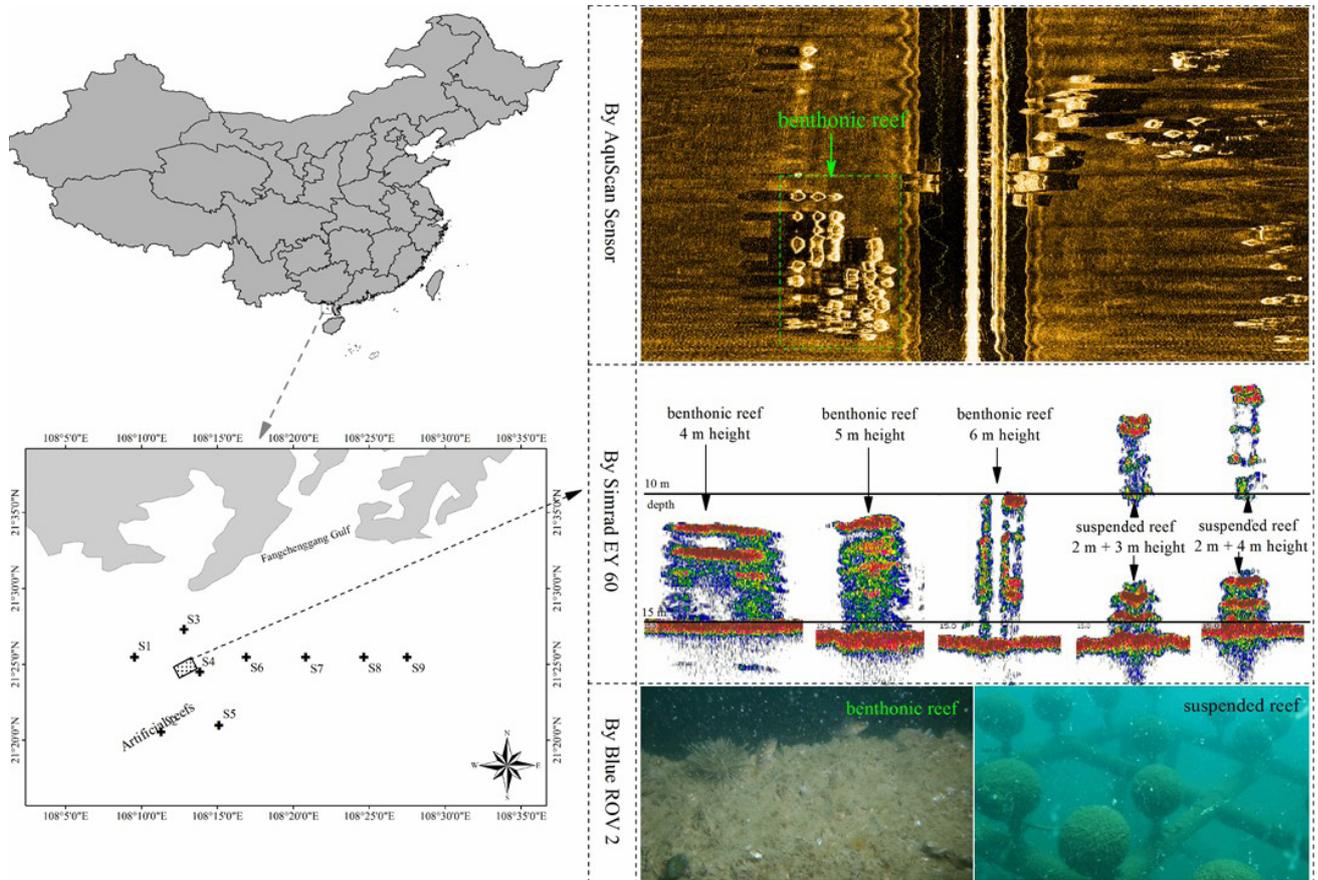


Fig. 1. The distribution of sample sites (S1~S9, left side) and artificial reefs (ARs) detected by different ways (right side).

proportion. The body length/fork length/mantle length and body weight of all specimens were measured accurately to 1 cm and 1 g, respectively.

These physicochemical parameters including water temperature (WT), dissolved oxygen (DO), salinity (Sal) and pH was tested by a calibrated YSI 6920V2-2 MPS probe in the bottom and surface water layer before trawling at each site. The determination of total inorganic nitrogen (TIN) and total phosphorus (TP) in each site were performed by the way of potassium persulfate oxidation in the laboratory. Water transparency (Tra) was measured using a 20 cm diameter Secchi disk with alternate black and white quadrants.

Data analysis

Spatial structure of nekton community

Nekton abundance from different sites were primarily transformed by $\log_{10}(x+1)$ to lessen the influence of prevalent species and increase the weight of rare species. Then, a Bray–Curtis similarity matrix based on nekton abundance of different sites was created for CLUSTER

and NMDS analysis which were used to explore the potential grouping structures (A, B, C etc.) and present an intuitive result among different sites with a stress value below 0.2. Furthermore, a SIMPROF permutation test was performed to confirm the clustering results statistically at 0.1% significance level. SIMPER analysis was conducted to assess the contribution rate of different species to similarities inner groups and dissimilarities between different groups. SIMPER analysis was a feasible way to find out the indicator species corresponding to habitat changes. Multivariate analysis on spatial structure of nekton community in the study area were performed by PRIMER 6 and PERMANOVA+.

Density and dominants

Nekton densities in each site were calculated according to the formula of $D = Y/A(1-E)$; Y means the total catches within the standardized time and speed (30 minutes at the average speed of 3 knots), A means the sweeping area within the standardized time and speed, E means the escape rate (0.5 in this research). Then, the

difference among different cluster groups (A, B, C, etc.) were tested by One-way ANOVA.

The dominants in different cluster groups (A, B, C, etc.) were evaluated by relative importance index (IRI) (Pinkas *et al.*, 1971), which was calculated as: $IRI = (N + W) \times F$; N was the quantity percentage of each species among the whole catches, W was the weight percentage of each species among the whole catches, and F was the occurrence frequency of each species in different cluster groups. Species with $IRI \geq 500$ were classified as dominants, $100 \leq IRI < 500$ were common species, $10 \leq IRI < 100$ were general species and $IRI < 10$ were rare species. Variation analysis were performed on the top five dominants of each cluster group.

Species similarity and biodiversity evaluation

Species similarity between different seasons and regions were evaluated by Jaccard coefficient (S_j) (Magurran, 1988). The calculation formula was defined as: $S_j = j/a+b-j$; j was the number of common species for different seasons and regions, a and b were the number of all nekton species in different surveys, respectively. The Jaccard coefficient indicates that nekton species in different surveys was very similar between 1.0 and 0.75, moderately similar between 0.75 and 0.5, moderately dissimilar between 0.5 and 0.25 and extremely dissimilar between 0.25 and 0.

The level of biodiversities in each sites were evaluated by Shannon-wiener diversity index H' and Pielou evenness index J' , which were defined as: $H' = -\sum_{i=1}^S p_i \ln p_i$ and $J' = H' / \ln S$; P_i was the quantity percentage of i species among all catches, and S was the number of species in each site. The spatial variation of biodiversity indexes among different cluster groups was tested by One-way ANOVA.

Relationship between nekton community and environmental factors

Linear model and unimodal model were usually used to reflect the response of species to successive environmental gradients. The linear model fitted better under the shorter environmental gradient, while the unimodal model was more suitable for the longer environmental gradient. The decision of model selection often depends on the analysis of the gradient length by detrending correspondence analysis (DCA). According to the result of DCA analysis, it was appropriate to choose the linear model (RDA), if the maximum value of the four-axis gradient was less than 3. Otherwise, it would be more suitable to select the unimodal model (CCA).

In this research, the maximum value of the four-axis gradient was 2.578 according to the DCA result. Therefore, the relationship between nekton community

and environmental factors was analyzed by redundancy analysis (RDA). Nekton variables were $\log_{10}(x+1)$ transformed firstly and the general and rare nekton species ($IRI < 100$) were excluded to reduce the weight of extreme values. Centralization treatment was necessary for nekton community data, for which may have great influence on RDA results. Environmental variables were also $\log_{10}(x+1)$ transformed to achieve approximate normal distributions. Furthermore, the partial Monte Carlo permutation test was used to evaluate the contribution of each alternative environmental variable to the interpretation of species variables. The environmental variables with remarkably higher interpretation values were usually regarded as the abiological indicators of habitat change. Analysis on relationship between nekton community and environmental factors were carried out through Canoco 4.5.

RESULTS

Temporal variations of nekton community between April and September

A total of 161 nekton species were collected in the study area in April and September 2017, including 101 species of fish, 27 species of crabs, 16 species of decapod, 9 species of squilloidea, and 8 species of cephalopods. The numbers of nekton species in April and September were 92 and 100 respectively, the number of fish species accounted for a relatively higher proportion in both surveys (Table I). However, the species similarity of different months was only 0.19 according to Jaccard coefficient, showing an obvious seasonal replacement feature. According to the results of IRI, ten dominants ($IRI \geq 500$) were selected from the nekton community both in the April and September respectively and part of them (*Psenopsis anomala*, *Alpheus brevicristatus*, *Saurida tumbil*, *Charybdis callianassa*, *Trachurus japonicus*, *et al.*) showed significant seasonal specificity (Table II).

Table I. The number of nekton species in the study area.

	April			September
	Group A	Group B	Total	
Fish	49	23	56	62
Crab	16	9	18	15
Decapod	4	4	8	10
Squilloidea	3	4	5	9
Cephalopoda	4	3	5	4
All species	76	43	92	100

Spatial structure of nekton community in the studied area

The spatial heterogeneity of nekton community was only observed in April, in which time the study area was

clearly divided into group A (S1-S6) and B (S7-S9) based on CLUSTER (Fig. 2) and NMDS (stress = 0.01) analysis, which separately represented the ecological effect region of AR and the control region. The spatial heterogeneity of nekton community in the study area was further confirmed by SIMPROF permutation test ($P_i = 7.08, P = 0.1\%$).

Table II. Dominants composition of nekton species in the study area.

	April			Sep- tember
	Group A	Group B	Total	
<i>Psenopsis anomala</i>	+++	+++	+++	--
<i>Alpheus brevicristatus</i>	+++	--	+++	--
<i>Orotosquilla interrupta</i>	--	--	--	+++
<i>Saurida tumbil</i>	++	+++	+++	--
<i>Leiognathus bindus</i>	+++	--	++	--
<i>Upeneus sulphureus</i>	--	--	--	+++
<i>Charybdis callianassa</i>	+++	--	+++	--
<i>Platycephalus indicus</i>	--	--	--	+++
<i>Orotosquilla nepa</i>	+++	--	+++	+++
<i>Trachurus japonicus</i>	++	+++	+++	--
<i>Leiognathus brevirostris</i>	++	++	++	+++
<i>Parargyrops edita</i>	+++	+++	+++	+
<i>Nemipterus virgatus</i>	+++	+++	+++	+
<i>Metapenaeus affinis</i>	--	+	+	+++
<i>Orotosquilla oratoria</i>	+++	+	++	+++
<i>Harpisquilla harpax</i>	+++	+	+++	+++
<i>Nemipterus japonicus</i>	+++	+	++	+
<i>Lactarius lactarius</i>	+	--	+	+++
<i>Leiognathus berbis</i>	+++	+++	+++	+++
<i>Charybdis truncata</i>	+	+++	++	+

*Note: +++, dominant species ($IRI \geq 500$); ++, common species ($100 < IRI < 500$); +, general and rare species ($IRI < 100$); --, absent species ($IRI = 0$).

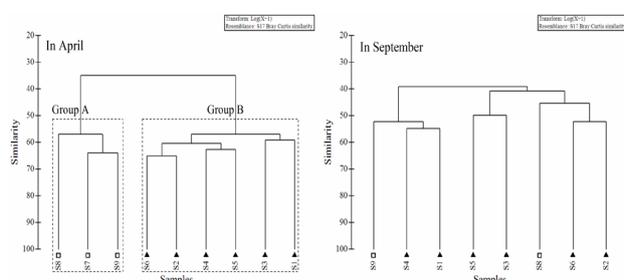


Fig. 2. Spatial pattern of nekton community in the study area based on CLUSTER analysis (Group A: artificial reefs ecological effect region; Group B: control region).

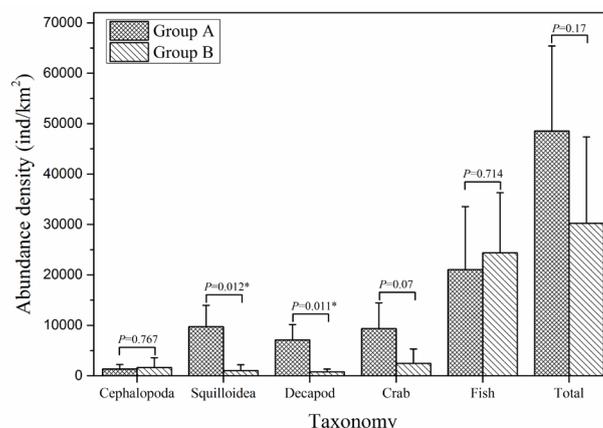


Fig. 3. Spatial variation of nekton densities (mean±SD, ind/km²) between group A and B.

In April, the number of nekton species in group A was 76, dramatically greater than that of 43 in group B (Table I). The species similarity between the two groups (A and B) was 0.29, at the moderately dissimilar level according to Jaccard coefficient. There were ten dominants in the study area in April, and four of them (*Alpheus brevicristatus*, *Leiognathus bindus*, *Charybdis callianassa* and *Orotosquilla nepa*) showed obvious geographical selectivity on AR area (Table II).

The average densities of the whole nekton species in group A and B were 48529 and 30220 ind/km² respectively, which was not significantly varied according to One-way ANOVA ($P = 0.17$). The average densities of fish, squilloidea, crab, decapod and cephalopoda in each group were presented in Figure 3, which demonstrated obviously higher values of crustaceans (crab, decapod and squilloidea) densities in group A than in group B. while, only decapod ($P = 0.011$) and squilloidea ($P = 0.012$) showed statistically differences between group A and B.

The Shannon-Wiener index (H') in group A and B were 2.74 and 2.3 respectively, which varied from 2.38 to 2.98 and 2 to 2.54 in different sites of two groups. The Pielou evenness index (J') in the study area presented the consistent results, with an average value of 0.78 (varied from 0.74 to 0.82) in group A and 0.7 (varied from 0.62 to 0.76) in group B. On the basis of one-way ANOVAs, the Shannon-Wiener index (H') ($P = 0.031$) and Pielou evenness index (J') ($P = 0.042$) in group A were significantly higher than that in group B (Table III).

Factors contributing to spatial heterogeneity of nekton community

Biological effect factors

The similarity of nekton community in group A was

58.89 in April 2017 based on SIMPER analysis and the key species were *Charybdis callianassa*, *Harpiosquilla harpax*, *Alpheus brevicristatus*, *Oratosquilla nepa*, and *Parapenaeopsis hardwickii*, which cumulatively contributed up to 30.39 (Table IV). The similarity of nekton community in group B was 59.27, which was mainly contributed by *Parargyrops edita*, *Trachurus japonicus*, *Leiognathus berbis*, *Saurida tumbil*, and *Psenopsis anomala*, with a cumulative contribution rate of 38.72. The dissimilarity of nekton community between group A and B was 65.05, and the cumulative contribution rate of *Alpheus brevicristatus*, *Charybdis callianassa*, *Harpiosquilla harpax*, *Oratosquilla nepa*, and *Parapenaeopsis hardwickii* reached 15.85. As a result, these Crustaceans species such as *Alpheus brevicristatus*, *Charybdis callianassa*, *Harpiosquilla harpax*, *Oratosquilla nepa*, and *Parapenaeopsis hardwickii* would be identified as the main biological effect factors contributing to the heterogeneity of nekton community between group A and B based on the SIMPER analysis and intuitively presented in Figure 4 (stress = 0.01).

Table III. Spatial variation of Shannon-Wiener diversity (H') and Pielou evenness (J') indexes between group A and B.

	Group A	Group B	F	P
Shannon-Wiener index (H')	2.74±0.21	2.3±0.28	7.243	0.031*
Pielou evenness (J')	0.78±0.03	0.7±0.07	6.18	0.042*

*Note: * means significant differences at $\alpha=0.05$ level.

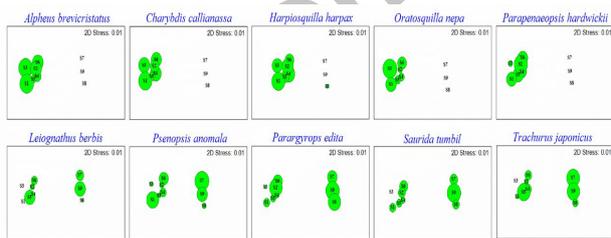


Fig. 4. Distribution pattern of the dominants contributing to spatial heterogeneity of nekton community based on NMDS analysis (the size of the bubbles showing the abundance of the dominants species).

Environmental effect factors

The mean values of water transparency (Tra), water temperature (WT), dissolved oxygen (DO), total inorganic nitrogen (TIN) and total phosphorus concentration (TP) in group A were 5.73 m, 18.96 °C, 7.10 mg/L, 276.36 ug/L and 3.88 ug/L respectively, which were relatively

higher than that of 3.27 m, 18.62 °C, 6.32 mg/L, 191.29 ug/L and 3.49 ug/L in group B. In contrast, salinity (Sal) and pH were relatively lower in group A (29.38 and 8.17, respectively) than that in group B (29.93 and 8.35, respectively). The difference of these parameters between group A and B were tested by one-way ANOVA, which revealed significantly higher values of Tra ($P=0.005$), DO ($P=0.001$), and TIN ($P=0.032$) in group A than that in group B (Table V).

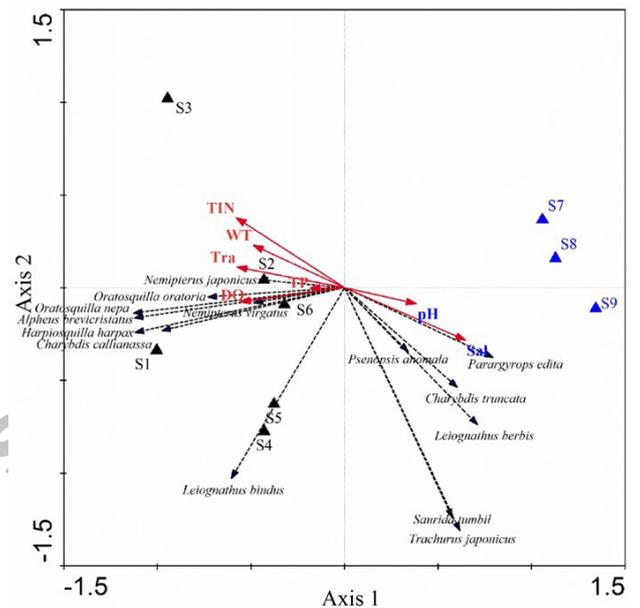


Fig. 5. RDA ordination of nekton community composition and the relationship with physicochemical environmental parameters in different sample sites (S1~S9).

RDA analysis based on the dominant nekton species matrix and the main physicochemical environmental parameters matrix revealed that axis 1 and axis 2 (Fig. 5) illustrate the spatial heterogeneity of nekton community between group A and B, and its corresponding relationship with the environmental factors. The interpretation rates of axis 1 and axis 2 for the spatial heterogeneity of nekton community between group A and B were 0.562 and 0.127, and the corresponding interpretation rates for the relationship between nekton community and physicochemical environmental parameters were 0.646 and 0.146, respectively. The dominant nekton species and physicochemical environmental parameters were generally distributed along the axis 1 and the sampling sites were roughly divided into two groups by axis 2. Crustaceans species including *Alpheus brevicristatus*, *Oratosquilla oratoria*, *Oratosquilla nepa*, *Charybdis callianassa*, and *Harpiosquilla harpax*, were mainly distributed in group A

Table IV. Dominants (Boldface) contribute to nekton community similarity inner groups and dissimilarity between group A and B based on SIMPER analysis.

	Group A		Group B		A versus B	
	Av. sim		Av. sim		Av. diss	
	58.89		59.27		65.05	
	Sim	Contrib	Sim	Contrib	Diss	Contrib
<i>Charybdis callianassa</i>	3.78	6.41	0	0	2.28	3.5
<i>Harpiosquilla harpax</i>	3.77	6.39	0	0	2.27	3.49
<i>Alpheus brevicristatus</i>	3.74	6.34	0	0	2.17	3.34
<i>Oratosquilla nepa</i>	3.51	5.95	0	0	1.91	2.94
<i>Parapenaeopsis hardwickii</i>	3.12	5.3	0	0	1.68	2.58
<i>Parargyrops edita</i>	3.1	5.27	5.42	9.14	0.4	0.61
<i>Trachurus japonicus</i>	1.88	3.2	4.7	7.93	0.69	1.06
<i>Leiognathus berbis</i>	1.27	2.15	4.37	7.37	1.01	1.55
<i>Saurida tumbil</i>	1.8	3.06	4.25	7.17	0.54	0.83
<i>Psenopsis anomala</i>	2.84	4.83	4.22	7.11	0.36	0.55

*Note: Region A represents artificial reefs and its adjacent area; Region B represents the control region; Av. Sim represents average similarity; Av. Diss represents average dissimilarity; contrib represents contribution.

Table V. One-way ANOVAs of physiochemical environmental parameters (mean \pm SD) between group A and B.

	Group A	Group B	F	P
Tra (m)	5.73\pm1.02	3.27 \pm 0.31	$F=15.833$	$P=0.005^{**}$
WT ($^{\circ}$ C)	18.96\pm0.37	18.62 \pm 0.16	$F=2.274$	$P=0.175$
Sal	29.38 \pm 0.38	29.93\pm0.32	$F=4.558$	$P=0.070$
pH	8.17 \pm 0.22	8.35\pm0.33	$F=1.708$	$P=0.233$
DO (mg/L)	7.10\pm0.22	6.32 \pm 0.21	$F=25.888$	$P=0.001^{**}$
TIN (ug/L)	276.36\pm50.07	191.29 \pm 29.02	$F=7.127$	$P=0.032^{*}$
TP (ug/L)	3.88\pm1.19	3.49 \pm 0.62	$F=0.271$	$P=0.619$

*Note: Tra, transparency; WT, water temperature; Sal, salinity; DO, dissolved oxygen; TIN, total inorganic nitrogen; TP, total phosphorus; *, significant difference at 0.05 level; **, significant difference at 0.01 level.

(S1-S6), which showed a positive relationship with WT, DO, TIN, TP and Tra. In contrast, fish species such as *Parargyrops edita*, *Psenopsis anomala*, *Saurida tumbil*, *Leiognathus berbis*, and *Trachurus japonicus*, were mainly distributed in group B (S7-S9) with relatively higher Sal and pH values. On the basis of partial Monte Carlo permutation test, the interpretation rate of DO, Tra and TIN to the spatial heterogeneity of nekton community were 0.44, 0.16 and 0.08 respectively, which were remarkably higher than other parameters included in this research. Therefore, parameters such as DO, Tra and TIN would be identified as the main environmental effect factors contributing to the heterogeneity of nekton

community between group A and B

DISCUSSION

Methods applied in ecological effect evaluation of AR

April and September are two very important periods for marine life in the South China Sea. The biological resources were relatively abundant in these periods, which was supposed to be the critical period of production and growth of some nekton species. In this research, the crustaceans and fishes in the breeding period accounted for a great proportion among the catches, such as *Leiognathus berbis*, *Trachurus japonicus*, *Upeneus sulphureus*, *Oratosquilla oratoria* etc. Therefore, a policy of strict prohibition of fishing in the South China Sea was set during May to late August to protect the reproductive parents and young and juvenile. The nekton species similarity evaluated by Jaccard coefficient was in the extremely dissimilar level (0.25 to 0) between April and September and the dominant species varied obviously with the only two common dominant species of *Harpiosquilla harpax* and *Oratosquilla nepa* during this period, which demonstrated varied ecological effects of AR in different seasons. According to the result of CLUSTER analysis, the spatial heterogeneity of nekton community was only demonstrated in April which confirmed the above conclusion further. Therefore, the ecological effects evaluation of AR was recommended to be carried out quarterly.

The conservation effect of AR on marine fishery

resources has always been concerned. How to evaluate the construction effect of AR scientifically and comprehensively is crucial to explore the mechanism of ecological effects of AR. Dynamic of fishery resources is the most direct response of AR's ecological effects and can be used as the basic parameter for the ecological effects evaluation of AR. In recent decades, many researches focusing on fishery resources dynamic have been carried out to evaluate the ecological effects of AR. Plenty of studies have shown that the bait effect, flow field effect and shelter effect produced by AR have improved the growth and habitat environment of marine organisms (Clynick *et al.*, 2008; Beeler, 2009; Burt *et al.*, 2010). As a result, the species, quantity and biomass of marine organisms in AR areas are generally higher than those in natural sea or reef areas, showing a good biological attraction effect (Rilov and Benayahu, 2000; Reed *et al.*, 2006). In this study, the density of nektons in group A (48529 ind/km²) was obviously higher than that in group B (30220 ind/km²), although the variation was not statistically significant ($P=0.17$). Further studies on different taxa of nekton community (fish, crabs, shrimps etc.) showed that there was no significant differences on densities of fish and cephalopod between group A and B ($P=0.714$ and 0.767 , respectively), while the density of crustaceans (crabs, shrimps and squid) in group A was dramatically higher than that in group B ($P=0.07$, 0.011^* , and 0.012^*). This indicated a clear biological attraction effect of AR on these crustaceans. Similar results were founded in the AR area of Dongshanghai, Huizhou, China in 2019 (unpublished). Therefore, the taxa of nekton community should be given full consideration on the ecological effects evaluation of AR.

It is well known that fishery resource dynamics often depend on a few dominant species. Therefore, comparative analysis of dominants composition and change characteristics based on AR area and the control area can be used as another important factors to evaluate the ecological effects of AR. As reported by Wang *et al.* (2010), the AR area not only retained the dominants in the adjacent control area, but also increased other rock species such as *Sebastes marmoratus* based on the study of fish and macroinvertebrates community structure in artificial habitat around Sanheng Isle, Shengsi, China. The result indicated that the attraction responses of AR on marine organisms were species-specific (Santos and Monteiro, 2007). Similar result was founded in this research according to the IRI analysis of nektons in group A and B. In addition to the dominants (*Psenopsis anomala*, *Parargyrops edita*, *Nemipterus virgatus*, and *Leiognathus berbis*) common to group A and B, there were other unique dominants in group A such as *Alpheus brevirostratus*, *Oratosquilla nepea*, *Charybdis callianassa* (Table II). In

other words, these crustaceans showing a distinct tendency towards AR reflected the ecological effects of AR to some extent. However, the composition of these species were not entirely consistent with those species that caused spatial heterogeneity of the nekton community between group A and B (Table IV). Therefore, the ecological effects of AR cannot be accurately reflected depending on the spatial distribution differences of dominants alone.

Biodiversity indexes have been considered as ecological effects indicators of oceanographic engineering for a long time (Castillo-Rivera and Zavala-Hurtado, 2002). However, a single biodiversity index cannot effectively describe the characteristics of biological communities in a specific habitat. Therefore, many scholars have proposed a series of biodiversity indexes, which reflect the biological community characteristics in the content of structural, taxonomic and functional (Nick *et al.*, 2010; Stuart-Smith *et al.*, 2013). Relevant studies have shown that the level of fish diversity generally increases in the short term after the deployment of AR (Folpp *et al.*, 2011). However, it is a dynamic change process over a long time, which may be higher or lower than that in natural waters (Nicoletti *et al.*, 2007). In this research, both the Shannon-Wiener index (H') and Pielou evenness index (J') in the AR region (A) were significantly higher than that in the control region (B), which implied an obvious ecological restoration effect since the ARs were deployed in 2009.

The construction of AR not only achieves the effects of conservation and recruitment of marine fishery resources (Schroepfer and Szedlmayer, 2006), but also make a change to the community structure of marine organisms. However, when a species in a community is replaced by another one with similar ecological niche, such changes are often not reflected by biomass or biodiversity indexes (Francisco *et al.*, 2008). In this research, the difference of nektons' density between group A and B was not significant in statistic. While, the spatial heterogeneity of nekton community was demonstrated between group A and B, which would be roughly regarded as the effect and non-effect zone of the AR according to CLUSTER analysis. That means the ecological effects range of AR reached 3 km in the study area since the ARs were deployed in 2009, and this range was obviously greater than the result of 50 m assessed based on the abundance difference of fishes in varied distance from the centre of ARs (Santos *et al.*, 2010). As mentioned above, the attraction responses of AR on marine organisms were species-specific. Therefore, the ecological effects range assessment of AR based only on spatial heterogeneity of fish abundance might be underestimated. Furthermore,

the key crustaceans (*Alpheus brevicristatus*, *Charybdis callianassa*, *Harpisquilla harpax*, *Oratosquilla nepa* and *Parapenaeopsis hardwickii*) extracted by SIMPER analysis, would be regarded as the main biological effect factors contributing to the spatial heterogeneity of nekton community between group A and B. While the spatial distribution of these fishes such as *Parargyrops edita*, *Leiognathus berbis*, *Saurida tumbil*, *Trachurus japonicus* and *Psenopsis anomala* were relatively uniform in the study area according to the NMDS plot, and these species contributed rarely to the dissimilarity of nekton community between group A and B. Hence, analysis based on community structure can reflect more information than density and biodiversity indexes, it not only describes the number of species in the habitat, but also considers the distribution pattern and niche relationship among different species (Legendre *et al.*, 2005; Socolar *et al.*, 2016). All in all, to make a comprehensive evaluation of ecological effects of AR, multidimensional evaluations including density, dominants, biodiversity, community structure *et al* should be combined.

Spatial heterogeneity of nekton community and its linkage with environmental factors

Multivariate statistical analysis including RDA, Bio-Evn, CCA *et al*, is a series of methods that study the relationship between multiple variable sets. These methods can reduce the complexity of the data matrix, extract important information, and intuitively represent the relationship between multiple variables (Clarke and Warwick, 2001). It is often used to analyze the spatial and temporal heterogeneity of biological community and their linkage relationship with environmental factors in different habitats (Simonsen *et al.*, 2013). In this study, the axis 1 and axis 2 in RDA plot (Fig. 5) can well explain the linkage relationship between the nekton community and the main environmental factors in the study area in April 2017. As a result, crustaceans such as *Alpheus brevicristatus*, *Oratosquilla oratoria*, *Charybdis callianassa*, *Harpisquilla harpax*, and *Oratosquilla nepa* showed a positive correlation with WT, DO, Tra, TIN and TP were mainly distributed along axis 1. While, species such as *Psenopsis anomala*, *Saurida tumbil*, *Parargyrops edita*, *Leiognathus berbis*, and *Trachurus japonicus* prefer to higher Sal and pH. As the construction of AR has changed the habitat of nekton community between group A and B, crustaceans showed a preference to the habitat with AR, which slow down water flow and provide a shelter (Danovaro, 2002) to the weak swimming species. In addition, previous studies indicated that the impact of environmental factors on the structure of nekton community varied from time to place (McQuoid, 2005;

Shukla and Bhat, 2018). As pointed out by Lunven *et al.* (2005), the plankton community in Skakai Bay is mainly regulated by phosphates in summer, while it was greatly affected by nitrates in summer. In this study, the analysis of preselected environmental factors by partial Monte Carlo test showed that DO, Tra and TIN were the main abiotic factors affecting the spatial heterogeneity of nekton community in April 2017. However, there were only seven physicochemical environmental factors considered in this research and more than half of these factors showed slight difference between group A and B. Therefore, the preference tropism effect of crustaceans on AR may be affected by other factors such as food coefficient, water flow characteristics and physical structural complexity (Coll *et al.*, 1998; Charbonnel *et al.*, 2002). For instance, the changes in the flow field of AR will lead to the changes in trophic level of the waters, thus changes the feeding conditions of the crustaceans, and has an impact on the spatial distribution of crustaceans through biological cascade effect. AR as an open ecosystem, are affected by many factors. It not only produces instantaneous ecological effects on phytoplankton community, but also has an influence on the structure of nekton community simultaneously through the bottom-up effect on phytoplankton zooplankton fish or other nektons linkage (Perkol-Finkel and Benayahu, 2005). Nektons, as the superior consumers in the food network of coastal marine ecosystem, are the concentrated expression of the comprehensive effects of other biological and abiotic factors in the ecosystem (Wu *et al.*, 2014). The study based on the spatial heterogeneity of nekton community is an indispensable part of the research on the ecological effects mechanism of AR which can be more comprehensive to explain the interaction between biological communities and environment.

CONCLUSION

In order to evaluate the ecological effects of AR scientifically, multidimensional evaluation methods concerning resources dynamic, dominants, biodiversity and community structure would be more appropriate. Furthermore, to achieve a more comprehensive understanding of the ecological cascade effect mechanism of AR, more physicochemical (such as the structure and material of ARs) and biological factors (such as phytoplankton and zooplankton) should be considered in future studies.

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Statement of conflict of interest

The authors have declared no conflict of interest.

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