

Biogeographical diet variation within and between the rabbitfishes *Siganus corallinus*, *Siganus virgatus*, *Siganus doliatus* and the endemic *Siganus trispilos*.

Zarco-Perello S.^{1, 2,*}, Martin S.¹, Hoey A.³

¹ Harry Butler Institute, Murdoch University, Perth, Australia.

² College of Life Sciences and Agriculture, University of New Hampshire, Durham, USA.

³ ARC Centre of Excellence for Coral Reef Studies and College of Science and Engineering, James Cook University, Townsville, Queensland, Australia

* Corresponding author: salvador.zarco.perello@gmail.com

Abstract

Feeding habits of herbivorous fish play an important role in the form and function of coastal marine ecosystems. Rabbitfishes (Siganidae) have been recognized as important consumers of macroalgae in the coral reefs of the Indo-Pacific region. However, it is unclear how their diet varies among and within species at biogeographical scales. The present study assessed the inter- and intra-specific diet variation of rabbitfishes (*Siganus trispilos*, *Siganus corallinus*, *Siganus virgatus*, and *Siganus doliatus*) factored by phylogenetic and morphological relatedness among populations from Ningaloo (western Australia), the Great Barrier Reef (GBR, eastern Australia) and Okinawa (Japan). Results showed a strong effect in diet by geographic distribution, effectively reducing the expected effect of phylogenetic and morphologic relatedness. While intra-specific differences were only significant when populations inhabited different regions as expected; inter-specific differences were not as predicted, with different morphotypes (non-sister species) having similar diets when populations inhabited the same regions. Differences were driven by higher consumption of corticated and filamentous macroalgae by populations from the GBR, higher consumption of foliose and membranous macroalgae by rabbitfishes in Okinawan reefs and higher diet proportions of leathery macroalgae in populations from Ningaloo reefs. The findings indicate that rabbitfishes possess a relatively high diet plasticity, potentially driven by regional differences in algal resources, and hence their functional role as mediators of competition between macroalgae and corals can change across biogeographic regions. This highlights the importance of considering local context when assessing the diet of herbivorous fishes to know their functional role in the ecosystem accurately. As climate change unfolds, shifts in the distribution, trophic behaviour and function of species are expected, making the study of trophic plasticity more crucial.

Keywords: trophic niche, trophic plasticity, herbivory, coral reefs, intraspecific variation, interspecific variation

Introduction

Primary consumption is an important ecological process that greatly influences energy flow and habitat structure in natural ecosystems. Herbivores link primary producers and higher-order consumers, initiating the transfer of energy across the trophic network and sustaining directly and indirectly the secondary productivity of higher trophic levels (Zarco-Perello *et al.* 2019). In instances of high abundances, herbivores can consume the majority of primary productivity, and influence the standing biomass and composition of benthic communities (Spadaro & Butler 2021). In coral reefs, fish species of the families Acanthuridae (surgeonfishes), Scarinae (parrotfishes) and Siganidae (rabbitfishes) are conspicuous and abundant consumers of macroalgae, and have been suggested to act as ecosystem engineers (Steneck, Bellwood & Hay 2017)(Clements *et al.* 2017). It is generally theorized that healthy communities of herbivorous fish strengthen the resilience of coral dominated states by preventing phase-shifts to macroalgal-dominated states following disturbances such as hurricanes or high temperatures that cause mass coral bleaching (Cheal *et al.* 2013); however, this will be highly dependent on the identity of the macroalgae and herbivorous fish species present in each reef system (Bellwood, Hughes & Hoey 2006; Puk *et al.* 2020). Inter-species comparisons of diet and feeding behaviour are widespread among all the most important families of herbivorous fish, delineating functional diversity and redundancy within fish communities (Johansson *et al.* 2013; Kelly *et al.* 2016). Species that feed importantly on leathery macroalgae (browsers) can shorten canopy height, enhancing light penetration to the benthos, and reduce physical damage on coral colonies (McCook, Jompa & Diaz-Pulido 2001), while consumers of short foliose and filamentous macroalgae (grazers) can clear benthic space and potentially facilitate coral recruitment (Korzen, Israel & Abelson 2011). Generally, studies have shown that while there are many species of herbivores that are grazers, only a few species have been identified as important browsers in each system of study (Puk, Ferse & Wild 2016).

The specific diet of different herbivorous fishes has been linked strongly to the morphological adaptations of specific clades of species. Parrotfishes are clearly distinguished from other families by its beak-like fused teeth that allows them to not only consume turf but also bite deep into the substrate and scoop sediment, detritus, and calcium carbonate; explaining the lack of species that feed on fleshy macroalgae, with a few exceptions in the genus *Sparisoma* spp., *Calatomus* spp. and *Leptoscarus* spp. (Bonaldo, Hoey & Bellwood 2014). Surgeonfishes present a higher diversity of morpho-functional adaptations for feeding specializations. For

instance, consumers of large leathery macroalgae are essentially restricted to the genus *Naso* spp. who have teeth adapted to perforate and rip pieces of the macroalgae thallus, while consumers of detritus and filamentous macroalgae are grouped in the genus *Ctenochaetus* spp. who evolved brush-like teeth which allows them to collect particulate material within turf mats (Tebbett, Siqueira & Bellwood 2022). Rabbitfishes are a particular case because all species are grouped within the genus *Siganus* spp. However, morphological and behavioural adaptations differentiate species in their trophic ecology. For instance, flat-snouted species such as *Siganus canaliculatus* and *Siganus virgatus* have been identified as the most important consumers of leathery macroalgae in Orpheus Island on the Great Barrier Reef (GBR; (Fox & Bellwood 2008; Bennett & Bellwood 2011), and coral reefs of the Indo-Pacific region respectively (Plass-Johnson *et al.* 2015; Bauman *et al.* 2017; Seah, Bauman & Todd 2021; Müller *et al.* 2021). On the other hand, species with more slender bodies and elongated snouts, such as *Siganus corallinus*, or *Siganus vulpinus*, have been identified as important consumers of filamentous algae in open and cryptic reef spaces, allowing them to exploit unique trophic resources in the ecosystem (Fox & Bellwood 2013; Brandl & Bellwood 2014).

Macroalgae consumption may not only depend on species identity, but also according to the environment where different populations reside. Species are regularly classified in trophic guilds, but feeding behaviour and prey items can change depending on the environmental and biological factors in different locations. Among fish, species of different trophic levels have shown feeding plasticity across space (Hamilton *et al.* 2011). Even species considered to have specialized diets have shown diet plasticity. Such as the corallivorous butterflyfish *Chaetodon octofasciatus*, who's populations can differ significantly in the number of coral species eaten and number of bites taken in each of them (Feary *et al.* 2018). Herbivorous species most likely are not the exception; however, this has been rarely assessed around the globe, particularly at large biogeographical scales. Among the few studies, the parrotfishes *Sparisoma viridae* and *Scarus vetula* have shown significant variation in bite rates among populations in coral reefs of Barbuda, Antigua and Bonaire (Wilson *et al.* 2021), and populations of *Leptoscarus vaigiensis* differed in diet diversity and breadth across coral reefs of Kenya (Locham *et al.* 2015), while the surgeonfishes *Acanthurus tractus* and *Acanthurus coeruleus* had different proportions of bites taken on different food items, such as turf, epiphytes, or sessile invertebrates among coral reefs of the Florida Keys (Duran *et al.* 2019). The spatial variability in the trophic ecology of rabbitfishes has only been assessed in the Great Barrier Reef, where diet analyses indicated clear inter-specific differences, however, a formal assessment of their intra-specific variability has not been done (Hoey, Brandl & Bellwood 2013). Thus, to-date, it is unclear how their consumption of macroalgae, and their functional role, can vary among and within species in different locations at large biogeographical scales. Addressing this

knowledge gap is crucial because coral reef ecosystems are managed at local level and it's important to understand the spatial variability of the ecological processes related to their stability. If herbivorous species change their feeding activity dramatically from one region of the world to the other, their functional role will change as well, impacting the capacity of the reef to suppress or recover from changes in the benthic community.

The present study aimed to shed light into the inter- and intra-specific diet variation factored by phylogenetic and morphological relatedness and biogeographic distribution by comparing populations of closely related species of rabbitfishes (*Siganus trispilos*, *Siganus coralinus*, *Siganus virgatus*, and *Siganus doliatus*) within the same regions and separated by thousands of kilometers: Ningaloo (western Australia), the Great Barrier Reef (GBR, eastern Australia) and Okinawa (Japan). We hypothesized that: (1) phylogenetically and morphologically similar species should have more similar diets and functional roles; however (2) intra- and inter-specific differences will increase between populations of different biogeographic regions, since they will likely face different physical and biological environments (Fig. 1).

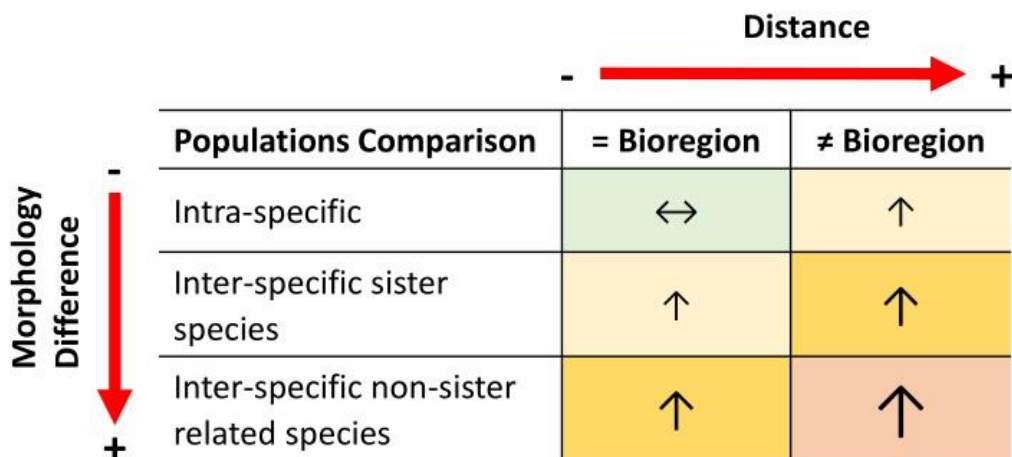


Figure 1. Hypothesized magnitudes of dissimilitude (size of arrows) in diet within (intra-specific) and among (inter-specific) species of rabbitfish as a factor of shared geographic distribution and phylogenetic relatedness (sister vs non-sister species).

Methods

Locations and species of study

The study focused on four closely-related species of rabbitfish (*Siganus virgatus*, *S. doliatus*, *S. corallinus*, and *S. trispilos*;(Siqueira, Bellwood & Cowman 2019) that have populations in three regions set apart by thousands of kilometres: Okinawa (Yaeyama Islands), the Great Barrier Reef (GBR; Turtle Group and Lizard Island), and Ningaloo Reef (Coral Bay; Fig. 2). *Siganus virgatus* and *S. doliatus* are sister species (Siqueira, Bellwood & Cowman 2019) that are distributed across the Indo-West Pacific and Western Pacific, respectively. *Siganus corallinus* and *S. trispilos* are very likely sister species (Woodland & Allen 1977), although their relatedness is yet to be confirmed by genetic analyses. *Siganus virgatus* and *S. doliatus* have regularly been observed to take bites from assays of the macroalga *Sargassum* (e.g., (Fox & Bellwood 2013); although whether they are targeting the *Sargassum* itself or epibiota is unknown. *S. corallinus* is distributed across the Indo-West Pacific and has been described as an “algal cropper” (Hoey *et al.* 2013); however, *S. trispilos* is endemic of northwestern Australia and his diet has not yet been described, but we hypothesize that it should be functionally similar to its sister species (Fox & Bellwood 2013).

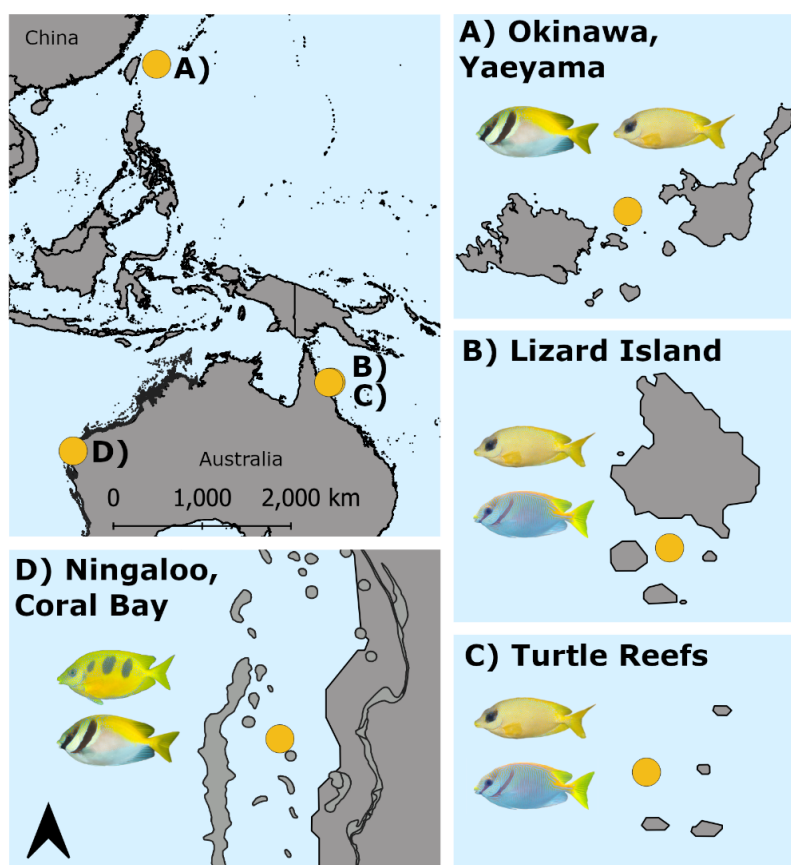


Figure 2. Locations in the world (Ningaloo, Okinawa, and inner-shelf (Turtle Reefs) and mid-shelf reefs (Lizard Island) of the Great Barrier Reef) and rabbitfish populations compared in the study: *Siganus corallinus* (fully yellow), *Siganus trispilos* (yellow with black blotches), *Siganus virgatus* (black facial bands), and *Siganus doliatus* (subtle facial bands).

Diet information

Specimens of *S. virgatus* and *S. trispilos* were collected by spear (Murdoch University ethics permit number R3349/21 and Department of Fisheries (DPIRD) exemption 3699) between 9 am to 11 am in backreef and lagoon habitats of Yalobia, Bateman and Five Fingers reefs during June 2022 and March-April 2023 (Table 1). Captured specimens were euthanized, stored in ice, and transferred to the research station of Murdoch University in Coral Bay, where their alimentary tract was removed, and stomach content preserved in a solution of 80% ethanol. Diet analyses of these samples were performed in the laboratory using a stereo microscope similarly to previous studies (Nanami 2018), where food items were spread evenly over a petri dish divided into a grid of 100 cells, identified to the lowest taxonomic level and assigned a proportion of the total gut content based on the proportional number of grid squares covered using the program ImageJ (rsb.info.nih.gov/ij/). Macroalgae in the diet were grouped into five morpho-functional groups: Foliose, membranous, corticated, filamentous, and leathery macroalgae, as well as seagrass, cyanobacteria, sessile invertebrates, and detritus. For the populations of rabbitfishes in the GBR, we used a diet database created in a previous study by (Hoey *et al.* 2013); while the diet of populations from Okinawa were obtained from the study of (Nanami 2018) using the software Datathief (Flower, McKenna & Upreti 2016); Table 1)

Table 1. Number of specimens and body size of each species and populations of rabbitfish included in the present study. N: number of specimens collected. GBR: Great Barrier Reef.

Species	Region	N	Fork Length (mm)	Reference
<i>Siganus corallinus</i>	Okinawa, Yaeyama Islands	10	126 – 205	(Nanami 2018)
<i>Siganus virgatus</i>		10	159 – 215	
<i>Siganus corallinus</i>	GBR mid-shelf (Lizard Island)	22	183 – 234	(Hoey <i>et al.</i> 2013)
	GBR inner-shlef (Turtle Group)	6		
<i>Siganus doliatus</i>	GBR, Lizard Island	20	165 – 250	
	GBR, Turtle Group	11		
<i>Siganus trispilos</i>	Ningaloo, Coral Bay	7	217 – 269	This study
<i>Siganus virgatus</i>		6	220 - 257	

Statistical Analysis

Differences in diet between species and populations across regions were analyzed with Non-metric Multidimensional Scaling (nmMDS) based on Bray-Curtis distances using the functions *vegdist* and *metaMDS* of the R package *vegan* (Oksanen *et al.* 2022). Statistical differences were tested with permutational analysis of variance (PERMANOVA), considering “Region” and “Taxa” with sister species nested in 2 levels (*S. virgatus* + *S. doliatus* vs *S. trispilos* + *S. corallinus*) as fixed factors, using the function *adonis2* of the R package *vegan* (Oksanen *et al.* 2022), followed by pairwise comparisons among populations using the function *pairwise.adonis2* of the R package *pairwiseAdonis* (Martinez 2017).

Results

Interspecific differences among and within regions

The diets of rabbitfishes differed among species and regions, with most of the variance being explained by geographic distribution (PERMANOVA; Region: pseudo- $F_{3, 83} = 27.0833$, $P = 0.0001$; Taxa: pseudo- $F_{1, 83} = 4.9176$, $P = 0.0001$; Species: pseudo- $F_{2, 83} = 3.0200$, $P = 0.0138$; Table S1; Table S2). *S. corallinus* had statistically significant different diets from all other species of rabbitfishes, except between *S. corallinus* from Turtle Reefs and *S. doliatus* from Lizard Island, both within the Great Barrier Reef (GBR), and between *S. corallinus* and *S. virgatus* within Okinawan reefs. *S. corallinus* and *S. doliatus* from the GBR had very similar diets dominated by corticated and filamentous algae ($63.2 \pm 3.4\%$, mean \pm se) followed by foliose and membranous macroalgae ($24.0 \pm 3.2\%$), whereas both Okinawan rabbitfishes also had similar diets but with an inverse pattern, having higher proportions of foliose and membranous macroalgae ($\sim 60.1 \pm 4.3\%$), and less corticated and filamentous algae ($\sim 27.4 \pm 2.8\%$; Fig. 3). *S. trispilos* had a diet with equal proportions of foliose/membranous ($41.3 \pm 5.1\%$) and corticated/filamentous algae ($45.6 \pm 9.1\%$), followed by leathery macroalgae ($13.1 \pm 4.4\%$; Fig. 3), making it significantly different from all the other rabbitfishes with the exception of the population of *S. virgatus* from Ningaloo, which also had high proportion of leathery macroalgae ($33.6 \pm 6.8\%$), followed by foliose/membranous ($49.5 \pm 6.7\%$) and corticated/filamentous algae ($16.9 \pm 7.6\%$; Fig. 3). Accounting for these exceptions, the diet of *S. doliatus* and *S. virgatus* also differed from all other rabbitfishes (Table S1; Table S2).

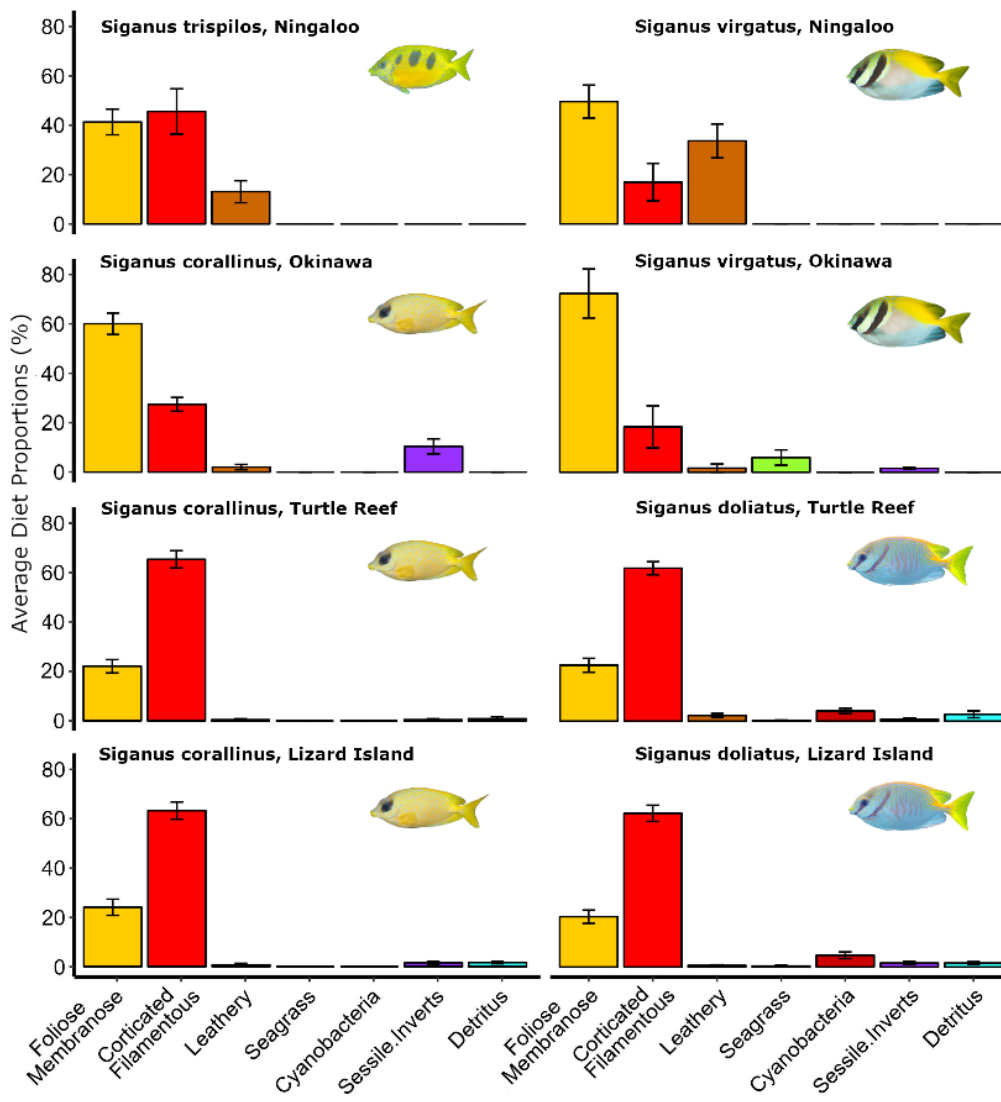


Figure 3. Barplots showing averages and standard errors (SE) of different diet items in the gut contents of different populations of the rabbitfishes *Siganus trispilos*, *S. corallinus*, *S. doliatus* and *S. virgatus* from different regions of the world: Great Barrier Reef (Turtle Reef and Lizard Island), Japan (Okinawa) and Western Australia (Ningaloo).

Intraspecific differences within and among regions

The diets of conspecific populations within the same region were not significantly different (Table S1; Table S2). Populations of *S. corallinus* from Lizard Island and Turtle Group within the GBR didn't differ significantly in their diets. Likewise, populations of *S. doliatus* within these two locations in the GBR didn't have statistically significant differences in their diets. These populations had diets dominated by corticated and filamentous algae seconded by foliose and membranous macroalgae; however, the trophic breadth of populations from Turtle Group were

smaller when compared at individual level (Fig. 4). In contrast, intraspecific differences in the diet of rabbitfishes separated at continental scales were highly significant (Table S1, Table 3). Both populations of *S. corallinus* from the GBR had significantly different diets than those from Okinawa, which tended to feed more on foliose and membranous macroalgae and sessile invertebrates, than individuals of the GBR (Fig. 3, Fig. 4). Likewise, populations of *S. virgatus* from Ningaloo and Okinawa also had marked differences in their diet, driven by higher consumption of leathery macroalgae by Ningaloo individuals and higher diet proportions of foliose and membranous macroalgae and seagrass by individuals from Okinawa (Fig. 3, Fig. 4).

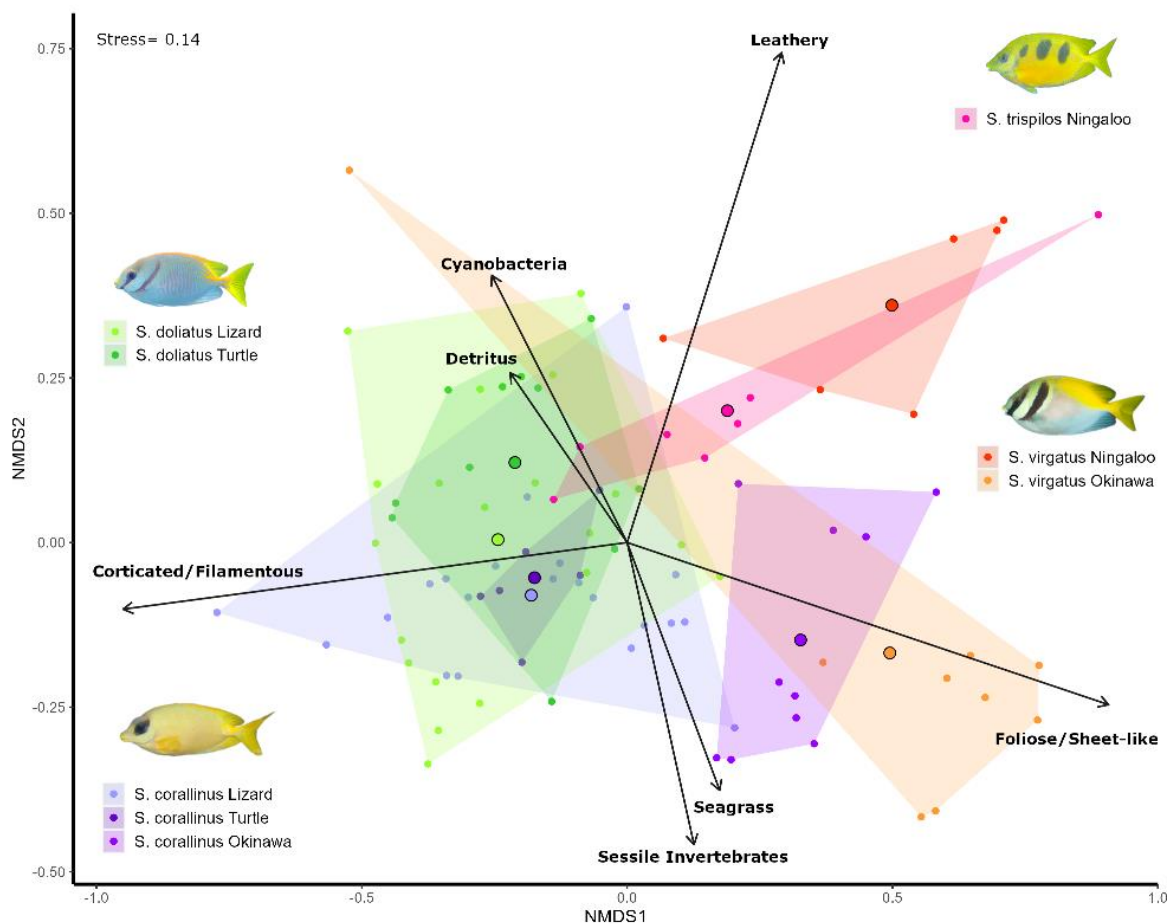


Figure 4. Non-metric multidimensional scaling ordination showing differences in diet composition between the rabbitfishes *Siganus trispilos*, *S. corallinus*, *S. doliaus* and *S. virgatus* from different regions of the world: Great Barrier Reef (Turtle Reef and Lizard Island), Japan (Okinawa) and Western Australia (Ningaloo). Bigger dots represent centroids of each population.

Discussion

Our study hypothesized that phylogenetically and morphologically similar species would have similar diets; however intra-and inter-specific differences will increase between populations of different biogeographic regions. We found that our results partially agreed with our hypotheses, as we found a strong effect in diet by geographic distribution, explaining 46% of the diet variation, effectively reducing the expected effect of phylogenetic and morphologic relatedness. While intra-specific differences were only significant when populations inhabited different regions as expected; inter-specific differences were not as predicted, with different morphotypes (non-sister species) having similar diets when populations inhabited the same regions.

The significant influence of the geographic distribution in rabbitfish diet can be attributed to their high trophic plasticity in response to shared local environmental conditions. Feeding behaviour and prey items can change depending on the environment in different locations, even for species considered to have specialized diets (Feary *et al.* 2018). Plasticity in herbivory among surgeonfish and parrotfish has been documented for a few species among sites within the same region (Locham *et al.* 2015; Duran *et al.* 2019). Trophic plasticity in herbivores can be influenced by multiple factors among biological interactions and habitat characteristics. Some studies, have shown that populations living in habitats with different structural complexity can differ in rates of consumption of different macroalgal resources (Vergés *et al.* 2011). This has been observed for the rabbitfishes *S. doliatus* and *S. canaliculatus* in the GBR, who's populations differed in their macroalgae consumption among the reef slope and reef crest, the latter having higher topographic complexity (Loffler, Bellwood & Hoey 2015). Habitat complexity also can interact with predator-prey dynamics and have important impacts in the foraging behaviour of herbivores, where the risk of predation suppressing herbivory more effectively in topographically complex areas; thus populations residing in ecosystems with differences in predator densities and composition can suffer significant changes in macroalgae consumption (Catano *et al.* 2016). For instance, the foraging of *Siganus virgatus* and *Siganus javus* in coral reefs of Singapore was heavily affected by the presence of a predator decoy, significantly reducing their consumption of *Sargassum* (Bauman *et al.* 2019).

Moreover, resource availability is one of the most important factors determining foraging plasticity in fish populations. The diet of fish species can vary depending on the abundance of dietary resources which are more or less preferred by the consumers; thus, populations residing in habitats with most palatable macrophyte species will likely differ from those residing in regions having less preferred resources (Barrientos *et al.* 2021). Such examples have been observed in some surgeonfish species, who generally preferred to feed on filamentous turf algae but consumption rates on fleshy and calcareous macroalgae increased at locations with higher abundance of them (Francini-Filho *et al.* 2010; Carlson *et al.* 2017) (Francini-Filho *et al.* 2010). The influence of resource availability on the diet of rabbitfish has only been documented in range-extending species, such as *Siganus luridus* and *Siganus rivulatus* in the Mediterranean Sea and *Siganus fuscescens* in western Australia, which consume a great variety of macrophytes in temperate reefs that are not found in their original habitats in coral reefs (Bariche 2006; Azzurro *et al.* 2007; Zarco-Perello *et al.* 2019). Our results document this phenomenon within tropical regions but separated at biogeographical scales. In the reef system of Ningaloo, *Sargassum* ssp. mats can be very abundant at the backreef and lagoon habitats, functioning as nursery habitats for some fish species (Evans *et al.* 2014). This may explain why *S. virgatus* from this region had higher gut contents of leathery macroalgae than the rest of the rabbitfish populations. This might be also the case for populations of *S. virgatus* inhabiting degraded coral reefs in Singapore and Thailand, where they have been observed feeding importantly on foliose and leathery macroalgae that are abundant in the ecosystems (Plass-Johnson *et al.* 2015; Bauman *et al.* 2017; Seah *et al.* 2021; Müller *et al.* 2021). Likewise, since *S. trispilos* is endemic of Western Australia, it might have adapted more to browse on this abundant resource than its sister species *S. coralinus* residing in mid- and outer-shelf reef habitats in the Great Barrier Reef, where *Sargassum* is less abundant (Wismer, Hoey & Bellwood 2009) and coral reefs in Okinawa which are dominated by branching *Acropora* spp. (Nanami 2018).

Consumption of new resources could be limited by morphological and physiological traits of each species. The specific trophic function of different herbivorous fishes has been linked strongly to their morphological traits (Bellwood *et al.* 2014a). We were expecting that only species with flat-snouted and more robust bodies would be consistently consuming important amounts of fleshy and leathery macroalgae (Fox & Bellwood 2008; Müller *et al.* 2021). On the other hand, species with more elongated snouts and slender bodies have been associated with feeding on filamentous algae in open and cryptic reef spaces (Fox & Bellwood 2013; Brandl & Bellwood 2014). The species of rabbitfish in our study belonged to both groups, each consisting of two sister species being more similar to each other in terms of snout length and

body shape (Nanami 2018). Yet, species of both morphological models ingested a mix of the same macroalgae types, similar to previous findings on surgeonfishes, where no relationship was found between morphology and diet specialization (Brandl, Robbins & Bellwood 2015). The fact that *S. trispilos* consumes significant amounts of leathery macroalgae shows that longer snouts are not an impediment to consume tough macrophytes, nor restricts the trophic behaviour of these rabbitfishes to grazing. Rather, it provides them with the ability to extend their trophic niche inside and outside cryptic reef spaces (Fox & Bellwood 2013; Brandl & Bellwood 2014). Dentition traits are important for the abilities of animals to consume certain types of food (Bellwood *et al.* 2014b), and although no study has made a dentition analysis in micro-detail among rabbitfish species, they seem to have similar traits, comprised of narrow incisor-like tricuspid teeth (Woodland 1990). Our results indicate that they may be suitable to crop a wide variety of resources.

The trophic plasticity of rabbitfishes has important repercussions for its functional roles in marine ecosystems. Herbivory can play an essential function in reinforcing the stability of coral dominated states in tropical reefs, reducing the abundance of macroalgae and favouring the development of coral colonies. However, whether herbivory can help the ecosystem to maintain and recover high abundances of scleractinean corals after suffering disturbances, is greatly determined by the specific trophic behaviours of different herbivorous fish species present in the community (Hoey & Bellwood 2009). The presence of populations of browsers and grazers that clear benthic space to facilitate coral recruitment is important to have high resilience in coral dominated states (Donovan *et al.* 2023). Currently, is common practice to assign trophic guild membership for herbivorous species based on studies conducted in a few different locations (Edwards *et al.* 2014). Our results stress the importance of local herbivory assessments and indicates that the behavioural plasticity of the species must be considered when assessing the intensity of different herbivory functions at global or biogeographic scales. In similitude to the concepts of fundamental vs realized niche, our study highlights the distinction between (a) *fundamental herbivory*: the ability of a species to perform herbivory functions and (b) *realized herbivory*: the function executed by one population under specific physical and biological conditions. For instance, the batfish *Platax pinnatus* which normally feeds on benthic and planktonic invertebrates, was found to consume high amounts of *Sargassum* sp. when it was extraordinarily presented with this resource in a herbivory-exclusion experiment (Bellwood *et al.* 2006). Thus, the trophic functions among the fish community must be carefully assessed at local scale to ensure that the species responsible for critical ecosystem processes are accurately identified and included in management strategies (Chung *et al.* 2019).

Our study explored the differences in diet between populations of rabbitfish across geographic distribution, phylogeny and morphology. We found significant trophic plasticity among the four rabbitfish species compared, where diets seemed to be strongly related to geographic distribution. The results suggest that the trophic role among rabbitfishes, and potentially species of other herbivorous fish families, is difficult to extrapolate across locations and across species, even when they are closely related. These results are particularly important as the assessment of coral reef resilience includes examining the abundance of key fish herbivorous guilds that can prevent and revert phase-shifts to coral dominated states based on what macroalgae they consume. As climate change unfolds, shifts in the distribution, trophic behaviour and function of species are expected, making the study of trophic plasticity more crucial.

Acknowledgements

We would like to thank Helen Armstrong and Thomas Cribb for their help on fish collection during the fieldtrips to Coral Bay, Ningaloo. Special thanks to Marie Maca Windstein for her awesome support during the diet analyses of specimens of *Siganus trispilos* and *Siganus virgatus* in the old spooky parasitology lab at Murdoch University.

Data availability

All data used in this paper will be publicly available via Dryad after acceptance for publication.

References

Azzurro, E., Fanelli, E., Mostarda, E., Catra, M. & Andaloro, F. (2007) Resource partitioning among early colonizing *Siganus luridus* and native herbivorous fish in the Mediterranean: an integrated study based on gut-content analysis and stable isotope signatures. *Journal of the Marine Biological Association of the UK*, **87**, 991.

Bariche, M. (2006) Diet of the Lessepsian fishes, *Siganus rivulatus* and *S. luridus* (Siganidae) in the eastern Mediterranean: A bibliographic analysis. *Cybium*, **30**, 41–49.

Barrientos, S., Zarco-Perello, S., Piñeiro-Corbeira, C., Barreiro, R. & Wernberg, T. (2021) Feeding preferences of range-shifting and native herbivorous fishes in temperate ecosystems. *Marine Environmental Research*, **172**, 105508.

- Bauman, A.G., Hoey, A.S., Dunshea, G., Feary, D.A., Low, J. & Todd, P.A. (2017) Macroalgal browsing on a heavily degraded, urbanized equatorial reef system. *Scientific Reports*, **7**, 8352.
- Bauman, A.G., Seah, J.C.L., Januchowski-Hartley, F.A., Hoey, A.S., Fong, J. & Todd, P.A. (2019) Fear effects associated with predator presence and habitat structure interact to alter herbivory on coral reefs. *Biology Letters*, **15**, 20190409.
- Bellwood, D.R., Goatley, C.H.R., Brandl, S.J. & Bellwood, O. (2014a) Fifty million years of herbivory on coral reefs: fossils, fish and functional innovations. *Proceedings. Biological Sciences / the Royal Society*, **281**, 20133046.
- Bellwood, D.R., Hoey, A.S., Bellwood, O. & Goatley, C.H.R. (2014b) Evolution of long-toothed fishes and the changing nature of fish-benthos interactions on coral reefs. *Nature Communications*, **5**, 3144.
- Bellwood, D.R., Hughes, T.P. & Hoey, A.S. (2006) Sleeping functional group drives coral-reef recovery. *Current Biology*, **16**, 2434–2439.
- Bennett, S. & Bellwood, D.R. (2011) Latitudinal variation in macroalgal consumption by fishes on the Great Barrier Reef. *Marine Ecology Progress Series*, **426**, 241–252.
- Bonaldo, R.M., Hoey, A.S. & Bellwood, D.R. (2014) The ecosystem roles of parrotfishes on tropical reefs. *Oceanography and marine biology: an annual review* (eds R.N. Hughes), D.J. Hughes, & I.P. Smith), pp. 81–132. CRC Press.
- Brandl, S.J. & Bellwood, D.R. (2014) Individual-based analyses reveal limited functional overlap in a coral reef fish community. *The Journal of Animal Ecology*, **83**, 661–670.
- Brandl, S.J., Robbins, W.D. & Bellwood, D.R. (2015) Exploring the nature of ecological specialization in a coral reef fish community: morphology, diet and foraging microhabitat use. *Proceedings. Biological Sciences / the Royal Society*, **282**.
- Carlson, P.M., Davis, K., Warner, R.R. & Caselle, J.E. (2017) Fine-scale spatial patterns of parrotfish herbivory are shaped by resource availability. *Marine Ecology Progress Series*, **577**, 165–176.
- Catano, L.B., Rojas, M.C., Malossi, R.J., Peters, J.R., Heithaus, M.R., Fourqurean, J.W. & Burkepile, D.E. (2016) Reefscapes of fear: predation risk and reef heterogeneity interact to shape herbivore foraging behaviour. *The Journal of Animal Ecology*, **85**, 146–156.
- Cheal, A.J., Emslie, M., Aaron, M.M., Miller, I. & Sweatman, H. (2013) Spatial variation in the functional characteristics of herbivorous fish communities and the resilience of coral reefs. *Ecological Applications*, **23**, 174–188.
- Chung, A.E., Wedding, L.M., Green, A.L., Friedlander, A.M., Goldberg, G., Meadows, A. & Hixon, M.A. (2019) Building coral reef resilience through spatial herbivore management. *Frontiers in Marine Science*, **6**.
- Clements, K.D., German, D.P., Piché, J., Tribollet, A. & Choat, J.H. (2017) Integrating ecological roles and trophic diversification on coral reefs: multiple lines of evidence identify parrotfishes as microphages. *Biological Journal of the Linnean Society*, **120**, 729–751.
- Donovan, M.K., Counsell, C.W.W., Donahue, M.J., Lecky, J., Gajdzik, L., Marcoux, S.D., Sparks, R. & Teague, C. (2023) Evidence for managing herbivores for reef resilience. *Proceedings. Biological Sciences / the Royal Society*, **290**, 20232101.

- Duran, A., Adam, T.C., Palma, L., Moreno, S., Collado-Vides, L. & Burkepile, D.E. (2019) Feeding behavior in Caribbean surgeonfishes varies across fish size, algal abundance, and habitat characteristics. *Marine Ecology*, **40**.
- Edwards, C.B., Friedlander, A.M., Green, A.G., Hardt, M.J., Sala, E., Sweatman, H.P., Williams, I.D., Zgliczynski, B., Sandin, S.A. & Smith, J.E. (2014) Global assessment of the status of coral reef herbivorous fishes: evidence for fishing effects. *Proceedings. Biological Sciences / the Royal Society*, **281**, 20131835.
- Evans, R.D., Wilson, S.K., Field, S.N. & Moore, J.A.Y. (2014) Importance of macroalgal fields as coral reef fish nursery habitat in north-west Australia. *Marine Biology*, **161**, 599–607.
- Feary, D.A., Bauman, A.G., Guest, J. & Hoey, A.S. (2018) Trophic plasticity in an obligate corallivorous butterflyfish. *Marine Ecology Progress Series*, **605**, 165–171.
- Flower, A., McKenna, J.W. & Upreti, G. (2016) Validity and reliability of graphclick and datathief III for data extraction. *Behavior modification*, **40**, 396–413.
- Fox, R.J. & Bellwood, D.R. (2008) Remote video bioassays reveal the potential feeding impact of the rabbitfish *Siganus canaliculatus* (f: Siganidae) on an inner-shelf reef of the Great Barrier Reef. *Coral Reefs*, **27**, 605–615.
- Fox, R.J. & Bellwood, D.R. (2013) Niche partitioning of feeding microhabitats produces a unique function for herbivorous rabbitfishes (Perciformes, Siganidae) on coral reefs. *Coral reefs (Online)*, **32**, 13–23.
- Francini-Filho, R.B., Ferreira, C.M., Coni, E.O.C., De Moura, R.L. & Kaufman, L. (2010) Foraging activity of roving herbivorous reef fish (Acanthuridae and Scaridae) in eastern Brazil: influence of resource availability and interference competition. *Journal of the Marine Biological Association of the UK*, **90**, 481–492.
- Hamilton, S.L., Caselle, J.E., Lantz, C.A., Egloff, T.L., Kondo, E., Newsome, S.D., Loke-Smith, K., Pondella, D.J., Young, K.A. & Lowe, C.G. (2011) Extensive geographic and ontogenetic variation characterizes the trophic ecology of a temperate reef fish on southern California (USA) rocky reefs. *Marine ecology progress series*, **429**, 227–244.
- Hoey, A.S. & Bellwood, D.R. (2009) Limited Functional Redundancy in a High Diversity System: Single Species Dominates Key Ecological Process on Coral Reefs. *Ecosystems (New York, N.Y.)*, **12**, 1316–1328.
- Hoey, A.S., Brandl, S.J. & Bellwood, D.R. (2013) Diet and cross-shelf distribution of rabbitfishes (f. Siganidae) on the northern Great Barrier Reef: implications for ecosystem function. *Coral Reefs*, **32**, 973–984.
- Johansson, C.L., van de Leemput, I.A., Depczynski, M., Hoey, A.S. & Bellwood, D.R. (2013) Key herbivores reveal limited functional redundancy on inshore coral reefs. *Coral Reefs*, **32**, 963–972.
- Kelly, E.L.A., Eynaud, Y., Clements, S.M., Gleason, M., Sparks, R.T., Williams, I.D. & Smith, J.E. (2016) Investigating functional redundancy versus complementarity in Hawaiian herbivorous coral reef fishes. *Oecologia*, **182**, 1151–1163.
- Korzen, L., Israel, A. & Abelson, A. (2011) Grazing Effects of Fish versus Sea Urchins on Turf Algae and Coral Recruits: Possible Implications for Coral Reef Resilience and Restoration. *Journal of marine biology*, **2011**, 1–8.

- Locham, A.G., Kaunda-Arara, B., Wakibia, J.G. & Muya, S. (2015) Diet and niche breadth variation in the marbled parrotfish, *Leptoscarus vaigiensis*, among coral reef sites in Kenya. *African journal of ecology*, **53**, 560–571.
- Loffler, Z., Bellwood, D.R. & Hoey, A.S. (2015) Among-habitat algal selectivity by browsing herbivores on an inshore coral reef. *Coral Reefs*, **34**, 597–605.
- Martinez, A.P. (2017) *_pairwiseAdonis: Pairwise Multilevel Comparison Using Adonis_*. Computer software, R.
- McCook, L., Jompa, J. & Diaz-Pulido, G. (2001) Competition between corals and algae on coral reefs: a review of evidence and mechanisms. *Coral reefs (Online)*, **19**, 400–417.
- Müller, M., Staab, C.F.K., Puk, L.D., Schoenig, E.M., Ferse, S.C.A. & Wild, C. (2021) The Rabbitfish *Siganus virgatus* as Key Macroalgae Browser in Coral Reefs of the Gulf of Thailand. *Diversity*, **13**, 123.
- Nanami, A. (2018) Spatial distributions, feeding ecologies, and behavioral interactions of four rabbitfish species (*Siganus unimaculatus*, *S. virgatus*, *S. corallinus*, and *S. puellus*). *PeerJ*, **6**, e6145.
- Oksanen, J., Blanchet, F.G., Friendly, M., Kindt, R., Legendre, P., McGlinn, D., Minchin, P.R., O'Hara, R.B., Simpson, G.L., Solymos, P., Stevens, M.H.H., Szoecs, E. & Wagner, H. (2022) *Vegan: Community Ecology Package*. Computer software, R.
- Plass-Johnson, J.G., Ferse, S.C.A., Jompa, J., Wild, C. & Teichberg, M. (2015) Fish herbivory as key ecological function in a heavily degraded coral reef system. *Limnology and Oceanography Letters*, **60**, 1382–1391.
- Puk, L.D., Cernohorsky, N., Marshall, A., Dwyer, J., Wolfe, K. & Mumby, P.J. (2020) Species-specific effects of herbivorous fishes on the establishment of the macroalga *Lobophora* on coral reefs. *Marine Ecology Progress Series*, **637**, 1–14.
- Puk, L.D., Ferse, S.C.A. & Wild, C. (2016) Patterns and trends in coral reef macroalgae browsing: a review of browsing herbivorous fishes of the Indo-Pacific. *Reviews in Fish Biology and Fisheries*, **26**, 53–70.
- Seah, J.C.L., Bauman, A.G. & Todd, P.A. (2021) Temporal variation in macroalgal removal: insights from an impacted equatorial coral reef system. *Marine Biology*, **168**, 2.
- Siqueira, A.C., Bellwood, D.R. & Cowman, P.F. (2019) The evolution of traits and functions in herbivorous coral reef fishes through space and time. *Proceedings. Biological Sciences / the Royal Society*, **286**, 20182672.
- Spadaro, A.J. & Butler, M.J. (2021) Herbivorous crabs reverse the seaweed dilemma on coral reefs. *Current Biology*, **31**, 853-859.e3.
- Steneck, R.S., Bellwood, D.R. & Hay, M.E. (2017) Herbivory in the marine realm. *Current Biology*, **27**, R484–R489.
- Tebbett, S.B., Siqueira, A.C. & Bellwood, D.R. (2022) The functional roles of surgeonfishes on coral reefs: past, present and future. *Reviews in Fish Biology and Fisheries*.
- Vergés, A., Vanderklift, M.A., Doropoulos, C. & Hyndes, G.A. (2011) Spatial patterns in herbivory on a coral reef are influenced by structural complexity but not by algal traits. *Plos One*, **6**, e17115.

- Wilson, M.W., Gaines, S.D., Stier, A.C. & Halpern, B.S. (2021) Variation in herbivore grazing behavior across Caribbean reef sites. *Marine Biology*, **168**, 53.
- Wismer, S., Hoey, A.S. & Bellwood, D.R. (2009) Cross-shelf benthic community structure on the Great Barrier Reef: relationships between macroalgal cover and herbivore biomass. *Marine Ecology Progress Series*, **376**, 45–54.
- Woodland, D.J. & Allen, G.R. (1977) *Siganus trispilos*, a New Species of Siganidae from the Eastern Indian Ocean. *Copeia*, **1977**, 617.
- Woodland, D.J. (1990) Revision of the fish family Siganidae with descriptions of two new species and comments on distribution and biology. *Indo-Pacific Fishes*, **19**.
- Zarco-Perello, S., Langlois, T.J., Holmes, T., Vanderklift, M.A. & Wernberg, T. (2019) Overwintering tropical herbivores accelerate detritus production on temperate reefs. *Proceedings. Biological Sciences / the Royal Society*, **286**, 20192046.

Supplementary Materials

Biogeographical diet variation within and between the rabbitfishes *Siganus corallinus*, *Siganus virgatus*, *Siganus doliatus* and the endemic *Siganus trispilos*.

Zarco-Perello, S. ^{1,2}, Martin, S. ¹, Hoey A. ³

¹ Harry Butler Institute, Murdoch University, Perth, Australia.

² College of Life Sciences and Agriculture, University of New Hampshire, Durham, USA.

³ ARC Centre of Excellence for Coral Reef Studies and College of Science and Engineering, James Cook University, Townsville, Queensland, Australia

Table S1. Results of the PERMANOVA, homogeneity of variances and pairwise comparisons in diet composition between different species and populations of the rabbitfishes *Siganus trispilos*, *Siganus corallinus* and *Siganus virgatus*.

Model:

adonis2(formula = Sig.diet.dbs ~ Location + Sister.taxa / Species, data = Sig.names, permutations = 9999, strata = Sister.taxa)

General Anova:

	Df	SumOfSqs	R2	F	Pr(>F)
Location	3	2.6984	0.46374	27.0833	0.0001 ***
Sister.taxa	1	0.1633	0.02807	4.9176	0.0001 ***
Sister.taxa:Species	2	0.2006	0.03447	3.0200	0.0138 *
Residual	83	2.7565	0.47372		
Total	89	5.8188	1.00000		

Homogeneity of multivariate dispersions by populations

	Df	Sum Sq	Mean Sq	F	N.Perm	Pr(>F)
Groups	7	0.0785	0.0112140	1.3263	9999	0.242
Residuals	82	0.6933	0.0084549			

Pairwise comparisons of populations:

\$`s. virgatus okinawa_vs_s. corallinus okinawa`

	Df	SumOfSqs	R2	F	Pr(>F)
Population.ID	1	0.08738	0.10459	1.9858	0.132
Residual	17	0.74808	0.89541		
Total	18	0.83547	1.00000		

\$`s. virgatus okinawa_vs_s. trispilos Ningaloo`

	Df	SumOfSqs	R2	F	Pr(>F)	
Population.ID	1	0.35606	0.31696	6.4967	0.008	**
Residual	14	0.76729	0.68304			
Total	15	1.12335	1.00000			

\$`S. virgatus Okinawa_vs_S. virgatus Ningaloo`

	Df	SumOfSqs	R2	F	Pr(>F)	
Population.ID	1	0.35164	0.33924	6.6744	0.007	**
Residual	13	0.68491	0.66076			
Total	14	1.03655	1.00000			

\$`S. virgatus Okinawa_vs_S. corallinus Lizard`

	Df	SumOfSqs	R2	F	Pr(>F)	
Population.ID	1	0.82694	0.42943	21.073	0.001	***
Residual	28	1.09874	0.57057			
Total	29	1.92568	1.00000			

\$`S. virgatus Okinawa_vs_S. corallinus Turtle`

	Df	SumOfSqs	R2	F	Pr(>F)	
Population.ID	1	0.46829	0.42731	9.6999	0.008	**
Residual	13	0.62761	0.57269			
Total	14	1.09589	1.00000			

\$`S. virgatus Okinawa_vs_S. doliatus Lizard`

	Df	SumOfSqs	R2	F	Pr(>F)	
Population.ID	1	0.93316	0.43822	21.062	0.001	***
Residual	27	1.19626	0.56178			
Total	28	2.12942	1.00000			

\$`S. virgatus Okinawa_vs_S. doliatus Turtle`

	Df	SumOfSqs	R2	F	Pr(>F)	
Population.ID	1	0.79244	0.49278	17.488	0.001	***
Residual	18	0.81565	0.50722			
Total	19	1.60809	1.00000			

\$`S. corallinus Okinawa_vs_S. trispilos Ningaloo`

	Df	SumOfSqs	R2	F	Pr(>F)	
Population.ID	1	0.22613	0.39694	9.8733	0.001	***
Residual	15	0.34355	0.60306			
Total	16	0.56968	1.00000			

\$`S. corallinus Okinawa_vs_S. virgatus Ningaloo`

	Df	SumOfSqs	R2	F	Pr(>F)	
Population.ID	1	0.35591	0.57677	19.079	0.001	***
Residual	14	0.26117	0.42323			
Total	15	0.61708	1.00000			

\$`S. corallinus Okinawa_vs_S. corallinus Lizard`

	Df	SumOfSqs	R2	F	Pr(>F)	
Population.ID	1	0.58283	0.46336	25.04	0.001	***
Residual	29	0.67500	0.53664			
Total	30	1.25783	1.00000			

\$`s. corallinus Okinawa_vs_S. corallinus Turtle`

	Df	SumOfSqs	R2	F	Pr(>F)
Population.ID	1	0.33051	0.61849	22.697	0.001 ***
Residual	14	0.20387	0.38151		
Total	15	0.53438	1.00000		

\$`s. corallinus Lizard_vs_S. corallinus Turtle`

	Df	SumOfSqs	R2	F	Pr(>F)
Population.ID	1	0.01123	0.01986	0.5065	0.651
Residual	25	0.55453	0.98014		
Total	26	0.56576	1.00000		

\$`s. corallinus Okinawa_vs_S. doliatus Lizard`

	Df	SumOfSqs	R2	F	Pr(>F)
Population.ID	1	0.67546	0.46648	24.482	0.001 ***
Residual	28	0.77252	0.53352		
Total	29	1.44799	1.00000		

\$`s. corallinus Okinawa_vs_S. doliatus Turtle`

	Df	SumOfSqs	R2	F	Pr(>F)
Population.ID	1	0.55738	0.58715	27.021	0.001 ***
Residual	19	0.39192	0.41285		
Total	20	0.94929	1.00000		

\$`s. trispilos Ningaloo_vs_S. virgatus Ningaloo`

	Df	SumOfSqs	R2	F	Pr(>F)
Population.ID	1	0.08403	0.2306	3.2968	0.097 .
Residual	11	0.28037	0.7694		
Total	12	0.36441	1.0000		

\$`s. trispilos Ningaloo_vs_S. corallinus Lizard`

	Df	SumOfSqs	R2	F	Pr(>F)
Population.ID	1	0.39737	0.36403	14.883	0.001 ***
Residual	26	0.69421	0.63597		
Total	27	1.09158	1.00000		

\$`s. trispilos Ningaloo_vs_S. corallinus Turtle`

	Df	SumOfSqs	R2	F	Pr(>F)
Population.ID	1	0.21519	0.491	10.611	0.003 **
Residual	11	0.22308	0.509		
Total	12	0.43826	1.000		

\$`s. trispilos Ningaloo_vs_S. doliatus Lizard`

	Df	SumOfSqs	R2	F	Pr(>F)
Population.ID	1	0.43762	0.35598	13.819	0.001 ***
Residual	25	0.79173	0.64402		
Total	26	1.22935	1.00000		

\$`s. trispilos Ningaloo_vs_S. doliatus Turtle`

	Df	SumOfSqs	R2	F	Pr(>F)
--	----	----------	----	---	--------

Population.ID	1	0.26530	0.39221	10.325	0.001	***
Residual	16	0.41112	0.60779			
Total	17	0.67643	1.00000			

\$`S. virgatus Ningaloo_vs_S. corallinus Lizard`

	Df	SumOfSqs	R2	F	Pr(>F)	
Population.ID	1	0.90477	0.59658	36.97	0.001	***
Residual	25	0.61183	0.40342			
Total	26	1.51660	1.00000			

\$`S. virgatus Ningaloo_vs_S. corallinus Turtle`

	Df	SumOfSqs	R2	F	Pr(>F)	
Population.ID	1	0.54308	0.79424	38.6	0.002	**
Residual	10	0.14069	0.20576			
Total	11	0.68377	1.00000			

\$`S. virgatus Ningaloo_vs_S. doliatus Lizard`

	Df	SumOfSqs	R2	F	Pr(>F)	
Population.ID	1	0.95829	0.57464	32.423	0.001	***
Residual	24	0.70935	0.42536			
Total	25	1.66763	1.00000			

\$`S. virgatus Ningaloo_vs_S. doliatus Turtle`

	Df	SumOfSqs	R2	F	Pr(>F)	
Population.ID	1	0.65801	0.66685	30.024	0.001	***
Residual	15	0.32874	0.33315			
Total	16	0.98675	1.00000			

\$`S. corallinus Lizard_vs_S. doliatus Lizard`

	Df	SumOfSqs	R2	F	Pr(>F)	
Population.ID	1	0.10277	0.08383	3.5684	0.021	*
Residual	39	1.12318	0.91617			
Total	40	1.22595	1.00000			

\$`S. corallinus Lizard_vs_S. doliatus Turtle`

	Df	SumOfSqs	R2	F	Pr(>F)	
Population.ID	1	0.14865	0.16679	6.0055	0.003	**
Residual	30	0.74258	0.83321			
Total	31	0.89123	1.00000			

\$`S. corallinus Turtle_vs_S. doliatus Lizard`

	Df	SumOfSqs	R2	F	Pr(>F)	
Population.ID	1	0.04698	0.06721	1.7293	0.193	
Residual	24	0.65205	0.93279			
Total	25	0.69903	1.00000			

\$`S. corallinus Turtle_vs_S. doliatus Turtle`

	Df	SumOfSqs	R2	F	Pr(>F)	
Population.ID	1	0.05588	0.17071	3.0878	0.027	*
Residual	15	0.27144	0.82929			
Total	16	0.32732	1.00000			

\$`s. doliatus Lizard_vs_S. doliatus Turtle`

	Df	SumOfSqs	R2	F	Pr(>F)
Population.ID	1	0.02394	0.0277	0.8263	0.53
Residual	29	0.84009	0.9723		
Total	30	0.86403	1.0000		

Table S2. Synthesis of statistical significance (p-values from PERMANOVA) in pairwise comparisons of differences in diet between species and populations of the rabbitfishes from the Great Barrier Reef (Turtle Reef and Lizard Island), Japan (Okinawa) and Western Australia (Ningaloo).

Population	<i>Siganus corallinus</i> Lizard	<i>Siganus corallinus</i> Turtle	<i>Siganus corallinus</i> Okinawa	<i>Siganus doliatus</i> Lizard	<i>Siganus doliatus</i> Turtle	<i>Siganus virgatus</i> Ningaloo	<i>Siganus virgatus</i> Okinawa
<i>S. corallinus</i> Turtle	0.651						
<i>S. corallinus</i> Okinawa	*** 0.001	*** 0.001					
<i>S. doliatus</i> Lizard	* 0.021	0.193	*** 0.001				
<i>S. doliatus</i> Turtle	** 0.003	* 0.027	*** 0.001	0.53			
<i>S. virgatus</i> Ningaloo	*** 0.001	** 0.002	*** 0.001	*** 0.001	*** 0.001		
<i>S. virgatus</i> Okinawa	*** 0.001	** 0.008	0.132	*** 0.001	*** 0.001	** 0.007	
<i>S. trispilos</i> Ningaloo	*** 0.001	** 0.003	*** 0.001	*** 0.001	*** 0.001	0.097	** 0.008