



Adaptation strategies of relatively high-latitude marginal reef corals in response to severe temperature fluctuations

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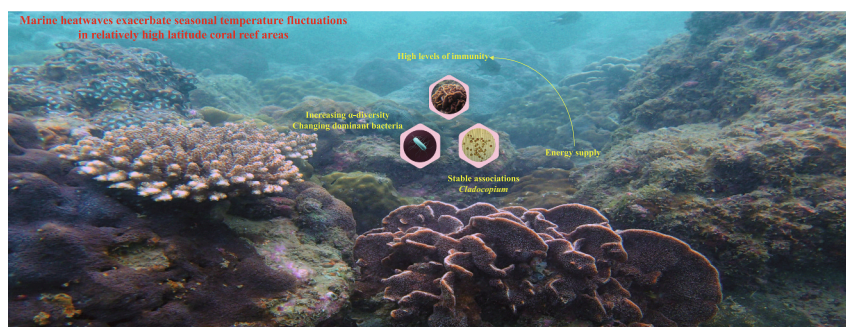
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HIGHLIGHTS

- We studied adaptation plasticity of scleractinian corals facing seasonal stress.
- Dynamic balance of immunity costs and energy supply was important.
- High temperatures in summer threaten relatively high latitude corals.
- Interspecific differences in adaptation strategies were observed.
- Coral had similar adaptation strategies in summer and winter.

GRAPHICAL ABSTRACT



ARTICLE INFO

Editor: Fang Wang

Keywords:

Climate change
South China Sea
Coral reef ecosystem
Coral holobionts
Adaptation strategies
Seasonal stress

ABSTRACT

The large seasonal temperature fluctuations caused by global warming and frequent marine heatwaves pose new challenges to survival of relatively high-latitude marginal reef corals. However, the adaptation strategies of high-latitude marginal corals are not fully understood. We employed integrated approach to investigate the response mechanism of hosts, Symbiodiniaceae, and symbiotic bacteria of marginal reef corals *Acropora pruinosa* and *Pavona decussata* in response to large seasonal temperature fluctuations. The coral holobiont maintained a high level of immunity to adapt to seasonal pressure by increasing Symbiodiniaceae energy supply. The symbiotic Symbiodiniaceae of two coral was dominated by C1 subgroup, and was stable across seasons. The α -diversity of symbiotic bacteria *P. decussata* and *A. pruinosa* in summer was higher than that in winter. The symbiotic bacterial community of two coral reorganized during different seasons. Scleractinian corals improve adaptability to seasonal stress by increasing energy supply to maintain high levels of immunity, increasing symbiotic bacterial α -diversity, and changing dominant bacteria. This study demonstrates the adaptation strategies of marginal reef corals to seasonal temperature fluctuations and provides novel insights into the study of the adaptation of corals and relatively high-latitude coral refuges in the context of global warming and intensified marine heatwaves.

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1. Introduction

Relatively high-latitude marginal reefs are considered as climate change refuges (Yao and Wang, 2022). However, the frequent occurrence of marine heatwaves (MHW) pushes the coral reef ecosystem at the edge of relatively high latitudes to the limit of its tolerance, and they may have a serious impact on future coral survival and coral reef development. According to previous research, the intensity of MHW in the high-latitude coral reef areas is higher, and it increased at a rate of 0.1–0.3 °C/count per decade (Yao and Wang, 2022). Large-scale coral bleaching events caused by abnormal high temperatures have been observed in many relatively high-latitude coral reef areas. For example, the high-latitude coral reef (33°S) suffered a severe bleaching event between 2010 and 2019 (Davis et al., 2020), and the high-latitude ~34°S coral community in eastern Australia experienced a severe bleaching event during 2015–2016 (Goyen et al., 2019). In 2015, 2016 and 2020, the marginal reefs in the South China Sea experienced severe albinism and death (Mo et al., 2022; Yu et al., 2021a, 2021b, 2021c). The high-latitude coral reefs in Okinawa experienced a sharp decline after bleaching in 1998 and 2001 (Hongo and Yamano, 2013). These events show that abnormal high temperature caused by global warming and MHW threaten high-latitude coral reefs (Mo et al., 2022). Moreover, previous studies have found that abnormal high temperature events such as MHW increase the sea surface temperature (SST) in coral reef areas at relatively high latitudes. Strong SST anomalies, along with the East Asian winter monsoon, have led to low winter SST, leading to increased seasonal temperature fluctuations (Feng et al., 2022; Zhou et al., 2022). Large seasonal temperature fluctuation stress is a serious challenge for relatively high-latitude reef areas that act as future coral reef refuges (Beger et al., 2014; Yu et al., 2021). In the context of global warming, the frequency of extreme events such as MHW may increase in the future, and the climate in relatively high-latitude reef areas may become unpredictable and complex (Jiang et al., 2021).

The ability to adapt to large seasonal temperature fluctuations is crucial for the future of corals. Some researchers have found that the growth of littoral corals from different fringing reef areas in the South China Sea has evident seasonal characteristics (Yu et al., 2005). The density of symbiotic Symbiodiniaceae of the five scleractinian corals in Hainan was the highest in summer and lowest in winter, and the photosynthetic rate of symbiotic Symbiodiniaceae was exactly the opposite (Xu et al., 2017). In addition, studies have reported that positive thermal deviation in summer is more unfavorable to the physiological maintenance and survival of *Acropora downingi* than the negative thermal deviation in winter from Hengam Island. The balance between net photosynthesis and dark respiration is lower under high temperature in summer than in winter (Vajedsamiei et al., 2015). Differences in trophic regimes in response to summer high temperatures exist for scleractinian corals in the northern South China Sea; for example, *P. lutea* and *Favia palauensis* rely on heterotrophy to maintain metabolic energy requirements. *Acropora millepora* and *Pocillopora damicornis* are more dependent on consuming their own lipids (Xu et al., 2022). Some studies have also found that there are differences in the symbiotic bacteria of scleractinian corals between different seasons, which may help them adapt to seasonal environmental fluctuations (Yu et al., 2021a, 2021b, 2021c). For example, during the transition of *Platygyra carnosa* and *Galaxea fascicularis* from winter to summer, the symbiotic bacteria related to sulfur oxidation, nitrogen fixation, and photosynthesis, experience an increase in bacteria associated with chemical synthesis; however, the bacteria potentially associated with ammonia oxidation and nitrite oxidation decrease from winter to summer (Cai et al., 2018). However, there is a lack of systematic research that focuses on long-term outcomes regarding the adaptation strategies of relatively high-latitude coral to stress caused by large seasonal temperature fluctuations.

Our study area was Weizhou Island, a relatively high-latitude coral reefs in the South China Sea, which is also been regarded as a potential refuge for corals under global warming (Yu et al., 2019). Under the

background of global warming, the SST at Weizhou Island shows an evident upward trend, with a rate of increase exceeding that of both the global and Northern Hemisphere SST (Zhang et al., 2020). Since 1997, there have been many peaks in the highest monthly mean SST in Weizhou Island in summer (Zhang et al., 2020). In addition, Weizhou Island frequently has several high temperature years and low temperature years, and the frequency and intensity of high and low temperature events are increasing (Zhang et al., 2020). Notably, the high average temperature, the weak East Asian winter monsoon, and the extension of the western Pacific subtropical high to the north and west have led to the frequent occurrence of MHW in Weizhou Island (Feng et al., 2022; Yao and Wang, 2022). Since 1983, there have been 112 MHW events in Weizhou Island, and the frequency and duration of MHW events have shown an evident increasing trend (Feng et al., 2022), particularly in the summer of 2020, and the abnormal high temperature events caused by MHW have led to a large area of coral bleaching death at Weizhou Island (Yu et al., 2021a, 2021b, 2021c). In our previous study, we have analyzed the environmental characteristics of Weizhou Island, and seawater surface temperature (SST) is one of the most important factors affecting the coral reefs of Weizhou Island (Yu et al., 2021c). Therefore, Weizhou Island is a suitable choice for studying adaptation strategies of scleractinian corals in response to large-scale seasonal temperature fluctuations. Two representative corals with significantly different tolerance to temperature stress, *A. pruinosa* and *P. decussata*, were used as research subjects. The results of surveys on the coral reef ecosystem of Weizhou Island over the past 30 years indicate that *P. decussata* is more tolerant to stress than *A. pruinosa* (Yu et al., 2019). An integrated approach was used to investigate the regulatory roles of coral hosts, symbiotic Symbiodiniaceae, and symbiotic bacteria in adaptation strategies. The results of this study will improve our understanding of the ability of subtropical transitional reef communities to adapt in the context of global warming, thereby providing reference basis for researchers in ensuring the continuity of these communities and their potential to act as refuges for tropical coral reef species.

2. Materials and methods

2.1. Collection and processing of coral samples

We studied two representative scleractinian corals (the branching coral *A. pruinosa* and the foliaceous coral *P. decussata*) of Weizhou Island (N21°8.27', E109°12.6'). Samples were collected in March 2019 to January 2020 in spring (March 2019, "Spr" group), summer (July 2019, "Sum" group), autumn (October 2019, "Aut" group) and winter (January 2020, "Win" group). All corals were sampled from the same depth (approximately 3–5 m). We made careful comparisons and collected morphologically distinct samples to minimize the chance of multiple sampling of the same ramet (Chen et al., 2021). Coral fragments were obtained by scuba diving with hammers and chisels. Coral tissue samples of approximately 2–4 cm in diameter were collected from the outer edge of each coral. All samples were immediately snap-frozen in liquid nitrogen and stored at –80 °C.

2.2. Analysis of symbiotic Symbiodiniaceae community composition

Genomic DNA was extracted from 100 to 200 mg of frozen coral tissue of 40 coral samples (five samples of each coral species per season) using the DNeasy Plant Mini Kit (QIAGEN, Hilden, Germany). Qualified DNA was used for subsequent experiments. We performed PCR amplification of the ITS2 sequence using a previously described method (Chen et al., 2019). ITS2 sequence data analysis and operational taxonomic unit (OTU) data were analyzed to assess symbiotic Symbiodiniaceae diversity and community composition. As previously described methods (Chen et al., 2019; Ziegler et al., 2017), BLASTn was used to compare all sequences with those deposited in the ITS2 database. We analysed symbiotic Symbiodiniaceae diversity and community composition. The

minimum cutoff greater in this analysis is set to 5 % and filtered the results (Ziegler et al., 2017). Subsequently, the ITS2 sequence was flattened (1000 reads) using MOTHUR. Sequences with retention alignment lengths >90 % were first clustered into OTUs and then further clustered under a 97 % similarity threshold (Arif et al., 2014).

2.3. Analysis of symbiotic bacterial diversity and community composition

Genomic DNA was extracted from 100 to 200 mg of frozen coral tissue of 28 *A. pruinosa* samples (6, 10, 6, and 6 samples in four seasons, respectively) and 35 *P. decussata* samples (10, 10, 9, and 6 samples in four seasons, respectively) using the TIANamp Marine Animal DNA Kit (Beijing Tiangen Biotechnology Co., Ltd., China). Qualified DNA was used for subsequent experiments. PCR amplification of bacterial 16S rRNA V3-V4 using primers 338F and 806R (Liang et al., 2017). The purified PCR products subjected to paired-end sequencing (2 × 300 bp) using the IlluminaMiSeq platform (Illumina, San Diego, USA) according to the standard protocol.

The original sequencing reads were demultiplexed, quality filtered, and merged as previously described methods and parameter settings (Yu et al., 2021a, 2021b, 2021c). OTUs with a 97 % similarity cutoff were clustered using UPARSE, and chimeric sequences were identified and removed. The classification of each OTU representative sequence was analyzed using the RDP classifier against a 16S rRNA database (e.g., SilvaSSU128) with a confidence threshold of 0.7. The OTU analyses of sequences, including coverage, ACE, Shannon index, and Simpson's index, were performed using MOTHUR software. Microbial community beta diversity was determined based on the Bray–Curtis distance matrix and visualized by principal coordinate analysis (PCoA) (Zaneveld et al., 2017). Differences in microbial communities were tested using analysis of similarity (ANOSIM) with 9999 permutations (Ziegler et al., 2019). To further determine whether specific individual bacterial taxa were differentially enriched across seasonal groups, we applied LEfSe analysis, which uses LDA and an effect size measure (LDA: 4, all-against-all (more strict)). Venn and collinear network analysis revealed the differences in symbiotic bacteria among different groups. A phylogenetic tree was constructed using the neighbor-joining method via MEGA software. Finally, QIIME2 was used to determinate the core microbiome bacteria, which were present in >80 % of the sample (Hernandez-Agreda et al., 2017).

2.4. Transcriptome analysis of coral hosts and symbiotic Symbiodiniaceae

Qualified RNA of 40 coral samples (five samples of each coral species per season) were used for subsequent experiments. Paired-end RNA-seq libraries were sequenced (2 × 150 bp read length, Illumina) using the HiSeqXTen platform.

Sequencing data for all samples were used for assembly. Raw paired-end reads were trimmed, and SeqPrep and Sickle with default parameters were used to verify quality. The clean data were then assembled de novo using Trinity. After assembly, the assembled unigenes were annotated by BLAST in Kyoto Encyclopedia of Genes and Genomes (KEGG), non-redundant (Nr), and Gene Ontology (GO). Based on previous studies (Yu et al., 2020a, 2020b; Yu et al., 2021a, 2021b, 2021c), BLASTx was used to distinguish corals host or Symbiodiniaceae (Bay and Palumbi, 2015; Mansour et al., 2016; Yu et al., 2020a, 2020b; Yu et al., 2021a, 2021b, 2021c).

We used RSEM to quantify gene abundance. Differentially expressed genes (DEGs) (FDR < 0.05) between groups were screened using the obtained BAMfiles and DESeq2 software (Love et al., 2014). Transcript expression levels were measured in terms of fragments per kilobase of transcript in the reads per million mapped approach.

In the KEGG enrichment analysis, we used over-representation analysis (ORA) using Fisher's exact test (Benjamini–Hochberg adjusted $p < 0.05$) (Désert et al., 2008). The Benjamini–Hochberg method for multiple testing correction (Benjamini and Hochberg, 1995; Désert

et al., 2008) was used to correct the p -values for the probability or frequency of errors in the overall inference results.

3. Results

3.1. Seasonal variability among the Symbiodiniaceae

In this study, we generated and analyzed 2,713,692 high-quality sequence reads. In this study, although there were slight differences between different samples, Clade C1 (Cladocopium) was the most dominant in *A. pruinosa* and *P. decussata* on Weizhou Island, with a mean relative abundance of 81.60 % (Fig. S1). Furthermore, there is no significant difference in OTU richness of Symbiodiniaceae between the different groups ($p < 0.05$). Raw reads were deposited into the NCBI sequence reads archive (SRA) database: PRJNA841798.

3.2. Plasticity of symbiotic bacteria

The results of symbiotic bacteria showed that 2,670,913 treated bacterial sequences were assigned to 1786 OTUs with a similarity of 97 % (Table S1). Raw reads were deposited into the NCBI SRA databases: PRJNA687160 and PRJNA841966. The average length of bacterial sequences were 420 bp (200 to 530 bp) (Table S2). Good's coverage was >98.76 % coverage of symbiotic bacteria for each sample. Bacterial community richness indices include Sob, Chao, and ACE indices (Table S3). The α -diversity of symbiotic bacteria in the summer samples of both corals was higher than that in the winter samples, and the number of OTUs of *P. decussata* was higher than that of *A. pruinosa* in summer and winter (Fig. 1, Student's test, $p < 0.05$). Consistent with the regularity of OTU numbers, the symbiotic bacterial community richness of *P. decussata* in both summer and winter was higher than that of *A. pruinosa*, and the α -diversity of symbiotic bacteria was higher in the summer samples than in winter samples (Fig. 1, Student's test, $p < 0.05$). PCoA shown the similarity of the symbiotic bacterial communities associated with *A. pruinosa* and *P. decussata*, accounting for 41.02 % and 30 % of the total variance, respectively ($R = 0.7838$, $p = 0.001$) (Fig. 1F). To compare the taxonomic composition of *A. pruinosa* and *P. decussata* in summer and winter, we performed a Venn analysis of OTU composition corresponding to four groups (Fig. 1E). Among them, 146, 48, 409, and 139 core OTUs were unique to Sum_Acro, Win_Acro, Sum_Pavo, and Win_Pavo, respectively, and 91 OTUs were shared by each group.

Corals are highly diverse and rich in microbial communities, which is also an adaptation way of coral holobionts (Ziegler et al., 2019). Fig. S2 shows the microbial composition of *A. pruinosa* and *P. decussata* at the genus level. For *A. pruinosa*, the dominant genus was *Ralstonia* in spring, accounting for 90.84 %. The dominant genera were BD1-7_clade and *Prosthecochloris* in summer, accounting for 47.31 % and 16.80 %, respectively. The dominant genera were BD1-7_clade and *Ralstonia* in autumn, accounting for 59.16 % and 18.22 %, respectively. The dominant genera were *Ralstonia* and *Acinetobacter* in winter, accounting for 62.88 % and 14.44 %, respectively. For *P. decussata*, BD1-7_clade, the dominant genera were *Cohaesibacter*, and *Fusibacter* in spring, accounting for 15.16 %, 11.73 %, and 10.75 %, respectively. The dominant genera were BD1-7_clade and *Vibrio* in summer, accounting for 43.68 % and 14.04 %, respectively. The dominant genus was BD1-7_clade in autumn, accounting for 92.39 %. The dominant genera were BD1-7_clade and *Ralstonia* in winter, accounting for 32.94 % and 30.95 %, respectively.

Circos analysis shown the corresponding abundance relationships between bacterial communities and samples at the genus level (Fig. 2). The three bacterial genera with the highest abundance were BD1-7_clade, *Ralstonia*, and *Prosthecochloris*. LEfSe analysis led to the identification of 11 of the 646 genera that were differentially enriched across groups (Fig. S3): *Ruegeria*, *Acinetobacter*, *Thermus*, BD1-7_clade, *Vibrio*, *Francisella*, *Labrenzia*, Sva0996_marine_group, *Pseudovibrio*,

Endozoicomonas, and JTB255_marine_benthic_group. In addition, the collinear network map revealed the differences in symbiotic bacteria of *A. pruinosa* and *P. decussata*. Only a few OTUs of the two groups were shared, and the unique OTUs of the *P. decussata* were far exceeded the shared OTUs (Fig. S4). Phylogenetic tree revealing the evolutionary relationship between the symbiotic bacteria of *A. pruinosa* and *P. decussata* (Fig. S5).

In this study, six identified bacterial core OTUs in four groups were associated with *A. pruinosa* and *P. decussata* (Fig. 3, Table S4). Although the abundance of the core microbial community fluctuated among different groups, the core microbial community of *P. decussata* contributed 38.69 % of the bacterial abundance, while Sum_Acro, Sum_Pavo, and Win_Acro contributed 30.17 %, 34.56 %, and 28.38 %, respectively (Fig. 3). Among the core bacteria, BD1-7_clade (OTU961) was most abundant in the Sum_Acro (59.04 %), Sum_Pavo (83.46 %), Win_Acro (11.53 %), and Win_Pavo (41.80 %) groups (Fig. 3). *Acinetobacter* (OTU4642) was the second most dominant bacterium, accounting for 17.99 % (Sum_Acro), 1.02 % (Sum_Pavo), 49.70 % (Win_Acro), and 18.23 % (Win_Pavo) of the core microbial community, (Fig. 3). In addition, *Thermus* (OTU734) dominated the core bacterial microbiome of winter in *A. pruinosa* and *P. decussata*, accounting for 25.65 % and 25.27 %, respectively (Fig. 3).

3.3. Differential transcriptional responses of coral hosts and symbiotic Symbiodiniaceae to large seasonal temperature fluctuations

In the analysis, 2,141,748,814 high-quality clean reads were obtained by filtering from 2,164,987,348 paired-end raw reads, of which Q20 bases accounted for 98.36 %, Q30 bases accounted for 94.71 %, and the GC content was 47.96 % (Table S5). Raw reads were deposited in the NCBI SRA database: PRJNA841791. In addition, de novo assembly yielded 158,308 unigenes with an N50 length of 1966 nt and an average length of 1148.55 nt (Table S6). Coral hosts for each sample had an alignment rate of 15.13–46.49 % (Table S7), while symbiotic Symbiodiniaceae had an alignment rate of 17.43–55.49 % (Table S8).

The expression levels of 158,308 coral host unigenes (Table S9) and 38,177 Symbiodiniaceae unigenes (Table S10) were quantitatively analyzed using RSEM with TPM as an indicator. The difference analysis software DESeq2 (significance level: p -adjust < 0.05, multiple test correction method: BH, up/down difference fold: 2.0) analyzed the differential genes of *A. pruinosa* and *P. decussata* in summer and winter. Compared with *A. pruinosa*, 49,048 upregulated coral host genes, 42,099 downregulated coral host genes, 616 upregulated symbiotic Symbiodiniaceae genes, and 667 downregulated symbiotic Symbiodiniaceae genes were identified in *P. decussata* in summer. In total, 41,924 upregulated coral host genes, 43,884 downregulated coral host genes, 172 upregulated symbiotic Symbiodiniaceae genes, and 184 downregulated symbiotic Symbiodiniaceae genes were identified in *P. decussata* in winter. This further confirms the mechanism of the adaptation of the two types of corals to the large seasonal temperature fluctuations in relatively high-latitude seas. Venn analysis revealed the DEGs; 36,472 coral genes and 73 symbiotic Symbiodiniaceae genes were significantly upregulated in summer and winter in *P. decussata*, and 39,089 coral genes and 71 symbiotic Symbiodiniaceae genes were significantly upregulated in summer and winter in *A. pruinosa* (Fig. S6).

To further understand the molecular process of the differential adaptation of two scleractinian corals to summer and winter environments, we performed KEGG functional enrichment analysis on the DEGs identified above. In summer, *P. decussata* upregulated host genes were enriched in seven pathways, and the most notably enriched pathways were the dorso-ventral axis formation (map04320), Th1 and Th2 cell differentiation (map04658), endocrine resistance (map01522), and steroid hormone biosynthesis (map00140). Upregulated symbiotic Symbiodiniaceae genes were enriched in endocytosis (map04144) and phagosome (map04145). Upregulated host genes of *A. pruinosa* were enriched in four pathways. The upregulated symbiotic Symbiodiniaceae

genes were enriched in one pathway: photosynthesis (map00195) (Fig. S7).

The upregulated host genes of *P. decussata* in winter were enriched in six pathways. The most notably enriched pathways were the dorso-ventral axis formation (map04320), Th1 and Th2 cell differentiation (map04658), endocrine resistance (map01522), and steroid hormone biosynthesis (map00140). Upregulated symbiotic Symbiodiniaceae genes were enriched in carbon fixation in photosynthetic organisms (map00710). The upregulated host genes of *A. pruinosa* were enriched in two pathways. The upregulated symbiotic Symbiodiniaceae genes were enriched in three pathways (Fig. S8).

Finally, we performed KEGG functional enrichment analysis on the DEGs shared by the two corals in summer and winter. The upregulated host genes of *A. pruinosa* were enriched in three pathways. Upregulated symbiotic Symbiodiniaceae genes were enriched in two pathways. The upregulated host genes of *P. decussata* were enriched in 10 pathways. Upregulated symbiotic Symbiodiniaceae genes were enriched in three pathways. We found that the two corals shared some key pathways in adaptation to both summer and winter environments. For example, the highly expressed host genes of *A. pruinosa* were significantly enriched in neuroactive ligand-receptor interaction (map04080), and symbiotic Symbiodiniaceae genes were significantly enriched in photosynthesis (map00195). *P. decussata* highly expressed host genes were significantly enriched in dorso-ventral axis formation (map04320), Th1 and Th2 cell differentiation (map04658), endocrine resistance (map01522), and steroid hormone biosynthesis (map00140). The upregulated genes of symbiotic Symbiodiniaceae were significantly enriched in endocytosis (map04144) and phagosome (map04145), which were found in *P. decussata* in summer (Fig. 4, Fig. S9).

4. Discussion

To reveal the interspecies differences in the adaptation strategies of marginal reefs to seasonal temperature fluctuations, we adopted a comprehensive approach to explore the regulatory role of coral host, symbiotic Symbiodiniaceae, and symbiotic bacteria in the differential tolerance of *A. pruinosa* and *P. decussata* in Weizhou Island, northern South China Sea.

4.1. Stable symbiotic relationship between coral and Symbiodiniaceae C1 helps to adapt to relatively high-latitude environments

The results in this study demonstrate that the symbiotic Symbiodiniaceae types of *P. decussata* and *A. pruinosa* were dominated by the C1 subgroup and were stable during the adaptation to the large seasonal temperature fluctuations in relatively high-latitude seas.

The symbiotic Symbiodiniaceae is an essential part of the coral holobiont and is important for its environmental adaptation (Little et al., 2004). Furthermore, specific symbiotic Symbiodiniaceae genotypes are typically the result of environmental selection and co-evolution (Chen et al., 2019). Differences between geographic locations generally have a greater impact on Symbiodiniaceae types than differences in hosts (Thornhill et al., 2017). Weizhou Island is located in the northern part of the South China Sea and belongs to the high-latitude marginal coral reef area in the Pacific Ocean. Scleractinian corals at high latitudes are considered more likely to form symbiotic relationships with *Cladocopium* (Chen et al., 2019). In particular, the C1 subgroup is associated with most coral species in high-latitude marginal corals of Jeju Island (South Korea) (De Palmas et al., 2015) and the temperate region (30°–35°N) of Japan (Lien et al., 2012).

Coral is in a balance between growth and survival, and the relatively high-latitude environment is one of the important driving factors of this balance. Long-term natural selection has driven the stable symbiotic relationship between corals and C1 at relatively high latitudes, to cause better growth in relatively high latitudes. The role of symbiotic

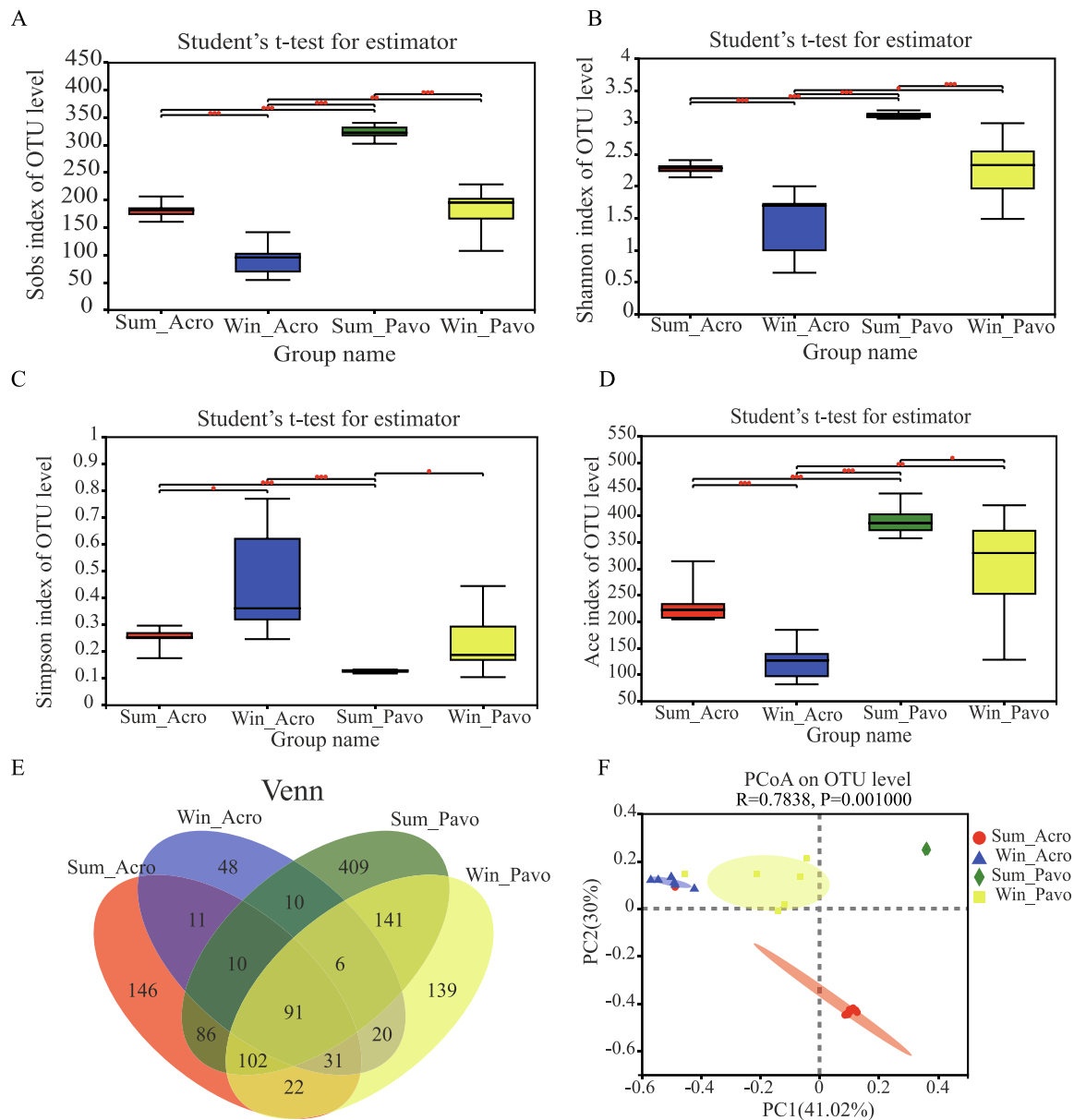


Fig. 1. Composition and diversity differences of each treatment on OUT lever. (A) Number of OTUs. (B) Alpha diversity of OTU level (Shannon Index). (C) Alpha diversity of OTU level (Simpson Index). (D) Ace index of OTU level. (E) Venn diagram of OTUs. (F) Principal Coordinates Analysis (PCoA) representing differences in community structure at the OTU level.

Symbiodiniaceae is also important in this process. Therefore, predicting how coral community members from different habitats and different environmental gradients respond to the increasing pressure under the background of global climate change is important. Under future climate conditions, the subtle phenotypic differences between symbiotic Symbiodiniaceae closely related to corals may significantly affect their diversity, richness, and community stability in the next few decades.

4.2. Increased symbiotic bacterial diversity and community reorganization contribute to seasonal adaptation

To reveal the regulatory mechanism of symbiotic bacteria in coral holobiont in response to seasonal temperature changes, we analyzed the differences in the diversity, community structure, and core microbiome of symbiotic bacteria between *A. pruinosa* and *P. decussata*. We found that α -diversity significantly differed between seasons, the symbiotic bacterial α -diversity of *P. decussata* was higher than that of *A. pruinosa* in both summer and winter, and the α -diversity of symbiotic bacteria in

summer samples of coral species was higher than that in winter samples. To further clarify the symbiotic bacteria in *A. pruinosa* and *P. decussata* in response to the large temperature difference between summer and winter in Weizhou Island, we combined the analysis of symbiotic bacterial community composition, LefSe differential species analysis, and core microbial communities analysis to screen the following potentially functional core symbiotic bacteria (over 10 % abundance within any group): BD1-7_clade, *Rhodococcus*, *Ralstonia*, *Prosthecochloris*, *Synechococcus*_CC9902, *Vibrio*, and *Acinetobacter*. The relative abundances of symbiotic bacteria BD1-7_clade, *Rhodococcus*, *Ralstonia*, and *Acinetobacter* were higher than those of *A. pruinosa* in summer in *P. decussata*, while those of *Prosthecochloris* and *Vibrio* were lower than those of *A. pruinosa*.

High abundance and α -diversity enable coral hosts to convert their predominant symbiotic bacteria into related species that perform better under specific conditions, and high α -diversity helps symbionts fight infection, absorb nutrients, and maintain the aggregation function of healthy microbiota (Flanagan et al., 2007; Pollock et al., 2019; Yu et al.,

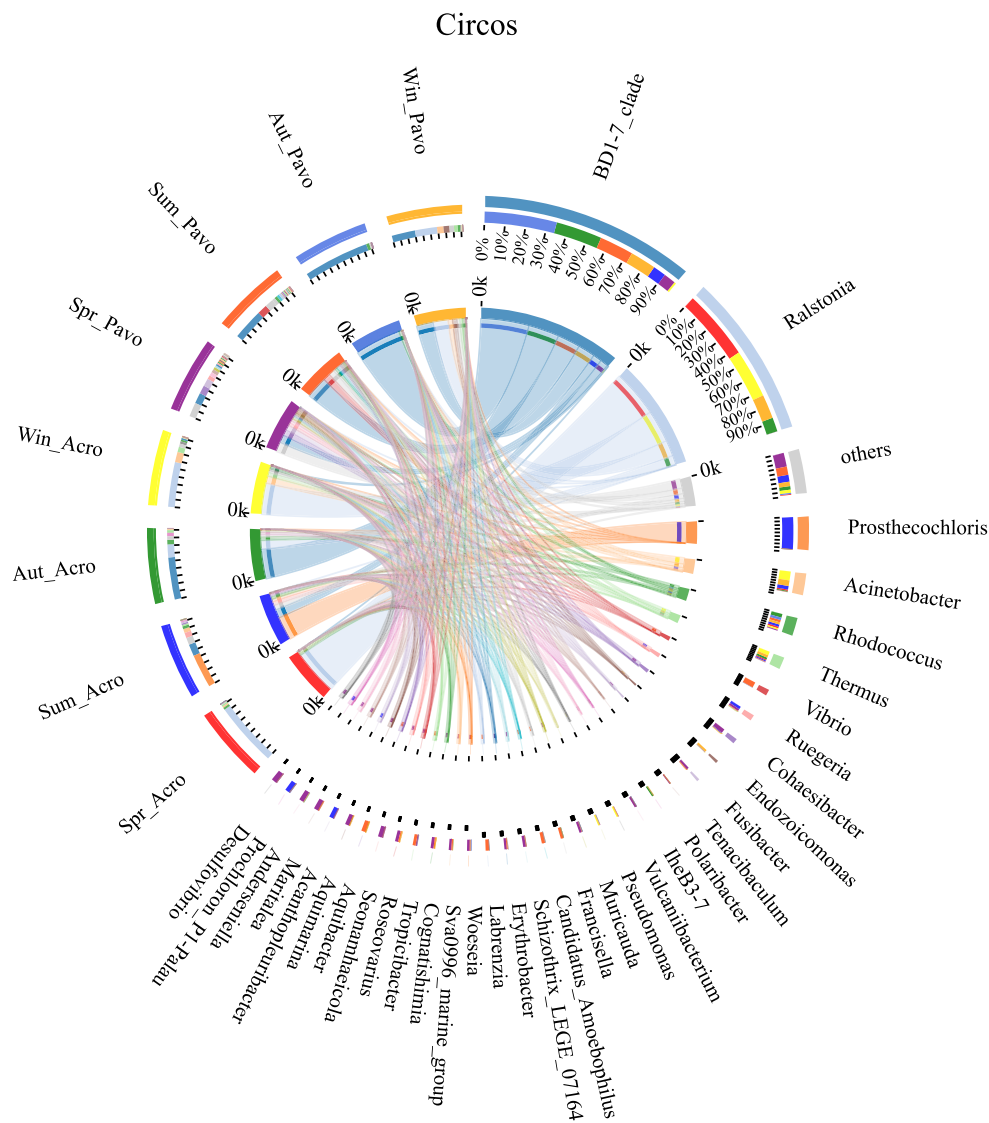


Fig. 2. Microbial community distribution of different groups at the genus level. Bar width indicates relative abundance in sample.

2020a, 2020b). This suggests that high α -diversity contributes to maintaining physiological functions throughout the organism, which may be associated with high tolerance.

In addition, the higher flexibility of bacterial community structure may contribute to better adaptation to the fluctuating reef area environment in the northern South China Sea. The dominant bacteria in summer were BD1-7 Clade, *Prosthecochloris*, and *Vibrio*. The dominant bacteria in winter were BD1-7 Clade, *Ralstonia*, and *Acinetobacter*. BD1-7_clade is a member of coral symbiotic bacteria *Spongiibacteraceae*, and it has been found in *Acropora muricata* (Doering et al., 2021). *Spongiibacteraceae* may be able to use the protein rhodopsin as an additional energy source for light (Spring et al., 2015), allowing it to take up increasingly important roles in global ocean carbon cycling and energy metabolism (Fenchel, 2001; Xie et al., 2021). *Prosthecochloris* is a potential nitrogen fixer and photoautotroph that may serve as a nitrogen and carbon source for coral holobionts (Yang et al., 2017). Previous studies have found that cyanobacteria exist in corals in Taiwan Green Island (Yang et al., 2017), Weizhou Island (Yu et al., 2021a, 2021b, 2021c), Luhuitou Marginal Reef (Li et al., 2013), and Hong Kong (Cai et al., 2018). Several studies also found very high relative abundances of the symbiotic *Prosthecochloris* of *A. pruinosa* at Weizhou Island (Yu et al., 2021a, 2021b, 2021c) and *G. fascicularis* and *Platygyra carnosa* in Hong Kong (Cai et al., 2018). Abiotic factors such as oxygen, temperature, and

light intensity may be decisive factors in determining the composition of symbiotic microorganisms (Yang et al., 2019). *Ralstonia* is a recognized minor symbiotic genus in corals and is closely related to dinoflagellate endosymbionts (D Ainsworth et al., 2015). It has been found in *Acropora granulosa* of the Coral Sea in northern Australia (D Ainsworth et al., 2015), endemic coral *Mussismilia hispida* of Brazilian (Leite et al., 2017), *A. pruinosa* in Weizhou Island (Yu et al., 2021a, 2021b, 2021c), and soft coral *Scleronephthya gracillimum* at the Kochi site (D Ainsworth et al., 2015). In addition, *Ralstonia* may have nitrogen fixation functions through a symbiotic relationship with the host (D Ainsworth et al., 2015) because ubiquitous subgroups are only conditionally less abundant in abundance and can grow under optimal conditions.

We found that in summer, the abundance of opportunistic bacteria in the symbiotic bacteria of *A. pruinosa* and *P. decussata* increased, but there were differences between the two corals, *Acinetobacter* and *Vibrio*, respectively. *Vibrio* is generally considered an opportunistic pathogen, and numerous studies have reported that *Vibrio* infection may lead to coral bleaching (Banin et al., 2003). *Acinetobacter* is also generally considered to be one of the important opportunistic pathogens. Moreover, it is symbiotic with scleractinian corals and can be transmitted vertically through mucus (Leite et al., 2017).

We also found differences in the two coral symbiotic bacteria during summer and winter. *Prosthecochloris* and *Synechococcus*_CC9902 may

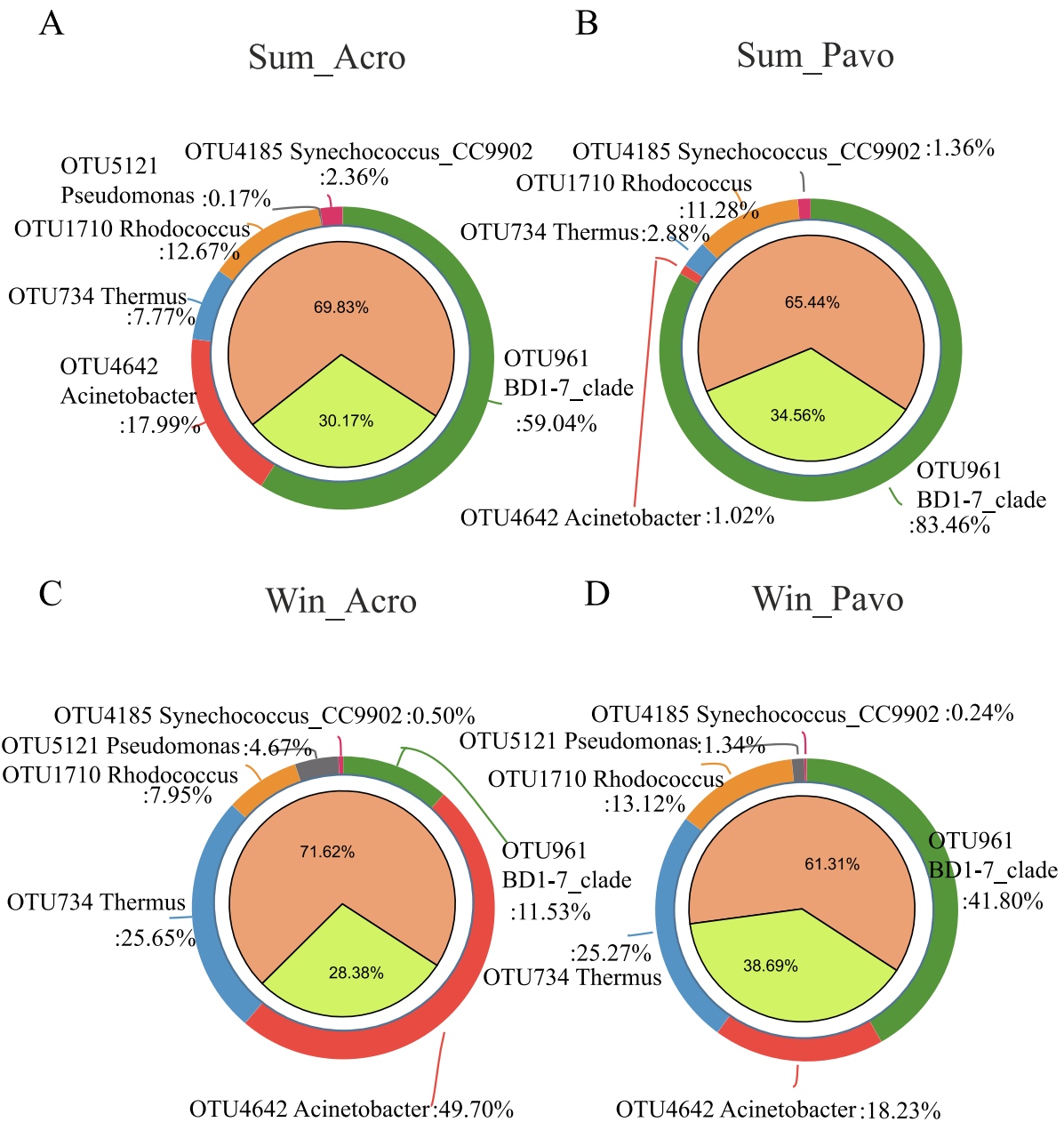


Fig. 3. The relative abundance and composition of the core bacterial microbiome. Internal pie charts demonstrating the proportion of core microbiome in coral bacterial community composition (green: core OTU, Orange: non core OTUs). The outer pie chart illustrates the core bacterial microbiome composition in the coral symbiotic functionalities. (A) Sum_Acro; (B) Sum_Pavo; (C) Win_Acro; (D) Win_Pavo.

provide nitrogen and carbon sources for *A. pruinosa*, and BD1-7_clade plays an increasingly important role in the global ocean carbon cycle and energy metabolism. *Rhodococcus* may help *P. decussata* resist pathogenic microbial invasion. The abundance of the symbiotic bacteria *Ralstonia* of *P. decussata* and *A. pruinosa* was relatively low in summer. In winter, *A. pruinosa* was mainly dominated by *Ralstonia*, followed by *Acinetobacter*, while *P. decussata* symbiotic bacteria BD1-7_clade and *Ralstonia* had the same abundance and dominated together, and the abundance of *Acinetobacter* was lower. *Ralstonia* may be involved in nitrogen fixation, and BD1-7_clade is involved in carbon cycling and energy metabolism. Therefore, we speculate that the symbiotic bacteria may have different roles in the energy metabolism of the two corals. The two types of corals may have different adaptation mechanisms to the lower sea temperature in relatively high latitudes. The psychrophilic marine bacterium *Rhodococcus* may help the two types of corals adapt to the low seawater temperature at Weizhou Island in winter but is more

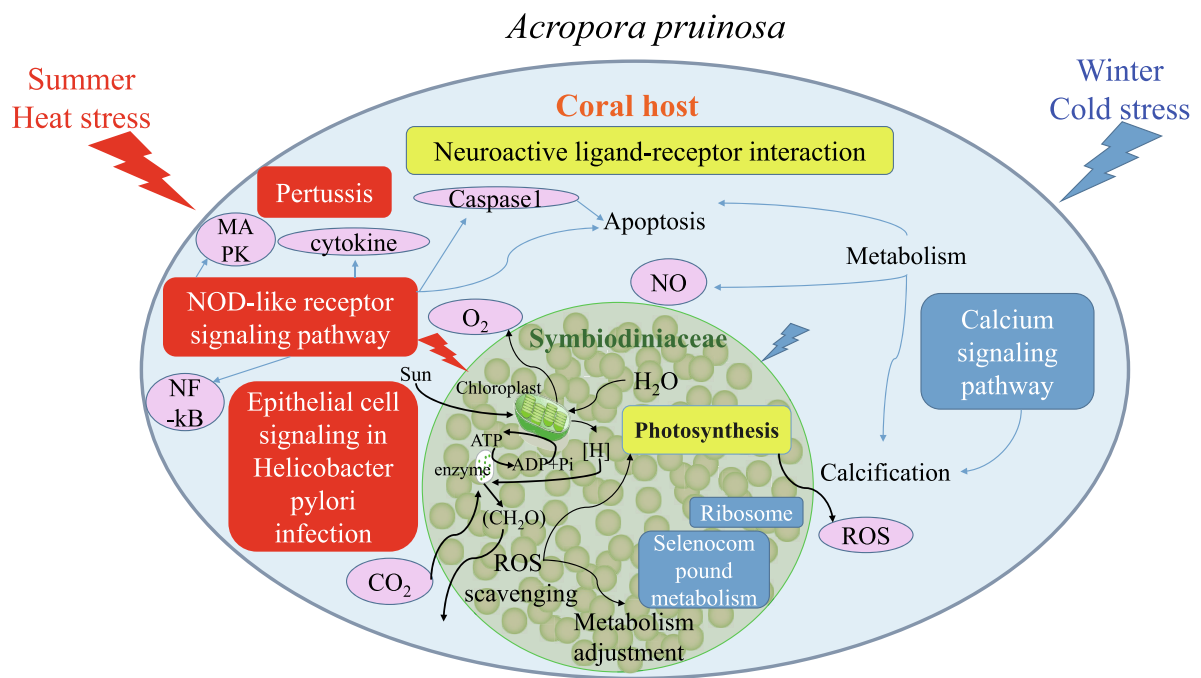
abundant in *P. decussata*. We also found that the relative abundances of *A. pruinosa* opportunistic bacteria *Vibrio* and *Acinetobacter* were significantly higher than those of *P. decussata*. This may be one of the reasons why *A. pruinosa* is more sensitive to environmental stress than *P. decussata*.

In summary, high α -diversity of symbiotic bacteria and high flexibility of bacterial community structure can improve adaptation to the fluctuating reef environment in the northern South China Sea.

4.3. Coral holobionts respond to seasonal stress by increasing energy supply to maintain a high level of immunity

In the present study, transcriptomes of *A. pruinosa* and *P. decussata* from Weizhou Island in different seasons were sequenced to understand their integrated transcriptional response to environmental stress and offer fundamental new insights into the adaptation of scleractinian

A



B

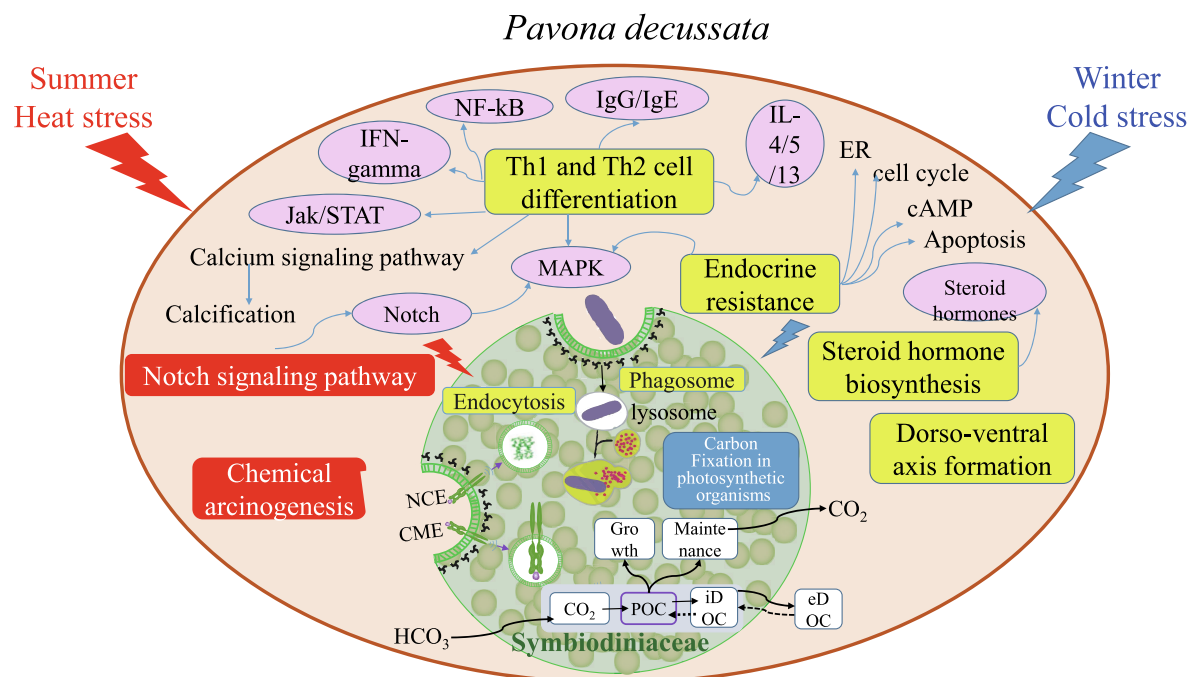


Fig. 4. Adaptation mechanism of *A. pruinosa* (A) and *P. decussata* (B) coral hosts, symbiotic Symbiodiniaceae to relative high latitude summer and winter stress.

corals to large seasonal temperature fluctuations in relatively high latitudes. The results showed that the two corals share some key pathways in adaptation to both summer and winter environments. For example, we found that the most enriched pathway of the *A. pruinosa* host was neuroactive ligand–receptor interaction (map04080), and for the symbiotic Symbiodiniaceae, it was photosynthesis (map00195).

Correspondingly, the enriched pathways of the *P. decussata* host were the immune-related pathways, such as dorso-ventral axis formation (map04320), Th1 and Th2 cell differentiation (map04658), endocrine resistance (map01522), and steroid hormone biosynthesis (map00140), and for the symbiotic Symbiodiniaceae, they were phagosome (map04145) and endocytosis (map04144).

The immune system is the provider of homeostasis, supporting the health and survival of coral holobionts, resisting destruction, and rebuilding homeostasis in the face of biological and abiotic disturbances. Therefore, a change in immunity may have a great impact on coral survival under the pressure of climate change. Large seasonal temperature fluctuations at relatively high latitudes produce an acute immune response. Higher immune levels help scleractinian corals adapt to environmental stress (Yu et al., 2020a, 2020b; Yu et al., 2021a, 2021b, 2021c), indicating that higher immune level is important in promoting coral response to high temperature and low temperature stress. In addition, in both summer and winter, coral symbiotic Symbiodiniaceae genes are enriched in energy supply related pathways, which can meet the high energy investment required for immune response to quickly eliminate threats. Previous studies on the symbiotic Symbiodiniaceae density of scleractinian coral in the South China Sea from May to August 2015 showed that the symbiotic Symbiodiniaceae densities of *P. decussata* in Weizhou Island are higher than that of *A. pruinosa* (Qin et al., 2019), and therefore, stronger photosynthesis may complement lower Symbiodiniaceae density to meet the nutritional needs of coral symbionts and meet the nutritional needs of coral holobionts of *A. pruinosa*. The upregulated genes of *P. decussata* symbiotic Symbiodiniaceae were significantly enriched in Endocytosis (map04144) and Phagosome (map04145), implying a stronger material exchange between the host and the symbiont. According to the theory of life history (Sheldon and Verhulst, 1996), coral's immune strategy is the result of a physiological trade-off that depends on the environment and has a plastic phenotype, which occurs within the predetermined limit of the evolutionary trade-off. The physiological cost that determines plasticity may occur during the maintenance or implementation of immunity. Coral homeostasis maintained by immunity may be directly related to energy availability and the ability to compensate costs. Therefore, the increase in energy supply of symbiotic Symbiodiniaceae to coral holobionts is helpful in meeting the energy demand at a high immune level.

We also found that there are interspecific differences in the adaptation mechanism of the *A. pruinosa* and *P. decussata* to relatively high-latitude coral reefs. Compared with *A. pruinosa*, *P. decussata* had a higher host immunity level when adapting to high temperature in summer and low temperature in winter at Weizhou Island. Coral with higher damage threshold, higher component immunity, large energy reserves, or the ability to compensate for depletion and autoimmune damage (for example, through eating and antioxidants) are more able to survive long-term disturbances such as bleaching events (Palmer, 2018). Therefore, for the *P. decussata* with a high immune level, there is a large buffer before the damage threshold is destroyed, and the pressure response may be delayed and not so serious. This may be the reason for the higher environmental tolerance of *P. decussata*.

Nevertheless, both *A. pruinosa* and *P. decussata* in summer were enriched in disease-related pathways. This is consistent with the increase in the abundance of opportunistic bacteria in the symbiotic bacteria of the two corals. Therefore, we speculated that the high temperature in summer had more serious damage to *A. pruinosa* and *P. decussata* than winter. With the increase of pressure level and duration of high temperature stress in summer, scleractinian coral will also reach the state of energy exhaustion. In this state, even the physiological mechanism of the tolerant whole organism can no longer make up for the damage. This indicates that the high temperature in summer is an important factor affecting scleractinian corals at Weizhou Island. Global warming has alleviated the inhibition of low temperature in winter on relatively high-latitude marginal reefs to some extent, but with the intensification of global warming and the increase of the frequency and duration of ocean heatwaves, relatively high-latitude coral reefs have been affected by high temperature in summer.

We speculate that coral holobionts respond to seasonal stress by increasing energy supply to maintain a high level of immunity. These adaptive strategies can be broadly defined as the increase of resistance after long-term exposure, which is a phenomenon of complete adaptive

immunity (Palmer, 2018). Coral has a certain plasticity under long-term periodic seasonal pressure, which can prevent future challenges. This is an attractive topic in this era of climate change and coral reef crisis (Palmer, 2018). Coral may adapt to more extreme conditions through immune memory or immune training, which is extremely important for the protection of coral reef ecosystem in the future (Milutinović and Kurtz, 2016). The mechanism of immune memory varies from organism to organism; however, it still must be clarified to a great extent, including how it relates to corals. Moreover, coral is still a challenge in revealing the adaptive process because of the synergistic effect of different symbionts. Therefore, further research is needed to gain understanding of symbiotic relationships involving coral.

5. Conclusions

In summary, we speculate that scleractinian corals have developed some plastic adaptation strategies under long-term periodic seasonal pressures. However, the high temperature in summer has severe harmful effects on scleractinian corals in relatively high-latitude seas and even affects the refuge function of relatively high-latitude seas. Scleractinian corals can improve their adaptability to seasonal temperature fluctuation stress by increasing energy supply to maintain high levels of immunity, increasing symbiotic bacterial α -diversity, and changing dominant bacteria. This study demonstrates the adaptation strategies of marginal reefs to seasonal temperature fluctuations. It provides new insights into the study of scleractinian coral adaptation and relatively high-latitude coral refuges in the context of future global warming and intensified MHW.

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.scitotenv.2023.166439>.

Data accessibility

Sequencing raw reads in this study were deposited into the NCBI sequence reads archive (SRA) database: PRJNA841798, PRJNA841791, PRJNA687160 and PRJNA841966.

CRediT authorship contribution statement

Xiaopeng Yu: Conceptualization, Methodology, Resources, Data curation, Visualization, Validation, Writing – original draft. **Kefu Yu:** Conceptualization, Resources, Methodology, Validation, Funding acquisition. **Zhiheng Liao:** Resources, Investigation, Writing – review & editing. **Biao Chen:** Software, Formal analysis, Visualization. **Zhenjun Qin:** Data curation, Validation, Formal analysis, Writing – review & editing. **Jiayuan Liang:** Project administration, Formal analysis, Writing – review & editing. **Xu Gao:** Methodology, Data curation.

Declaration of competing interest

We declare we have no competing interests.

Data availability

Data will be made available on request.

Acknowledgments

This work was supported by the National Natural Science Foundation of China (Nos. 42090041 and 42030502), the Guangxi Scientific Projects (No.AD17129063 and AA17204074).

References

- Arif, C., Daniels, C., Bayer, T., Banguera-Hinestroza, E., Barbrook, A., Howe, C.J., Lajeunesse, T.C., Voolstra, C.R., 2014. Assessing symbiodinium diversity in

- scleractinian corals via next-generation sequencing-based genotyping of the *its2* rdna region. *Mol. Ecol.* 23 (17), 4418–4433. <https://doi.org/10.1111/mec.12869>.
- Banin, E., Vassilakos, D., Orr, E., Martinez, R.J., Rosenberg, E., 2003. Superoxide dismutase is a virulence factor produced by the coral bleaching pathogen *Vibrio shiloi*. *Curr. Microbiol.* 46 (6), 418–422. <https://doi.org/10.1007/s00284-002-3912-5>.
- Bay, R.A., Palumbi, S.R., 2015. Rapid acclimation ability mediated by transcriptome changes in reef-building corals. *Genome Biol. Evol.* 7 (6), 1602–1612. <https://doi.org/10.1093/gbe/evv085>.
- Beger, M., Sommer, B., Harrison, P.L., Smith, S.D.A., Pandolfi, J.M., 2014. Conserving potential coral reef refuges at high latitudes. *Divers. Distrib.* 20 (3), 245–257. <https://doi.org/10.1111/ddi.12140>.
- Benjamini, Y., Hochberg, Y., 1995. Controlling the false discovery rate: a practical and powerful approach to multiple testing. *J. R. Stat. Soc. Ser. B Methodol.* 57 (1), 289–300. <https://doi.org/10.1111/j.2517-6161.1995.tb02031.x>.
- Cai, L., Zhou, G., Tong, H., Tian, R., Zhang, W., Ding, W., Liu, S., Huang, H., Qian, P., 2018. Season structures prokaryotic partners but not algal symbionts in subtropical hard corals. *Appl. Microbiol. Biotechnol.* 102 (11), 4963–4973. <https://doi.org/10.1007/s00253-018-8909-5>.
- Chen, B., Yu, K., Liang, J., Huang, W., Wang, G., Su, H., Qin, Z., Huang, X., Pan, Z., Luo, W., Luo, Y., Wang, Y., 2019. Latitudinal variation in the molecular diversity and community composition of symbiodiniaceae in coral from the South China Sea. *Front. Microbiol.* 10.
- Chen, B., Yu, K., Liao, Z., Yu, X., Qin, Z., Liang, J., Wang, G., Wu, Q., Jiang, L., 2021. Microbiome community and complexity indicate environmental gradient acclimatization and potential microbial interaction of endemic coral holobionts in the South China Sea. *Sci. Total Environ.* 765, 142690 <https://doi.org/10.1016/j.scitotenv.2020.142690>.
- D Ainsworth, T., Krause, L., Bridge, T., Torda, G., Raina, J., Zakrzewski, M., Gates, R.D., Padilla-Gamiño, J.L., Spalding, H.L., Smith, C., Woolsey, E.S., Bourne, D.G., Bongarts, P., Hoegh-Guldberg, O., Leggat, W., 2015. The coral core microbiome identifies rare bacterial taxa as ubiquitous endosymbionts. *ISME J.* 9 (10), 2261–2274. <https://doi.org/10.1038/ismej.2015.39>.
- Davis, K.L., McMahon, A., Correa, R.E., Santos, I.R., 2020. Calcification and organic productivity at the world's southernmost coral reef. *Mar. Chem.* 227, 103870 <https://doi.org/10.1016/j.marchem.2020.103870>.
- De Palmas, S., Denis, V., Ribas-Deulofeu, L., Loubeyres, M., Woo, S., Hwang, S.J., Song, J. I., Chen, C.A., 2015. Symbiodinium spp. associated with high-latitude scleractinian corals from Jeju Island, South Korea. *Coral Reefs* 34 (3), 919–925. <https://doi.org/10.1007/s00338-015-1286-y>.
- Désert, C., Duclos, M.J., Blavy, P., Lecerf, F., Moreews, F., Klopp, C., Aubry, M., Herault, F., Le Roy, P., Berri, C., Douaire, M., Diot, C., Lagarrigue, S., 2008. Transcriptome profiling of the feeding-to-fasting transition in chicken liver. *BMC Genomics* 9 (1), 611. <https://doi.org/10.1186/1471-2164-9-611>.
- Doering, T., Wall, M., Putcham, L., Rattanawongwan, T., Schroeder, R., Hentschel, U., Roik, A., 2021. Towards enhancing coral heat tolerance: a “microbiome transplantation” treatment using inoculations of homogenized coral tissues. *Microbiome* 9 (1), 102. <https://doi.org/10.1186/s40168-021-01053-6>.
- Fenchel, T., 2001. Marine bugs and carbon flow. *Science* 292 (5526), 2444–2445. <https://doi.org/10.1126/science.1062799>.
- Feng, Y., Bethel, B.J., Dong, C., Zhao, H., Yao, Y., Yu, Y., 2022. Marine heatwave events near weizhou island, beibu gulf in 2020 and their possible relations to coral bleaching. *Sci. Total Environ.* 823, 153414 <https://doi.org/10.1016/j.scitotenv.2022.153414>.
- Flanagan, J.L., Brodie, E.L., Weng, L., Lynch, S.V., Garcia, O., Brown, R., Hugenholtz, P., DeSantis, T.Z., Andersen, G.L., Wiener-Kronish, J.P., et al., 2007. Loss of bacterial diversity during antibiotic treatment of intubated patients colonized with *Pseudomonas aeruginosa*. *J. Clin. Microbiol.* 45, 1954–1962. <https://doi.org/10.1128/JCM.02187-06>.
- Goyen, S., Camp, E.F., Fujise, L., Lloyd, A., Nitschke, M.R., Lajeunesse, T., Kahlke, T., Ralph, P.J., Suggett, D., 2019. Mass coral bleaching of *P. Versipora* in Sydney harbour driven by the 2015–2016 heatwave. *Coral Reefs* 38 (4), 815–830. <https://doi.org/10.1007/s00338-019-01797-6>.
- Hernandez-Agreda, A., Gates, R.D., Ainsworth, T.D., 2017. Defining the core microbiome in corals' microbial soup. *Trends Microbiol.* 25 (2), 125–140. <https://doi.org/10.1016/j.tim.2016.11.003>.
- Hongo, C., Yamano, H., 2013. Species-specific responses of corals to bleaching events on anthropogenically turbid reefs on Okinawa island, Japan, over a 15-year period (1995–2009). *PLoS One* 8 (4), e60952.
- Jiang, L., Yu, K., Han, T., Tao, S., Zhang, H., 2021. Coral perspective on temperature seasonality and interannual variability in the northern South China Sea during the roman warm period. *Glob. Planet. Chang.* 207, 103675 <https://doi.org/10.1016/j.gloplacha.2021.103675>.
- Leite, D.C.A., Leão, P., Garrido, A.G., Lins, U., Santos, H.F., Pires, D.O., Castro, C.B., van Elsas, J.D., Zilberberg, C., Rosado, A.S., Peixoto, R.S., 2017. Broadcast spawning coral *Mussismilia hispida* can vertically transfer its associated bacterial core. *Front. Microbiol.* 8.
- Li, J., Chen, Q., Zhang, S., Huang, H., Yang, J., Tian, X., Long, L., 2013. Highly heterogeneous bacterial communities associated with the South China Sea reef corals *Porites lutea*, *Galaxea fascicularis* and *Acropora millepora*. *PLoS One* 8 (8), e71301.
- Liang, J., Yu, K., Wang, Y., Huang, X., Huang, W., Qin, Z., Pan, Z., Yao, Q., Wang, W., Wu, Z., 2017. Distinct bacterial communities associated with massive and branching scleractinian corals and potential linkages to coral susceptibility to thermal or cold stress. *Front. Microbiol.* 8, 979. <https://doi.org/10.3389/fmicb.2017.00979>.
- Lien, Y.T., Fukami, H., Yamashita, Y., 2012. Symbiodinium clade C dominates zooxanthellate corals (Scleractinia) in the temperate region of Japan. *Zool. Sci.* 29 (3), 173–180. <https://doi.org/10.2108/zsj.29.173>.
- Little, A.F., Oppen, M.J., Willis, B.L., 2004. Flexibility in algal endosymbioses shapes growth in reef corals. *Science* 304, 1492–1494. <https://doi.org/10.1126/science.1095733>.
- Love, M.I., Huber, W., Anders, S., 2014. Moderated estimation of fold change and dispersion for RNA-seq data with DESeq2. *Genome Biol.* 15 (12), 550. <https://doi.org/10.1186/s13059-014-0550-8>.
- Mansour, T.A., Rosenthal, J.J.C., Brown, C.T., Roberson, L.M., 2016. Transcriptome of the Caribbean stony coral *Porites astreoides* from three developmental stages. *Gigascience* 5 (1), 33. <https://doi.org/10.1186/s13742-016-0138-1>.
- Milutinović, B., Kurtz, J., 2016. Immune memory in invertebrates. *Semin. Immunol.* 28 (4), 328–342. <https://doi.org/10.1016/j.smim.2016.05.004>.
- Mo, S., Chen, T., Chen, Z., Zhang, W., Li, S., 2022. Marine heatwaves impair the thermal refugia potential of marginal reefs in the northern South China Sea. *Sci. Total Environ.* 825, 154100 <https://doi.org/10.1016/j.scitotenv.2022.154100>.
- Palmer, C.V., 2018. Immunity and the coral crisis. *Commun. Biol.* 1 (1), 91. <https://doi.org/10.1038/s42003-018-0097-4>.
- Pollock, F.J., Lamb, J.B., van de Water, J.A.J.M., Smith, H.A., Schaffelke, B., Willis, B.L., Bourne, D.G., 2019. Reduced diversity and stability of coral-associated bacterial communities and suppressed immune function precedes disease onset in corals. *R. Soc. Open Sci.* 6 (6), 190355 <https://doi.org/10.1098/rsos.190355>.
- Qin, Z., Yu, K., Wang, Y., Xu, L., Huang, X., Chen, B., Li, Y., Wang, W., Pan, Z., 2019. Spatial and intergenerative variation in physiological indicators of corals in the South China Sea: insights into their current state and their adaptability to environmental stress. *J. Geophys. Res. Oceans* 124, 3317–3332. <https://doi.org/10.1029/2018JC014648>.
- Sheldon, B.C., Verhulst, S., 1996. Ecological immunology: costly parasite defences and trade-offs in evolutionary ecology. *Trends Ecol. Evol.* 11 (8), 317–321. [https://doi.org/10.1016/0169-5347\(96\)10039-2](https://doi.org/10.1016/0169-5347(96)10039-2).
- Spring, S., Scheuner, C., Göker, M., Klenk, H., 2015. A taxonomic framework for emerging groups of ecologically important marine gamma-proteobacteria based on the reconstruction of evolutionary relationships using genome-scale data. *Front. Microbiol.* 6.
- Thornhill, D.J., Howells, E.J., Wham, D.C., Steury, T.D., Santos, S.R., 2017. Population genetics of reef coral endosymbionts (*Symbiodinium*, *Dinophyceae*). *Mol. Ecol.* 26 (10), 2640–2659. <https://doi.org/10.1111/mec.14055>.
- Vajedsamiei, J., Saleh, A., Shirvani, A., Kayal, M., 2015. Photosynthetic response of Persian gulf acroporid corals to summer versus winter temperature deviations. *PeerJ* 3. <https://doi.org/10.7717/peerj.1062>.
- Xie, H., Chen, J., Feng, L., He, L., Zhou, C., Hong, P., Sun, S., Zhao, H., Liang, Y., Ren, L., Zhang, Y., Li, C., 2021. Chemotaxis-selective colonization of mangrove rhizosphere microbes on nine different microplastics. *Sci. Total Environ.* 752, 142223 <https://doi.org/10.1016/j.scitotenv.2020.142223>.
- Xu, L., Yu, K., Li, S., Liu, G., Tao, S., Shi, Q., Chen, T., Zhang, H., 2017. Interseasonal and interspecies diversities of symbiodinium density and effective photochemical efficiency in five dominant reef coral species from Luhuitou fringing reef, northern South China Sea. *Coral Reefs* 36 (2), 477–487. <https://doi.org/10.1007/s00338-016-1532-y>.
- Xu, S., Men, Z., Yu, K., Chen, H., Mo, H., Chen, B., Qin, Z., 2022. Interspecific differences in the response of coral trophic status to the decrease in symbiotic zooxanthellae in summer. *Mar. Ecol. Prog. Ser.* 694, 73–87.
- Yang, S., Tseng, C., Huang, C., Chen, C., Tandon, K., Lee, S.T.M., Chiang, P., Shiu, J., Chen, C.A., Tang, S., 2017. Long-term survey is necessary to reveal various shifts of microbial composition in corals. *Front. Microbiol.* 8.
- Yang, S., Tandon, K., Lu, C., Wada, N., Shih, C., Hsiao, S.S., Hsiao, J., Lee, W., Lee, T., Yang, C., Liu, C., Denis, V., Wu, Y., Wang, L., Huang, L., Lee, D., Wu, Y., Yamashiro, H., Tang, S., 2019. Metagenomic, phylogenetic, and functional characterization of predominant endolithic green sulfur bacteria in the coral *Isopora palifera*. *Microbiome* 7, 3.
- Yao, Y., Wang, C., 2022. Marine heatwaves and cold-spells in global coral reef zones. *Prog. Oceanogr.* 209, 102920 <https://doi.org/10.1016/j.pocean.2022.102920>.
- Yu, K., Zhao, J., Wei, G., Cheng, X., Wang, P., 2005. Mid-late holocene monsoon climate retrieved from seasonal $\delta^{18}O$ and $\delta^{18}O$ records of *Porites lutea* corals at Leizhou peninsula, northern coast of South China Sea. *Glob. Planet. Chang.* 47 (2), 301–316. <https://doi.org/10.1016/j.gloplacha.2004.10.018>.
- Yu, W., Wang, W., Yu, K., Wang, Y., Huang, X., Huang, R., Liao, Z., Xu, S., Chen, X., 2019. Rapid decline of a relatively high latitude coral assemblage at weizhou island, northern South China Sea. *Biodivers. Conserv.* 28 (14), 3925–3949. <https://doi.org/10.1007/s10531-019-01858-w>.
- Yu, X., Yu, K., Huang, W., Liang, J., Qin, Z., Chen, B., Yao, Q., Liao, Z., 2020a. Thermal acclimation increases heat tolerance of the scleractinian coral *Acropora pruinosa*. *Sci. Total Environ.* 733, 139319 <https://doi.org/10.1016/j.scitotenv.2020.139319>.
- Yu, X., Yu, K., Liao, Z., Liang, J., Deng, C., Huang, W., Huang, Y., 2020b. Potential molecular traits underlying environmental tolerance of *Pavona decussata* and *Acropora pruinosa* in weizhou island, northern South China Sea. *Mar. Pollut. Bull.* 156, 111199 <https://doi.org/10.1016/j.marpolbul.2020.111199>.
- Yu, X., Yu, K., Chen, B., Liao, Z., Liang, J., Yao, Q., Qin, Z., Wang, H., Yu, J., 2021a. Different responses of scleractinian coral *Acropora pruinosa* from weizhou island during extreme high temperature events. *Coral Reefs* 40 (6), 1697–1711. <https://doi.org/10.1007/s00338-021-02182-y>.
- Yu, X., Yu, K., Chen, B., Liao, Z., Qin, Z., Yao, Q., Huang, Y., Liang, J., Huang, W., 2021b. Nanopore long-read rDNA reveals regulatory mechanisms of thermally variable reef environments promoting heat tolerance of scleractinian coral *Pocillopora damicornis*. *Environ. Res.* 195, 110782 <https://doi.org/10.1016/j.envres.2021.110782>.

- Yu, X., Yu, K., Liao, Z., Chen, B., Deng, C., Yu, J., Yao, Q., Qin, Z., Liang, J., 2021c. Seasonal fluctuations in symbiotic bacteria and their role in environmental adaptation of the scleractinian coral *Acropora pruinosa* in high-latitude coral reef area of the South China Sea. *Sci. Total Environ.* 792, 148438 <https://doi.org/10.1016/j.scitotenv.2021.148438>.
- Zaneveld, J.R., Mcminds, R., Vega Thurber, R., 2017. Stress and stability: applying the anna karenina principle to animal microbiomes. *Nat. Microbiol.* 2 (9), 17121. <https://doi.org/10.1038/nmicrobiol.2017.121>.
- Zhang, W., Zheng, Z., Zhang, T., Chen, T., 2020. Strengthened marine heatwaves over the beibu gulf coral reef regions from 1960 to 2017. *Haiyang Xuebao* 5 (42), 41–48. <https://doi.org/10.3969/j.issn.0253-4193.2020.05.005>.
- Zhou, P., Yan, H., Shi, G., Liu, C., Luo, F., Han, T., Wang, G., Wen, H., Zhao, N., Dodson, J., Li, Y., Zhou, W., 2022. Sea surface temperature seasonality in the northern South China Sea during the middle holocene derived from high resolution sr/ca ratios of tridacna shells. *Quat. Res.* 105, 37–48. <https://doi.org/10.1017/qua.2021.28>.
- Ziegler, M., Arif, C., Burt, J.A., Dobretsov, S., Roder, C., Lajeunesse, T.C., Voolstra, C.R., 2017. Biogeography and molecular diversity of coral symbionts in the genus symbiodinium around the arabian peninsula. *J. Biogeogr.* 44 (3), 674–686. <https://doi.org/10.1111/jbi.12913>.
- Ziegler, M., Grupstra, C.G.B., Barreto, M.M., Eaton, M., Baomar, J., Zubier, K., Al-Sofyani, A., Turki, A.J., Ormond, R., Voolstra, C.R., 2019. Coral bacterial community structure responds to environmental change in a host-specific manner. *Nat. Commun.* 10 (1), 3092. <https://doi.org/10.1038/s41467-019-10969-5>.