Factors constraining natural recovery of *Diadema antillarum* following a mass die-off: a case study near Saba, Caribbean Netherlands (#114861)

First submission

Guidance from your Editor

Please submit by **9 Apr 2025** for the benefit of the authors (and your token reward) .

Structure and Criteria
Please read the 'Structure and Criteria' page for guidance.

Custom checks
Make sure you include the custom checks shown below, in your review.

Raw data check
Review the raw data.

Image check
Check that figures and images have not been inappropriately manipulated.

If this article is published your review will be made public. You can choose whether to sign your review. If uploading a PDF please remove any identifiable information (if you want to remain anonymous).

Files

Download and review all files from the materials page.

4 Figure file(s)
1 Table file(s)
1 Raw data file(s)
1 Other file(s)

Custom checks

Field study

Have you checked the authors field study permits?

Are the field study permits appropriate?

Structure and Criteria

Structure your review

The review form is divided into 5 sections. Please consider these when composing your review:

1. **BASIC REPORTING**
2. **EXPERIMENTAL DESIGN**
3. **VALIDITY OF THE FINDINGS**
4. General comments
5. Confidential notes to the editor

🗋 You can also annotate this PDF and upload it as part of your review

When ready submit online.

Editorial Criteria

Use these criteria points to structure your review. The full detailed editorial criteria is on your guidance page.

BASIC REPORTING

- ✓ Clear, unambiguous, professional English language used throughout.
- ✓ Intro & background to show context. Literature well referenced & relevant.
- ✓ Structure conforms to PeerJ standards, discipline norm, or improved for clarity.
- ✓ Figures are relevant, high quality, well labelled & described.
- ✓ Raw data supplied (see PeerJ policy).

EXPERIMENTAL DESIGN

- ✓ Original primary research within Scope of the journal.
- ✓ Research question well defined, relevant & meaningful. It is stated how the research fills an identified knowledge gap.
- ✓ Rigorous investigation performed to a high technical & ethical standard.
- ✓ Methods described with sufficient detail & information to replicate.

VALIDITY OF THE FINDINGS

- ℹ **Impact and novelty is not assessed.** Meaningful replication encouraged where rationale & benefit to literature is clearly stated.
- ✓ All underlying data have been provided; they are robust, statistically sound, & controlled.
- ✓ Conclusions are well stated, linked to original research question & limited to supporting results.

Standout reviewing tips

The best reviewers use these techniques

Tip	*Example*
Support criticisms with evidence from the text or from other sources	*Smith et al (J of Methodology, 2005, V3, pp 123) have shown that the analysis you use in Lines 241-250 is not the most appropriate for this situation. Please explain why you used this method.*
Give specific suggestions on how to improve the manuscript	*Your introduction needs more detail. I suggest that you improve the description at lines 57- 86 to provide more justification for your study (specifically, you should expand upon the knowledge gap being filled).*
Comment on language and grammar issues	*The English language should be improved to ensure that an international audience can clearly understand your text. Some examples where the language could be improved include lines 23, 77, 121, 128 – the current phrasing makes comprehension difficult. I suggest you have a colleague who is proficient in English and familiar with the subject matter review your manuscript, or contact a professional editing service.*
Organize by importance of the issues, and number your points	*1. Your most important issue* *2. The next most important item* *3. …* *4. The least important points*
Please provide constructive criticism, and avoid personal opinions	*I thank you for providing the raw data, however your supplemental files need more descriptive metadata identifiers to be useful to future readers. Although your results are compelling, the data analysis should be improved in the following ways: AA, BB, CC*
Comment on strengths (as well as weaknesses) of the manuscript	*I commend the authors for their extensive data set, compiled over many years of detailed fieldwork. In addition, the manuscript is clearly written in professional, unambiguous language. If there is a weakness, it is in the statistical analysis (as I have noted above) which should be improved upon before Acceptance.*

Factors constraining natural recovery of *Diadema antillarum* following a mass die-off: a case study near Saba, Caribbean Netherlands

Alwin Hylkema Corresp., 1, 2 , **Oliver J Klokman** 1, 2

1 Van Hall Larenstein University of Applied Sciences, Leeuwarden, Netherlands
2 Marine Animal Ecology group, Wageningen University and Research, Wageningen, Netherlands

Corresponding Author: Alwin Hylkema
Email address: alwin.hylkema@hvhl.nl

After a mass mortality in 1983-1984, recovery of the keystone herbivore *Diadema antillarum*, from here on *Diadema*, has been slow to non-existent. Due to the loss of grazing pressure, Caribbean coral reefs became covered with algae, which inhibit coral recruitment and reduce the resilience of Caribbean coral reefs. To understand recovery dynamics and to achieve effective restoration, an increased insight into factors constraining population establishment is necessary. *Diadema* settlement rates, predation pressure and shelter availability might all affect recovery. The presence of adult *Diadema* potentially interact with these factors, as adults might provide settlement cues, enhance shelter, and remove macroalgae which harbor micro-predators. The 2022 *Diadema* die-off, wiping out adult populations, provided the opportunity to study factors potentially constraining recovery without the interactive effect of adults. In this case study, we compared four locations near Saba, Caribbean Netherlands in settlement rates, predator density, shelter availability and *Diadema* recovery after the 2022 die-off. One location, an artificial reef made of rocks, previously had high *Diadema* densities, while the three other natural reef locations all had sporadic recruitment but no population establishment. After the 2022 die-off, all locations had close to zero *Diadema*. One year later, the artificial reef location had a substantial higher *Diadema* density compared to the other locations. Although the artificial reef had a high density of fishes predating on *Diadema*, densities of Spanish hogfish and smaller wrasses was lower than at natural reef locations. Other studies indicated that especially these fishes, in combination with the queen triggerfish, might be essential in determining *Diadema* population recovery. Invertebrate predator density was similar at all locations, except for the red night shrimp and the spotted spiny lobster, which were more abundant at the artificial reef location. Spotted spiny lobster are known predators of smaller *Diadema*, but the large percentage of crabs in their diet might result in a net positive effect on *Diadema* populations. Finally, the shelter availability, depth, and shape appeared most favorable at the artificial reef location. We conclude that *Diadema* population establishment is likely determined by a combination of predation pressure and shelter availability. Settlement of *Diadema* larvae and presence of adult *Diadema* appear to be less important factors. Repeating this study on a regional scale, including multiple islands and locations with and without *Diadema,* might help to further detangle the factors affecting natural recovery, which is essential to identify reefs with a high chance of successful *Diadema* restoration.

Factors constraining natural recovery of *Diadema antillarum* following a mass die-off: a case study near Saba, Caribbean Netherlands

Alwin Hylkema[1,2], Oliver J. Klokman[1,2]

[1] Van Hall Larenstein University of Applied Sciences, P.O. Box 1528, 8901 BV Leeuwarden, the Netherlands
[2] Marine Animal Ecology group, Wageningen University, Wageningen University & Research, P.O. Box 338, 6700 AH Wageningen, The Netherlands

Corresponding Author:
Alwin Hylkema[1,2]
Email address: Alwin.hylkema@hvhl.nl

Abstract

After a mass mortality in 1983-1984, recovery of the keystone herbivore *Diadema antillarum*, from here on *Diadema*, has been slow to non-existent. Due to the loss of grazing pressure, Caribbean coral reefs became covered with algae, which inhibit coral recruitment and reduce the resilience of Caribbean coral reefs. To understand recovery dynamics and to achieve effective restoration, an increased insight into factors constraining population establishment is necessary. *Diadema* settlement rates, predation pressure and shelter availability might all affect recovery. The presence of adult *Diadema* potentially interact with these factors, as adults might provide settlement cues, enhance shelter, and remove macroalgae which harbor micro-predators. The 2022 *Diadema* die-off, wiping out adult populations, provided the opportunity to study factors potentially constraining recovery without the interactive effect of adults. In this case study, we compared four locations near Saba, Caribbean Netherlands in settlement rates, predator density, shelter availability and *Diadema* recovery after the 2022 die-off. One location, an artificial reef made of rocks, previously had high *Diadema* densities, while the three other natural reef locations all had sporadic recruitment but no population establishment. After the 2022 die-off, all locations had close to zero *Diadema*. One year later, the artificial reef location had a substantial higher *Diadema* density compared to the other locations. Although the artificial reef had a high density of fishes predating on *Diadema*, densities of Spanish hogfish and smaller wrasses was lower than at natural reef locations. Other studies indicated that especially these fishes, in combination with the queen triggerfish, might be essential in determining *Diadema* population recovery. Invertebrate predator density was similar at all locations, except for the red night shrimp and the spotted spiny lobster, which were more abundant at the artificial reef location. Spotted spiny lobster are known predators of smaller *Diadema*, but the large percentage of crabs in their diet might result in a net positive effect on *Diadema* populations. Finally, the shelter availability, depth, and shape appeared most favorable at the artificial reef location. We conclude that *Diadema* population establishment is likely determined by a combination of predation pressure and shelter availability. Settlement of *Diadema* larvae and presence of adult *Diadema* appear to be less important factors. Repeating this study on a regional scale, including multiple islands and locations with and without *Diadema,* might help to further detangle the factors affecting natural recovery, which is essential to identify reefs with a high chance of successful *Diadema* restoration.

Key words: Sea urchin, Echinoid, Settlement, Predation, Shelter availability

Introduction

The long-spined sea urchin *Diadema antillarum*, commonly known and from here on referred to as *Diadema*, was the most abundant herbivore on Caribbean coral reefs (Lessios et al. 2001). A mass die-off in 1983-1984 reduced *Diadema* population densities by 98% (Lessios 2016) and demonstrated that no other species was able to fill their grazing niche. Herbivorous fishes, the only other abundant group of herbivores, were already severely overfished in most of the Caribbean (Pandolfi et al. 2003). Although some herbivorous fishes increased in abundance after the *Diadema* die-off (Carpenter 1990a, Robertson 1991), they were not able to compensate for the loss of grazing pressure (Levitan 1988, Carpenter 1990a). Within days after the *Diadema* died and the grazing pressure was suddenly reduced, turf algae started to increase in biomass (Carpenter 1988). In the years following the *Diadema* mass mortality, turf and macroalgae became the dominant benthic groups (de Ruyter van Steveninck & Bak 1986, Hughes et al. 1987, Carpenter 1990b). Other threats, mostly climate change induced heatwaves and diseases, resulted in wide-scale coral mortality in the subsequent decades (Riegl et al. 2009), freeing up space for the expanding algae.

Turf and macroalgae compete with corals for light, space, and nutrients (McCook et al. 2001). The proliferating algae inhibited coral recruitment and reduced the resilience of Caribbean coral reefs (Mumby et al. 2007), which resulted in a stepwise degradation which continues today. After the 1980s die-off, Diadema recovery has been very poor, with populations estimated at around 12% of their former densities in 2015 (Lessios 2016). This slow recovery in combination with a new *Diadema* die-off in 2022 (Hylkema et al. 2023) sustained the functional extinction of *Diadema* on most Caribbean coral reefs.

The slow *Diadema* recovery is likely the result of a multitude of factors. Initially, Allee effects might have prevented effective fertilization (Lessios 1988). In addition, the removal of many populations resulted in downstream reductions in *Diadema* larval densities and settlement rates (Bak 1985). More recent studies focusing on *Diadema* settlement on artificial substrates demonstrated a lack of larval influx continued to constrain *Diadema* recovery in certain locations (Miller et al. 2009; Feehan et al. 2016). However, settlement rates in other locations were in the same order of magnitude as pre-die-off densities on Curacao (Williams et al. 2009, Vermeij et al. 2010, Hylkema et al. 2022a, Klokman & Hylkema 2024). On these locations, a high post-settlement and post-recruitment mortality, caused by high predation pressure (Harborne et al. 2009, Hylkema et al. 2022b) and/or low shelter availability (Bodmer et al. 2021), were the most likely causes for constrained *Diadema* recovery.

Settlement rates, predation pressure and shelter availability interact with the abundance of adult *Diadema*. Adults create a suitable habitat for juvenile conspecifics, for example by creating shelter for juveniles with their spine canopy (Miller et al. 2007). The intense grazing of adults creates bare substrate covered with a fresh biofilm, which is both an important settlement cue (Wijers et al. 2024) and an important food source for the settlers (Vermeij et al. 2010). On the other hand, a reef without adult *Diadema* is a less suitable habitat for settlers. Without the intense grazing of adults, turf and macroalgae fill small shelter spaces and reduce shelter availability (Spadaro & Butler 2021) while at the same time offering shelter to *Diadema* micro-predators (Bechtel et al. 2006) that reduce post-settlement survival.

To which extent settlement rates, predation pressure, and shelter availability affect *Diadema* recovery is essential knowledge to guide *Diadema* restoration approaches. As the removal of *Diadema* populations is undesired from a nature conservation perspective, it is difficult to study population recovery on locations that already have high *Diadema* densities, because the adults potentially interact with these factors. The 2022 *Diadema* die-off, with a lethality of 99% on some locations (Hylkema et al. 2023), provided the opportunity to study factors affecting natural recovery with minimal interacting effects of adult populations. In this case study we compared four locations near Saba, Caribbean Netherlands in settlement rates, predator density, shelter availability, and *Diadema* recovery after the 2022 die-off.

Methods

Locations

All research was conducted at four dive sites, Diadema City, Tent Reef, Ladder Bay, and Torrens Point, located at the southwestern side of Saba, Caribbean Netherlands (Figure 1). The dive sites were located within the Saba Marine Park and permission for this study was given by the Saba Conservation Foundation, management authority of the Saba Marine Park. Diadema City (Figure 2) consists of a former breakwater which was destroyed and turned into an artificial reef by hurricane Hugo in 1989. The breakwater was made of locally available rock in a variety of sizes. Diadema City is 100m long and 10-20m wide. Three semi-permanent 30m transects were established in a row from east to west on hard substrate at 8.1, 7.9 and 7.0m depth. Tent Reef consists of a reef plateau running east to west along the shore at 4-8m deep. Three semi-permanent 30m transects were established on the reef plateau at 5.8, 6.1 and 5.6m depth. Ladder Bay mostly consists of encrusted boulders of volcanic origin. Three semi-permanent transects were established over these boulders, aiming for hard substrate, at 10.0, 8.4 and 7.4m depth. Torrens Point consists of several lava fingers of 5-10m wide and up till 50m long, interspersed with volcanic boulders. Semi-permanent transects were established on the lava fingers at 9.4, 8.9 and 7.6m depth.

Diadema populations

Prior to the 2022 die-off, Diadema City contained the largest *Diadema* population around Saba (Hylkema et al. 2023). At Tent Reef, Ladder Bay, and Torrens Point, *Diadema* were occasionally observed prior to the 2022 die-off, but large populations were absent. *Diadema* populations were assessed at each location approximately one month after the 2022 *Diadema* die-off, in the period March – May 2022, and again one year after the die-off in March 2023. At each location, all *Diadema* within the three 30x2m semi-permanent transects were counted by two scuba-diving researchers.

Diadema settlement rates

Diadema settlement rates were determined monthly during the *Diadema* settlement season, which runs from April-October in this part of the Caribbean (Hylkema et al. 2022a, Klokman & Hylkema 2024), in 2022. At each location a sub-surface buoy and anchor were used to vertically keep a rope with pre-made loops at 8.0, 8.5, 9.0, 9.5 and 10.0m in the water column, following Klokman & Hylkema (2024). A settlement collector, consisting of 15 bio-balls strung on a nylon fishing line, was attached to each loop. Every month, the five settlement collectors deployed at each location were replaced with new collectors. Collected bio-balls were analyzed in the lab for *Diadema* settlers by carefully rinsing them in plastic trays following Hylkema et al. (2022a).

Fish predator density and presence

To determine the density of fish predators, three monitoring dives were conducted per location in the period May – October 2023. To select *Diadema* predators we used fish stomach content analyses from previous studies (Randall et al. 1964, Randall 1967) and included all fishes with *Diadema* or other echinoid material in their stomach (Table 1). At every survey, fish predators were counted and size estimated on all three semi-permanent 30x2m belt transects per location. The researcher was swimming with a constant speed of around 6min per transect using scuba.

After completion of the belt transects, a 20min roving diver survey was conducted by the same researcher to determine fish predators in a wider area (around 100x50m). The roving diver survey gave the opportunity to include shy species which typically are not observed in belt transects, such as queen triggerfish, and to include additional habitat such as ledges and sandy areas. During the roving diver survey, a researcher covered the indicated area in a zig-zag pattern swimming at a constant speed. All fishes from Table 1 were recorded and size estimated, except for bluehead and yellowhead wrasse, two very abundant small wrasses which were well represented within the belt transects.

Invertebrate predator presence and density

Invertebrate predators were surveyed during three nocturnal monitoring dives per location in the period May – October 2023. These dives were conducted at least 1h after sunset using scuba. During each survey, macro and micro invertebrate predators were counted and size estimated on the three 30x2m semi-permanent transects. Macro-invertebrate predators of *Diadema* include the spiny lobster *Panulirus argus* (Randall et al. 1964), the spotted spiny lobster *P. guttatus* (Kintzing & Butler 2014), the king helmet *Cassis tuberosa* (Levitan & Genovese 1989), the queen helmet *Cassis madagascariensis* (Randall et al. 1964) and the batwing crab *Carpilius corallinus* (Sharp & Reckenbeil 2022). Little is known about micro-predators of *Diadema*, but it can be assumed that most smaller crustaceans and fireworms predate on sea urchin settlers (Scheibling & Robinson 2008, Jennings & Hunt 2011) so these groups were included in the survey. During the monitoring dives, a single researcher inspected all shelter spaces along the transect using a Bigblue AL1300WP video light, recording all invertebrate predators.

Shelter availability

Shelter availability was determined using a modified version of the Point Intercept Contour (PIC) device modelled after Yanovski et al. (2017). The PIC device (Figure 3) reflects deviations from an artificial horizon by inserting 21 one-meter shafts at 5cm intervals, allowing detection of shelter spaces at ecologically relevant spatial scales for *Diadema* (Bodmer et al. 2021). The PIC device was deployed every 3m on each of the 30m semi-permanent transects, resulting in 10 deployments per transect and 27 deployments per location. At every deployment, the PIC device was placed on the reef, all shafts were inserted, and a photo was made from the side.

Analysis

Generalized Linear Mixed Models (GLMMs) with a Poisson distribution (glmer function with family = Poisson in the R package “lme4” (Bates et al. 2015) were used to test the effect of fixed factors year and location on the *Diadema* density. Transect_ID was added to the model as a random factor, to account for the fact that the same transects were surveyed in 2022 and 2023. Model selection was done based on Zuur et al. (2009) and Bolker et al. (2009). The model with the lowest Aikaike information criterion (AIC) was the model including both year, location, and their interaction. Model validation revealed that the model was not overdispersed. Wald $\chi 2$ tests were performed for statistical inference of the fixed factors (Bolker et al. 2009), using the Anova function of the R package “Car” (Fox & Weisberg 2019).

Generalized Linear Models (GLMs) with a Poisson distribution (glm function with family = Poisson in the R package “lme4”) were used to test the effect of fixed factors location, month and depth on the *Diadema* settlement rate per month. Model selection was done based on Zuur et al. (2009). The model with the lowest AIC was the model including only location and month, without depth or any interaction. Model validation revealed that the model was not overdispersed. Statistical inference was performed with likelihood ratio tests using the Anova function.

Fish predation pressure per survey and transect was calculated by using abundance data per species (density per $100m^2$) as well as known length-weight relationships (Bohnsack & Harper 1988) to get the *Diadema* predator biomass per species. Concurrently, the biomass per species was weighted using the average fraction of echinoid material in the stomach content of this specific species as described by Randall (1967), which is a slightly adapted version of the method developed by Harborne et al. (2009), who used the fraction of fish with *Diadema* contents to weigh their biomass estimates. We acknowledge that the study from Randall (1967) has been conducted in a time when *Diadema* were much more abundant, but more recent data about fish stomach contents was not available. The predation pressure per species per 100 m^2 was summed to get a total predation pressure per transect. Linear Mixed Models (LMMs, lmer function in the R package “lme4”) were used to test the effect of fixed factors location, survey number and their interaction on the fish predation pressure calculated from the transect surveys. Survey had a value of 1-3, as three surveys were conducted on all three transects of a location. To account for this dependency, transect_ID was added to the model as a random factor. Initial models showed a strong mean to variance relationship, which was solved by cube-root transforming the data. Model selection revealed that the model with location as only fixed factor had the lowest AIC (Zuur et al. 2009). For statistical inference, an F-test with Kenward-Roger’s approximation to degrees of freedom was performed.

For the invertebrate predators, a distinction was made between micro-predators (<3cm) and macro-predators (>3cm). Counts per taxonomic group were summed to get total micro- and macro-invertebrate densities per transect. General Linear Mixed Models (GLMMs) with a negative-binomial error distribution (glmer.nb fuction in the R package “lme4”) were used to test the effect of fixed factors location and survey number on the micro- and macro-invertebrate abundance. Survey had a value of 1-3, as three surveys were conducted on all three transects of a location. To account for this dependency, transect_ID was added to the model as a random factor. Model selection was done based on Zuur et al. (2009) and Bolker et al. (2009). The model with the lowest AIC was the model including both location, survey number and their interaction. Initially we used GLMMs with a Poisson distribution, but model validation (Zuur et al. 2009, Bolker et al. 2009) revealed substantial overdispersion, which was solved with using the negative-binomial distribution. Statistical inference was performed using likelihood ratio tests (Bolker et al. 2009) conducted with the drop1 function.

Photos from the PIC deployments were analyzed using ImageJ software. Based on Bodmer et al. (2021), we counted shelters that were at least 5cm deep and 5-15cm wide. Thus, negative deviations of the shafts to neighboring shafts greater than 5cm depth and consisting of one to a maximum of three shafts were considered a shelter (Figure 3). A one-way ANOVA, followed by Tukey *post-hoc* tests, was performed to compare shelter availability and depth per location. Model validation revealed non-normal distributed residuals and heterogeneity of variances for shelter depth, which was solved with a cube root transformation.

For LMM, GLMs and GLMMs, pairwise comparisons with a Tukey adjustment for multiple comparisons were conducted to examine significance of location using estimated marginal means (EMM) from the package “emmeans”. To compare the *Diadema* density between years per location, pairwise comparisons were conducted with location grouped within year. All analyses were done with R version 4.3.0 using Rstudio 2023.3.1.446. Graphs were made with the package “ggplot2”. Provided values are means ± standard error, while P-values <0.05 were considered statistically significant.

Results

Diadema populations

Location ($\chi2=23.6$, df=3, P<0.001), year ($\chi2=7.5$, df=1, P=0.006) and their interaction ($\chi2=23.4$, df=3, P<0.001) had a significant effect on the Diadema population. After the 2022 die-off, *Diadema* densities were less than 0.03 ± 0.02 *Diadema* per m^2 at all four locations and did not differ among each other (Figure 4A). One year later, in 2023, *Diadema* densities were very similar, except for Diadema City, where *Diadema* densities had increased to 1.4 ± 0.5 *Diadema* per m^2. This was significantly higher than all other locations (P<0.001 for all comparisons), which did not differ among each other.

Diadema settlement

Over the course of the study, 175 *Diadema* settlers were collected from the bio-ball collectors. Location (LRT=77.7, df=3, P<0.001) and month (LRT=217.9, df=5, P<0.001) had a significant effect on the number of *Diadema* settlers per collector (Figure 4B). Average settlement at Diadema City was 0.1 ± 0.1 *Diadema* per collector per month, which was significantly lower compared to all other locations (P<0.001 for all comparisons). Settlement at Ladder Bay was 2.6 ± 0.7 *Diadema* per collector per month, which was significantly higher than the 1.3 ± 0.4 found at Tent Reef (P=0.003), while settlement at Torrens Point (1.5 ± 0.4 *Diadema* per collector per month) did not differ from either Tent Reef or Ladder Bay.

Fish predators

Predation pressure, in this study calculated as the biomass of *Diadema* predators on the transects, weighted by the fraction of their stomach content consisting of echinoid remains (Randall 1967), ranged from 25 ± 9 gr per m^2 at Diadema City to 143 ± 73 gr per m^2 at Torrens Point (Figure 4C). Survey (not included in best fitting model) and location (F=1.4, df=3, P=0.260) did not significantly affect the predation pressure per m^2. At Diadema City, Ladder Bay and Torrens Point, grunts (Haemulidae), specifically Caesar grunt, black margate, and, to a lesser extent, French grunt, contributed most to the predation pressure (Table S2). At Tent Reef, wrasses (Labridae), specifically Spanish hogfish, puddingwife, and bluehead, contributed most to the predation pressure.

The results of the roving diver surveys were used to calculate predation pressure of the wider area, which ranged from 390 ± 175 gr per survey at Tent Reef to 2596 ± 498 gr per survey at Diadema City (Figure 4D). As roving diver predation pressure was the result of three surveys of the same area per location, statistical inference was not possible. At Diadema City, the predation pressure per survey was dominated by the black margate, followed by the Caesar grunt (Table S3). At Tent Reef, the Spanish hogfish contributed most to the predation pressure, while at Ladder Bay and Torrens Point both grunts and Spanish hogfish contributed to the predation pressure. During the roving diver surveys only one species, the porcupinefish *D. hystrix*, was observed in addition to the species already observed on the transects.

Invertebrate predators

Micro-predator (< 3cm) abundance (Figure 4E) was significantly affected by location (LRT=389.0, df=3, P<0.001), survey number (LRT=6.2, df=2, P=0.046), and the interaction between location and survey number (LRT=90.4, df=6, P<0.001). *Post-hoc* testing revealed that micro-predator abundance was highest at Diadema City, where 902 ± 147 micro-predators were recorded per 100 m^2. This was significantly higher compared to Tent Reef, Ladder Bay, and Torrens Point (P<0.0001 for all comparisons). At Tent Reef 59 ± 12 micro-predators per 100 m^2 were recorded, which was significantly more than the 31 ± 10 at Ladder Bay (P=0.003) and the

24 ± 9 at Torrens Point (P<0.001). Locations Ladder Bay and Torrens Point did not differ among each other. The majority of the micro-predators at each location were shrimp (Table S4) and the large difference between Diadema City and the other locations is explained by the much higher shrimp abundance at Diadema City.

Macro-predators (>3cm) abundance (Figure 4F) was not significantly affected by location (LRT=5.8, df=3, P=0.121) or survey number (not included in best fitting model) and ranged from 0.9 ± 0.3 macro-predators per 100 m^2 at both Tent Reef and Ladder Bay to 2.6 ± 0.7 macro-predators per 100m^2 at Diadema City. Spotted spiny lobster *P. guttatus* contributed most to the macro-predators abundance, followed by hermit crabs, Caribbean spiny lobster *P. argus*, and the king helmet *C. madagascariensis* (Table S4).

Shelter availability and depth

Shelter availability differed significantly per location (F=20.6, df=3, P<0.001, Figure 4G). Average shelter availability was with 1.3 ± 0.2 and 1.2 ± 0.1 shelter per meter highest at Torrens Point and Diadema City respectively. These locations did not differ significantly from each other but had significantly higher shelter availability compared to the 0.3 ± 0.1 shelter per m found at Tent Reef. Ladder Bay, with 0.8 ± 0.1 shelter per m did not differ from any of the other locations.

Shelter depth differed significantly per location (F=4.0, df=3, P<0.01, Figure 4H) and was highest at Diadema City where average shelter depth was 18.6 ± 2.0 cm. This was significantly higher compared to Tent Reef (P=0.034) and Torrens Point (P=0.0384), but not to Ladder Bay. These three locations all had average shelter depths around 10-12 cm.

Discussion

After the 2022 *Diadema* die-off, all locations had very low *Diadema* densities. One year later, Diadema City harbored, once again, a substantial *Diadema* population, while *Diadema* densities at all other locations remained similar to the year before. The recovery at Diadema City shows that the presence of adults is, in this case, not necessary to start population recovery. Based on this case study, the establishment of new *Diadema* populations is likely due to certain site characteristics, such as larval influx, predation pressure or shelter availability, which determine settlement, post-settlement survival, and population establishment. It has to be seen if the population at Diadema City will continue to recover. After the 1983-1984 die-off, *Diadema* populations in Panama first recovered slightly and then decreased further when recruitment remained low (Lessios 1988).

Settlement rates around Saba in 2022 (this study) were in the same order of magnitude as previous years (Hylkema et al. 2022b, Klokman & Hylkema 2024), which could be due to populations spawning before or even during the 2022 die-off or to spawning population located upstream and that have not been affected by the scuticociliate responsible for the mass mortality (Hewson et al. 2023). Settlement at Diadema City was significantly lower than at the other three locations, meaning larval influx was not a large determining factor in population recovery. This is in line with multiple other studies, showing high settlement on reefs without adult populations (Williams et al. 2010, Bodmer et al. 2015, Hylkema et al. 2022a, Klokman & Hylkema 2024). Williams et al. (2010) concluded that settlement is not a major determinant for recruitment, because settlement was relatively low on reefs with the highest adult populations and *vice versa*. However, as echinoids are known for their spatial and temporal variation in recruitment (Balch & Scheibling 2000), the observed discrepancy between settlement and adult populations could also have been the result of high settlement in previous years, followed by population establishment and an unrelated drop in settlement rates the year after. In the current study, the 2022 die-off killed 99% of the *Diadema* at Diadema City (Hylkema et al. 2023) and all surrounding reefs (personal observation both authors), ensuring that the gross majority of *Diadema* recorded in 2023 were the result of recruitment in 2022. This is, to our knowledge, the first record of *Diadema* population recovery under relatively low settlement rates.

Fish predation pressure measured on the semi-permanent transects was lowest at Diadema City, although it did not differ significantly from the other locations. Interestingly, the results of the roving diver survey showed the highest predation pressure per survey at Diadema City. This inverse pattern could be explained by the fish species contributing most to the predation pressure, which differed strongly per location. At Diadema City, grunts contributed most to the predation pressure, while at the other three locations wrasses had a more prominent role. While wrasses are diurnal and spread out over the reef during the day, grunts are nocturnal and spend the day near wall, overhang, or cave habitat. These habitats were mostly absent from the semi-permanent transects but included in the roving diver survey. Diadema City, constructed of large boulders, has a lot of this habitat and large schools of grunts, especially black margate and Caesar grunt, resulted in high roving diver predation pressure values at this location.

Predation pressure was calculated by multiplying estimated fish biomass per species with the fraction of echinoid content in their stomach (Randall 1967). This method has certain disadvantages. First, it assumes that preference of fishes did not change significantly compared to the situation in the 1960s, when *Diadema* were much more abundant (Harborne et al. 2009). After the 1983-1984 die-off, fishes had to shift their diet to other prey (Reinthal et al. 1984, Robertson 1987). It is however unclear if these species still retain their original preference for *Diadema* (Lessios 1988). A study identifying predators of restocked *Diadema* on artificial reefs revealed that only queen triggerfish and Spanish hogfish attacked the restocked *Diadema*, while grunts did not seem to be attracted by the introduced animals (de Breuyn et al. 2023). However, only daytime observations were made, possibly excluding the predation behavior of the mostly nocturnal grunts. A more recent study, following 200 restocked *Diadema* on Saba in 2023, included both day- and night-time observations from divers and cameras (Wijers et al. 2024). All recorded attacks were conducted by the Spanish hogfish. Multiple grunts were observed in the close vicinity of the restocking reef, but these fishes did not seem to be attracted by the *Diadema* (Wijers et al. 2024). It could be that dietary preference of grunts shifted to other species after the 1983-1984 *Diadema* die-off, meaning that the current calculation method overestimates their role in the predation pressure. At the same time, Wijers et al. (2024) clearly show that Spanish hogfish can be detrimental to the restocked *Diadema*. According to the roving diver survey, Spanish hogfish were almost absent at Diadema City, but abundant on all three other locations. It could be that Spanish hogfish densities are an essential factor in *Diadema* population establishment but that its effect on the calculated predation pressure was compensated for by the abundance of grunts at Diadema City.

The second disadvantage of the method employed for predation pressure is that it does not distinguish between different life stages of *Diadema*. Some species, like certain wrasses, prefer small *Diadema* juveniles (Lessios 1988), while others, like the queen triggerfish, prefer larger individuals (de Breuyn et al. 2023). However, predating on a large number of settlers might remove less *Diadema* biomass, but have a bigger effect on the *Diadema* population compared to the removal of a few larger individuals. With the present calculation method, abundant but small wrasses, with a stomach content containing only a small fraction of echinoid material (Randall 1967), have a minimal effect on the calculated predation pressure, while larger grunts or triggerfish which are likely to prey on larger adult *Diadema*, have a far larger effect. There are some indications that wrasses, such as blueheads and yellowhead wrasses predate on small juveniles and hamper recovery of *Diadema* (Lessios 1988, Rodríguez-Barreras et al. 2015) and their effect might be underestimated in the current metrics. Blueheads and yellowhead wrasses were the most abundant predators on all locations. However, their abundance at Diadema City was 2-5 times lower compared to all other locations, which might contribute to the population recovery at this artificial reef location.

Invertebrate micro-predators (<3cm) such as shrimps, crabs, and fireworms are often assumed to have an inhibiting role on *Diadema* recovery (Williams et al. 2011, Hylkema et al. 2022b), although the magnitude of their role is not yet understood (Bechtel et al. 2006). Of the four locations included in this study, Diadema City had a significantly higher invertebrate micro-predator abundance. This difference was entirely driven by the shrimp density. Shrimps were the most numerous micro-predators at all locations but were 20-30 times more abundant at Diadema City. However, this difference can largely be attributed to the high abundance of red night shrimps (*Cinetorhynchus manningi*) sheltering and foraging underneath the spines of the larger *Diadema*. During the invertebrate surveys, conducted in 2023 when the *Diadema* population at Diadema City had partly recovered, most adult *Diadema* had 5-10 red night shrimp around them, which retracted underneath the *Diadema* spines when disturbed (personal observation both authors). Little is known about the dietary preferences of red night shrimp or other Rhynchocinetidae (Dumont et al. 2009), but they are likely to be opportunistic feeders grazing on detritus and sessile invertebrates such as bryozoans and ascidians (Dumont et al. 2009). However, it cannot be excluded that they also prey on *Diadema* setters. Although micro-predators were recorded per infraorder and not per species, removing the red night shrimps from the observations would very likely have removed the difference among locations.

Other micro-invertebrates such as crabs (Harrold et al. 1991, Scheibling & Robinson 2008, Clemente et al. 2013) and fireworms (Simonini et al. 2017) are known to be important predators of juvenile other sea urchin species. Fireworms have also been observed eating *Diadema* which were still alive by both authors. However, on our transects, few of these groups of micro-predators were encountered. Densities of hermit crabs and other crabs were 0-2 per 100 m^2, while not a single fireworm was observed during the study. The lack of observations of these probably abundant micro-invertebrates can possibly be explained by their cryptic nature; even at night they might be hiding and could easily be overlooked. If this is the case, the employed method is not adequate and other methods should be used to acquire an idea of their abundance. Micro-invertebrate traps could be a suitable alternative. These traps could be baited or just provide refuge (Osawa et al. 2015, Moraes et al. 2022). Refuge traps might provide the best representation of the local micro-invertebrate abundance and diversity, although the catch will remain a sub-sample of the actual assemblage (Moreira-Ferreira et al. 2020).

Observations of invertebrate macro-predators (>3cm) included spotted spiny lobster, hermit crabs, Caribbean spiny lobster, and king helmet. Location did not significantly affect the macro-predator abundance. Outside the transects, the large crevices at Diadema City supported high numbers of both spotted and Caribbean spiny lobsters. Often, Caribbean spiny lobsters were seen in the same crevice as adult *Diadema*, literally 20cm from each other. These observations make it unlikely that Caribbean spiny lobsters are an important predator of adult *Diadema*. This confirms the findings of Kintzing & Butler (2014), who showed that *Diadema* flee for the scent of spotted spiny lobster, but not from Caribbean spiny lobsters. Although Caribbean spiny lobsters forage mostly off the reef, it remains possible that they consume smaller *Diadema*, as was also reported by Randall (1964). Contrary to its larger relative, spotted spiny lobster forage mostly on the reef and are known to predate on smaller *Diadema* (Kintzing & Butler 2014). However, the high densities of spotted spiny lobster at Diadema City did not prevent *Diadema* population recovery. Possibly, their role as *Diadema* predator is overestimated, but it could also be that their predation pressure on *Diadema* is mitigated by their predation on crabs, which form most of their diet (Butler & Kintzing 2015). Paradoxically, this would mean that the high spotted spiny lobster abundance at Diadema City might result in a lower net predation pressure on *Diadema* compared to other locations.

Shelter availability and shelter depth varied highly among and within locations. Diadema City and Torrens Point had the highest shelter availability, while Diadema City and Ladder Bay had the deepest shelters. These beneficial shelter characteristics might have been positive for the recovering *Diadema* population at Diadema City, as habitat complexity positively correlated with *Diadema* densities at other locations (Bodmer et al. 2015, Bodmer et al. 2021). Shelter availability for *Diadema* is difficult to parameterize and we realize that the by us used shelter density and depth are a good approximation at best. A parameter that we did not include, is almost impossible to quantify, but might be very important is the shape of shelters. We observed that Diadema City had a high availability of wedge formed shelters, a result of two or more rocks piled on top of each other. These wedge-shelters were not often observed on the other locations. While a rounded cup-formed shelter will only provide adequate shelter to a certain size class of *Diadema* (Bodmer et al. 2021), a wedge-shaped shelter provides shelter for a large range of size classes. *Diadema* settlers can retract all the way to the end of the wedge and slowly emerge while they grow, continuously being protected by the same shelter. We hypothesize that not only the high shelter availability and large depth, but also the shelter shape at Diadema City has positively affected *Diadema* population recovery.

A lingering question is why *Diadema* have difficulties recovering at the same reefs on which they were so abundant before the 1980s die-off. A possible explanation for this is that *Diadema* predators might have a larger effect when the population size of their prey is smaller (Ricklefs 2001). However, the quick recovery at Diadema City after the 2022 die-off tells there is more to the story. Caribbean coral reefs have severely degraded and the reefs of the 2020's are widely different to those of the 1970s. The decrease in scleractinian coral cover has resulted in a severe reduction in coral reef complexity, reducing shelter availability (Alvarez-Filip et al. 2009, Magel et al. 2019). Possibly, the wedge-shaped shelter provided by the piled rocks at Diadema City was previously provided by scleractinian corals, for example plate forming *Agaricia* sp. or *Orbicella* sp. (Bodmer et al. 2015). Another possible explanation is changes in fish assemblages caused by fishing and by changes in the earlier described complexity of the reef. While overfishing *Diadema* predators might be favorable for *Diadema* recovery (Harborne et al. 2009), overfishing of certain top predators might relax predation pressure over meso-predatory wrasses, which concurrently might result in increased predation on *Diadema* (Rodríguez-Barreras et al. 2015).

This study provides some indications why *Diadema* recovered at a man-made artificial reef, despite having lower *Diadema* settlement rates, and not at nearby natural reefs. Although the artificial reef had a high density of *Diadema* fish predators, densities of Spanish hogfish and smaller wrasses were lower than at natural reef locations. Other studies indicated that especially these fishes, in combination with the queen triggerfish, might be essential in determining *Diadema* population recovery. Invertebrate predator density was similar at all locations, except for the red night shrimp and the spotted spiny lobster, which were more abundant at the artificial reef. Spotted spiny lobster are known predators of smaller *Diadema*, but the large percentage of crabs in their diet might result in a net positive effect on *Diadema* populations. Finally, the shelter availability and depth were more favorable at the artificial reef location. In addition, we hypothesize that the large number of wedge-shaped shelters at the artificial reef also positively affected Diadema population recovery. It cannot be excluded that suitable shelter characteristics compensate for increased predation pressure at the artificial reef. We conclude that *Diadema* population establishment is likely determined by distinct habitat characteristics. A more regional approach, including multiple islands and locations with and without *Diadema,* might help to further detangle the factors affecting natural recovery. If this would be pursued, we advise to analyze predation pressure by wrasses and queen triggerfish separately, to avoid larger bodied predators cancelling out potential differences in predation pressure. Micro-invertebrate predation should be quantified by different means, for example by refuge traps, while the shelter shape quantification should be included if possible. The identification of factors hindering natural recovery are essential to select future *Diadema* restocking locations, thereby increasing the success chance of *Diadema* restoration, and enabling an increase of Caribbean coral reefs resilience.

Acknowledgements

We are grateful to the SCF staff, especially Marijn van der Laan, for their assistance in the field and to Rosemarijn van de Lint, Jasper Raijmakers, Bai Klap, Jelle van der Velde, Jethro van't Hul, Pauline Raimbault, and Peter Johnson who helped with collecting the data.

References

Alvarez-Filip, L., Dulvy, N. K., Gill, J. A., Côté, I. M., & Watkinson, A. R. (2009). Flattening of Caribbean coral reefs: region-wide declines in architectural complexity. *Proceedings of the Royal Society B: Biological Sciences*, *276*(1669), 3019-3025.

Bak, R. P. M. (1985). Recruitment patterns and mass mortalities in the sea urchin *Diadema antillarum*. *Proc. 5th int. coral reef congress*, Vol. 5. (Tahiti),267–272.

Balch, T., & Scheibling, R. E. (2000). Temporal and spatial variability in settlement and recruitment of echinoderms in kelp beds and barrens in Nova Scotia. *Marine Ecology Progress Series*, *205*, 139-154.

Bates, D., Maechler, M., Bolker, B., Walker, S., Christensen, R. H. B., Singmann, H., ... & Bolker, M. B. (2015). Package 'lme4'. *convergence*, *12*(1), 2.

Bechtel, J. D., Gayle, P., & Kaufman, L. (2006, June). The return of Diadema antillarum to Discovery Bay: patterns of distribution and abundance. In *Proc 10th Int Coral Reef Symp* (Vol. 1, pp. 367-375).

Bodmer, M. D., Rogers, A. D., Speight, M. R., Lubbock, N., & Exton, D. A. (2015). Using an isolated population boom to explore barriers to recovery in the keystone Caribbean coral reef herbivore Diadema antillarum. *Coral Reefs*, *34*, 1011-1021.

Bodmer, M. D. V., Wheeler, P. M., Anand, P., Cameron, S. E., Hintikka, S., Cai, W., ... & Exton, D. A. (2021). The ecological importance of habitat complexity to the Caribbean coral reef herbivore Diadema antillarum: Three lines of evidence. *Scientific reports*, *11*(1), 9382.

Bohnsack, J. A., & Harper, D. E. (1988). Length-weight relationships of selected marine reef fishes from the southeastern United States and the Caribbean.

Bolker, B. M., Brooks, M. E., Clark, C. J., Geange, S. W., Poulsen, J. R., Stevens, M. H. H., & White, J. S. S. (2009). Generalized linear mixed models: a practical guide for ecology and evolution. *Trends in ecology & evolution*, *24*(3), 127-135.

de Breuyn, M., van der Last, A. J., Klokman, O. J., & Hylkema, A. (2023). Diurnal predators of restocked lab-reared and wild Diadema antillarum near artificial reefs in Saba. *PeerJ*, *11*, e16189.

Butler IV, M. J., & Kintzing, M. D. (2015). An exception to the rule: top-down control of a coral reef macroinvertebrate community by a tropical spiny lobster. *Bulletin of Marine Science*, *92*(1), 137-152.

Carpenter, R. C. (1988). Mass mortality of a Caribbean sea urchin: immediate effects on community metabolism and other herbivores. *Proceedings of the National Academy of Sciences*, *85*(2), 511-514.

Carpenter, R. C. (1990a). Mass mortality of Diadema antillarum: II. Effects on population densities and grazing intensity of parrotfishes and surgeonfishes. *Marine Biology*, *104*, 79-86.

Carpenter, R. C. (1990b). Mass mortality of Diadema antillarum: I. Long-term effects on sea urchin population-dynamics and coral reef algal communities. *Marine Biology*, *104*, 67-77.

Clemente, S., Hernández, J. C., Montaño-Moctezuma, G., Russell, M. P., & Ebert, T. A. (2013). Predators of juvenile sea urchins and the effect of habitat refuges. *Marine Biology*, *160*, 579-590.

Dumont, C. P., Urriago, J. D., Abarca, A., Gaymer, C. F., & Thiel, M. (2009). The native rock shrimp Rhynchocinetes typus as a biological control of fouling in suspended scallop cultures. *Aquaculture*, *292*(1-2), 74-79.

Feehan, C. J., Brown, M. S., Sharp, W. C., Lauzon-Guay, J. S., & Adams, D. K. (2016). Fertilization limitation of Diadema antillarum on coral reefs in the Florida Keys. Ecology, 97(8), 1897-1904. doi: 10.1002 /ecy.1461

Fox, J., & Weisberg, S. (2020). Using car and effects Functions in Other Functions. *Using Car Eff. Funct. Other Funct*, *3*, 1-5.

Harborne, A. R., Renaud, P. G., Tyler, E. H. M., & Mumby, P. J. (2009). Reduced density of the herbivorous urchin Diadema antillarum inside a Caribbean marine reserve linked to increased predation pressure by fishes. *Coral Reefs*, *28*, 783-791.

Harrold, C., Lisin, S., Light, K. H., & Tudor, S. (1991). Isolating settlement from recruitment of sea urchins. *Journal of Experimental Marine Biology and Ecology*, *147*(1), 81-94.

Hewson, I., Ritchie, I. T., Evans, J. S., Altera, A., Behringer, D., Bowman, E., ... & Breitbart, M. (2023). A scuticociliate causes mass mortality of Diadema antillarum in the Caribbean Sea. *Science Advances*, *9*(16), eadg3200.

Hughes, T. P., Reed, D. C., & Boyle, M. J. (1987). Herbivory on coral reefs: community structure following mass mortalities of sea urchins. *Journal of Experimental Marine Biology and Ecology*, *113*(1), 39-59..

Hylkema, A., Debrot, A. O., Pistor, M., Postma, E., Williams, S. M., & Kitson-Walters, K. (2022a). High peak settlement of *Diadema antillarum* on different artificial collectors in the Eastern Caribbean. *Journal of Experimental Marine Biology and Ecology*, *549*, 151693.

Hylkema, A., Debrot, A. O., Van De Pas, E. E., Osinga, R., & Murk, A. J. (2022b). Assisted natural recovery: a novel approach to enhance *Diadema antillarum* recruitment. *Frontiers in Marine Science*, *9*, 929355.

Hylkema, A., Kitson-Walters, K., Kramer, P. R., Patterson, J. T., Roth, L., Sevier, M. L., ... & Lang, J. C. (2023). The 2022 Diadema antillarum die-off event: Comparisons with the 1983-1984 mass mortality. *Frontiers in Marine Science*, *9*, 1067449.

Jennings, L. B., & Hunt, H. L. (2011). Small macrobenthic invertebrates affect the mortality and growth of early post-settlement sea urchins and sea stars in subtidal cobble habitat. *Marine Ecology Progress Series*, *431*, 173-182.

Kintzing, M. D., & Butler IV, M. J. (2014). Effects of predation upon the long-spined sea urchin Diadema antillarum by the spotted spiny lobster *Panulirus guttatus*. *Marine Ecology Progress Series*, *495*, 185-191.

Klokman, O. J., & Hylkema, A. (2024). Spatial distribution of settlement of Diadema antillarum around Saba, Dutch Caribbean. *PeerJ*, *12*, e17291.

Lessios, H. A. (1988). Population dynamics of Diadema antillarum (Echinodermata: Echinoidea) following mass mortality in Panama. *Marine Biology*, *99*, 515-526.

Lessios, H. A. (2016). The great Diadema antillarum die-off: 30 years later. *Annual review of marine science*, *8*, 267-283.

Lessios, H. A., Garrido, M. J., & Kessing, B. D. (2001). Demographic history of Diadema antillarum, a keystone herbivore on Caribbean reefs. *Proceedings of the Royal Society of London. Series B: Biological Sciences*, *268*(1483), 2347-2353.

Levitan, D. R. (1988). Algal-urchin biomass responses following mass mortality of Diadema antillarum Philippi at Saint John, US Virgin Islands. *Journal of Experimental Marine Biology and Ecology*, *119*(2), 167-178.

Levitan, D. R., & Genovese, S. J. (1989). Substratum-dependent predator-prey dynamics: patch reefs as refuges from gastropod predation. *Journal of Experimental Marine Biology and Ecology*, *130*(2), 111-118.

Magel, J. M., Burns, J. H., Gates, R. D., & Baum, J. K. (2019). Effects of bleaching-associated mass coral mortality on reef structural complexity across a gradient of local disturbance. *Scientific Reports*, *9*(1), 2512.

McCook, L., Jompa, J., & Diaz-Pulido, G. (2001). Competition between corals and algae on coral reefs: a review of evidence and mechanisms. *Coral reefs*, *19*, 400-417.

Miller, R. J., Adams, A. J., Ebersole, J. P., & Ruiz, E. (2007). Evidence for positive density-dependent effects in recovering Diadema antillarum populations. *Journal of experimental marine biology and ecology*, *349*(2), 215-222.

Miller, M. W., Kramer, K. L., Williams, S. M., Johnston, L., & Szmant, A. M. (2009). Assessment of current rates of Diadema antillarum larval settlement. *Coral Reefs*, 28(2), 511-515. DOI: 10.1007/s00338-008-0458-4

Moraes, I. R., Davanso, T. M., Silva, A. R. D., Cobo, V. J., Alves, D. F., Santana, W., ... & Castilho, A. L. (2022). A first report of decapod crustaceans (Anomura and Brachyura) from Laje de Santos: a no-take marine reserve in the southeast coast of Brazil. *Revista mexicana de biodiversidad*, *93*.

Moreira-Ferreira, B., Selhorst, Y., Almeida, L. C. D., Amaral, J. R., Silva, É. F. D., Queiroz, I. R., ... & Zandonà, E. (2020). Evaluating two sampling methodologies for shrimp density and biomass estimates in streams. *Nauplius*, *28*, e2020042.

Mumby, P. J., Hastings, A., & Edwards, H. J. (2007). Thresholds and the resilience of Caribbean coral reefs. *Nature*, *450*(7166), 98-101.

Osawa, Y., Aoki, M. N., Bauer, R. T., & Thiel, M. (2015). Numbers and sizes of the shrimp Rhynchocinetes Uritai (Decapoda: Caridea) caught in bait and refuge traps. *Journal of Crustacean Biology*, *35*(6), 768-775.

Pandolfi, J. M., Bradbury, R. H., Sala, E., Hughes, T. P., Bjorndal, K. A., Cooke, R. G., ... & Jackson, J. B. (2003). Global trajectories of the long-term decline of coral reef ecosystems. *Science*, *301*(5635), 955-958.

Randall, J. E., Schroeder, R. E., & Starck, W. A. (1964). Notes on the biology of the echinoid *Diadema antillarum*. *Caribbean Journal of Science*, *4*(2-3), 421-33.

Randall, J. E. (1967). *Food habits of reef fishes of the West Indies* (Vol. 5, pp. 665-847). Coral Gables: Institute of Marine Sciences, University of Miami.

Reinthal, P. N., Kensley, B., & Lewis, S. M. (1984). Dietary shifts in the queen triggerfish, Balistes vetula, in the absence of its primary food item, Diadema antillarum. *Marine Ecology*, *5*(2), 191-195.

Ricklefs, R.E. 2001. The Economy of Nature, Chapter 18: Dynamics of Predation. 5th edition. New York: W.H. Freeman and Company. 550 pages.

Riegl, B., Bruckner, A., Coles, S. L., Renaud, P., & Dodge, R. E. (2009). Coral reefs: threats and conservation in an era of global change. *Annals of the New York Academy of Sciences*, *1162*(1), 136-186.

Robertson, D. R. (1987). Responses of two coral reef toadfishes (Batrachoididae) to the demise of their primary prey, the sea urchin Diadema antillarum. *Copeia*, 637-642.

Robertson, D. R. (1991). Increases in surgeonfish populations after mass mortality of the sea urchin Diadema antillarum in Panama indicate food limitation. *Marine Biology*, *111*, 437-444.

Rodríguez-Barreras, R., Durán, A., Lopéz-Morell, J., & Sabat, A. M. (2015). Effect of fish removal on the abundance and size structure of the sea urchin Diadema antillarum: a field experiment. *Marine Biology Research*, *11*(10), 1100-1107.

De Ruyter van Steveninck, E. D., & Bak, R. P. M. (1986). Changes in abundance of coral-reef bottom components related to mass mortality of the sea urchin Diadema antillarum. *Marine Ecology Progress Series*, 87-94.

Scheibling, R. E., & Robinson, M. C. (2008). Settlement behaviour and early post-settlement predation of the sea urchin Strongylocentrotus droebachiensis. *Journal of Experimental Marine Biology and Ecology*, *365*(1), 59-66.

Sharp, W. C., & Reckenbeil, B. A. (2022). Predation on a newly-stocked long-spined sea urchin (Diadema antillarum) by the batwing coral crab (Carpilius corallinus) on a coral reef restoration site. *Bulletin of Marine Science*, *98*(1), 99-100.

Simonini, R., Righi, S., Maletti, I., Fai, S., & Prevedelli, D. (2017). Bearded versus thorny: the fireworm Hermodice carunculata preys on the sea urchin Paracentrotus lividus. *Ecology*, *98*(10), 2730-2732.

Spadaro, A. J., & Butler, M. J. (2021). Herbivorous crabs reverse the seaweed dilemma on coral reefs. *Current Biology*, *31*(4), 853-859.

Vermeij, M. J., Debrot, A. O., van der Hal, N., Bakker, J., & Bak, R. P. (2010). Increased recruitment rates indicate recovering populations of the sea urchin Diadema antillarum on Curaçao. *Bulletin of Marine Science*, *86*(3), 719-725.

Wijers, T., Hylkema, A., Pilnick, A. R., Murk, A. J., & Patterson, J. T. (2023). Novel shaker bottle cultivation method for the long spined sea urchin (Diadema antillarum; Philippi, 1845) results in high larval survival and settlement rates. *Aquaculture*, *562*, 738855.

Wijers, T., van Herpen, B., Mattijssen, D., Murk, A. J., Patterson, J. T., & Hylkema, A. (2024). Implications of changing Caribbean coral reefs on Diadema antillarum larvae settlement. *Marine Biology*, *171*(2), 48.

Wijers, T., Klokman, O., Murk, A. J., Sijbrandij, J., & Hylkema, A. (2024). Low retention of restocked laboratory-reared long-spined sea urchins Diadema antillarum due to Spanish hogfish Bodianus rufus predation. *Restoration Ecology*, e14264.

Williams, S. M., García-Sais, J. R., & Capella, J. (2009). Temporal variation of early larval stages of the long-spined sea urchin Diadema antillarum in La Parguera, Puerto Rico. *Caribbean Journal of Science*, *45*(1), 110-117.

Williams, S. M., Yoshioka, P. M., & García Sais, J. R. (2010). Recruitment pattern of D*iadema antillarum* in la parguera, Puerto Rico. *Coral Reefs*, *29*, 809-812.

Williams, S. M., García-Sais, J. R., & Yoshioka, P. M. (2011). Spatial variation of *Diadema antillarum* settlement in la parguera, puerto rico. *Bulletin of Marine Science*, *87*(3), 531-540.

Yanovski, R., Nelson, P. A., & Abelson, A. (2017). Structural complexity in coral reefs: examination of a novel evaluation tool on different spatial scales. *Frontiers in Ecology and Evolution*, *5*, 27.

Zuur, A. F., Ieno, E. N., Walker, N. J., Saveliev, A. A., & Smith, G. M. (2009). *Mixed effects models and extensions in ecology with R* (Vol. 574, p. 574). New York: springer.

Table 1(on next page)

Diadema predators

List of fish species that are known to predate on *Diadema* (Randall et al. 1964, Randall 1967) and the percentage echinoid material in their stomach based on Randall (1967).

Table 1: List of fish species that are known to predate on Diadema (Randall et al. 1964, Randall 1967) and the percentage echinoid material in their stomach based on Randall (1967).

Common name	**Scientific name**	**Echinoid material in stomach (%)**
Queen triggerfish	*Balistes vetula*	72.8
Ocean triggerfish	*Canthidermis maculata*	25.0
Spanish hogfish	*Bodianus rufus*	14.4
Puddingwife	*Halichoeres radiatus*	19.9
Slippery dick	*Halichoeres bivittatus*	17.9
Yellowhead wrasse	*Halichoeres garnoti*	3.0
Black-ear wrasse	*Halichoeres poeyi*	6.8
Bluehead	*Thalassoma bifasciatum*	1.5
Porcupinefish	*Diodon hystrix*	34.6
Spotted trunkfish	*Lactophrys bicaudalis*	10.0
Smooth trunkfish	*Lactophrys triqueter*	2.3
Black margate	*Anisotremus surinamensis*	53.5
Spanish grunt	*Haemulon macrostomum*	86.8
Caesar grunt	*Haemulon carbonarium*	10.9
French grunt	*Haemulon flavolineatum*	1.5
Bluestriped grunt	*Haemulon sciurus*	8.7
White grunt	*Haemulon plumierii*	12.4
Joltheat porgy	*Calamus bajonado*	45.2
Saucereye porgy	*Calamus calamus*	8.9
Bandtail puffer	*Sphoeroides spengleri*	6.9
Sharpnose puffer	*Canthigaster rostrata*	3.8

Figure 1

Map of study locations

Map indicating the four locations near Saba, Caribbean Netherlands, and the location of Saba in the Caribbean region.

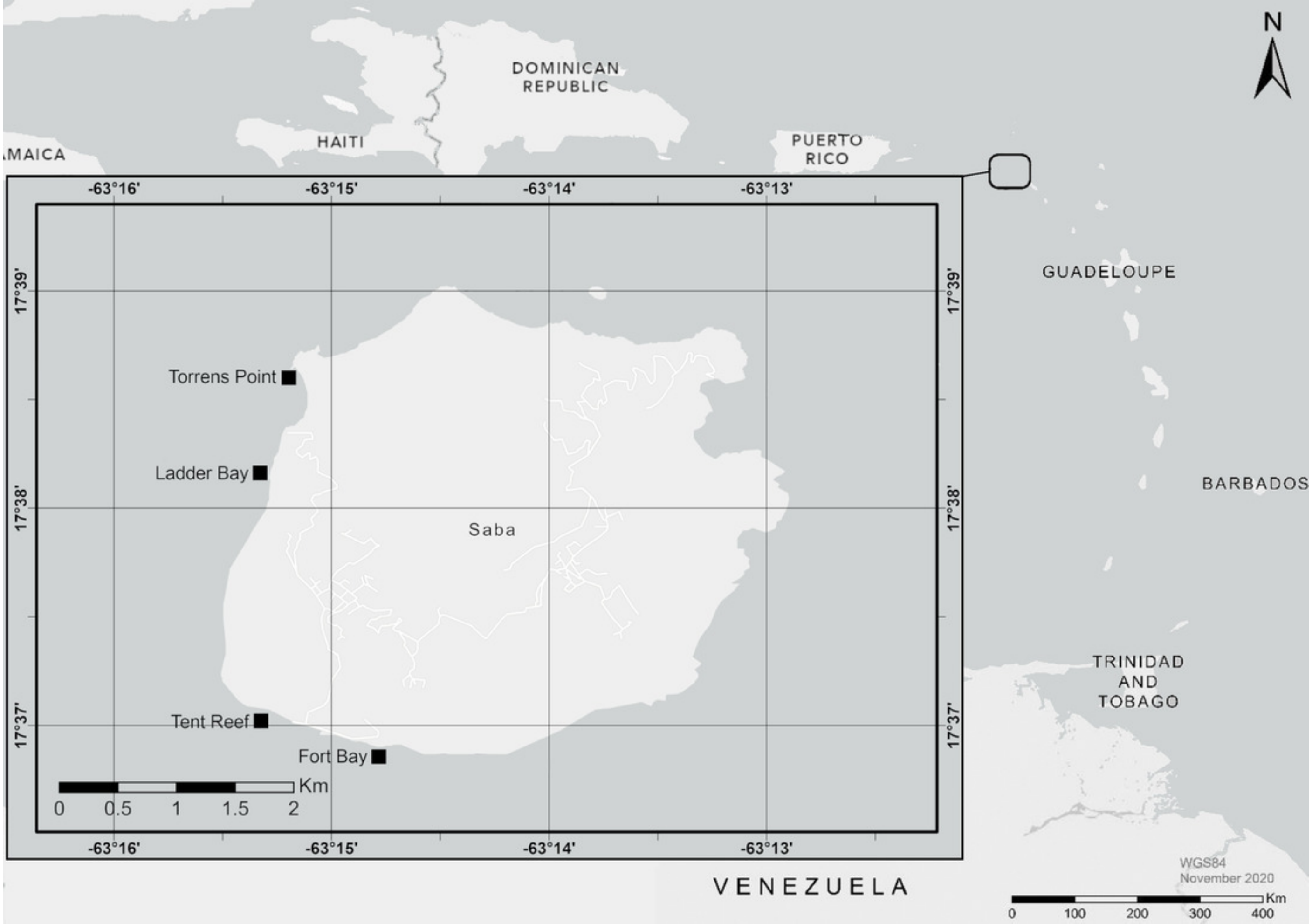


Figure 2

Four research locations

Overview photos of the four research locations: Diadema City (A), Tent Reef (B), Ladder Bay (C), and Torrens Point (D).

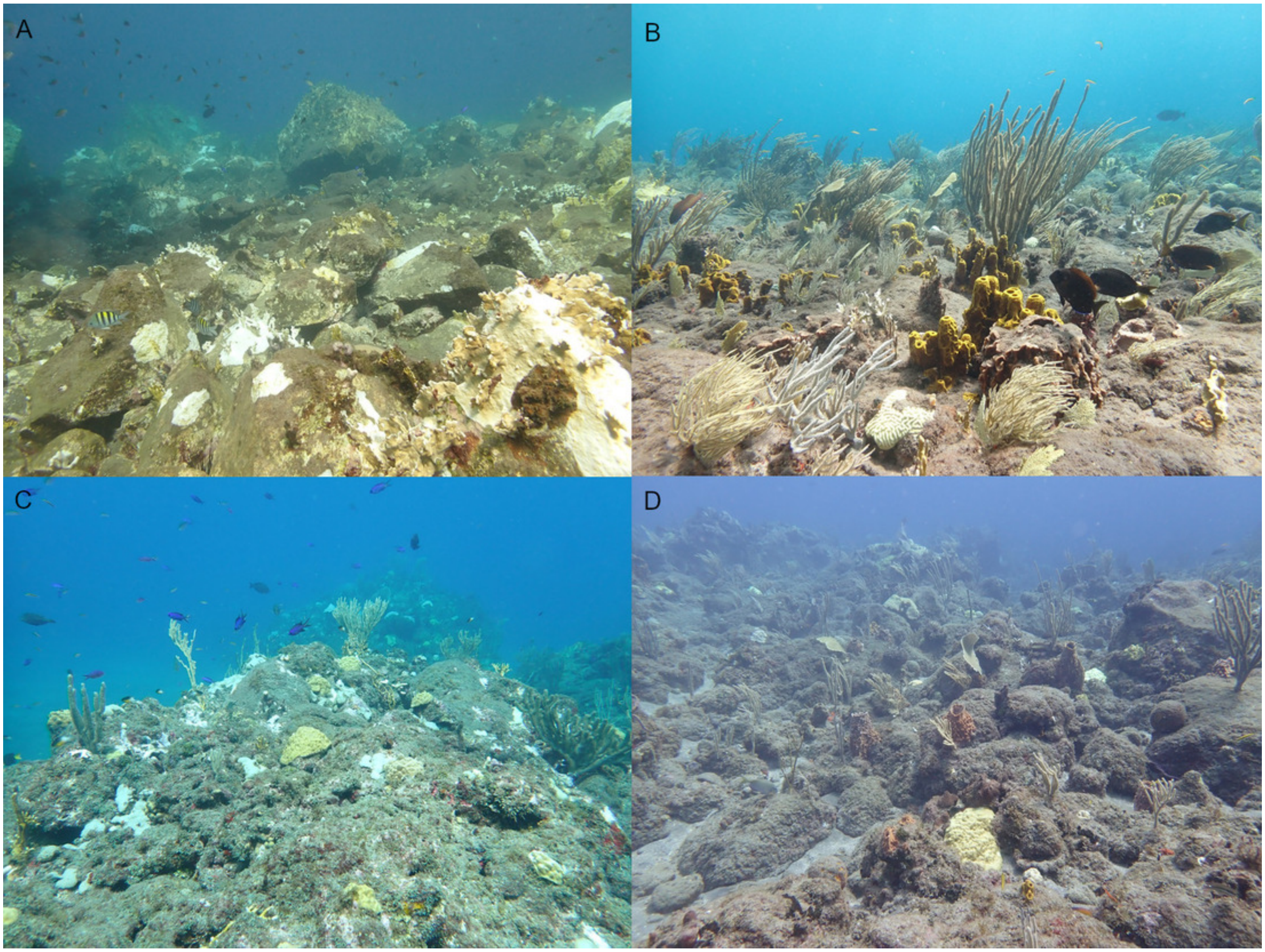


Figure 3

Point Intercept Countour device

Deployed Point Intercept Countour device modelled after Yanovski et al. (2017). Shafts are 1m in length and marked areas are 10cm. Shelter for Diadema was defined as at least 5cm deep and 5-15cm wide (Bodmer et al. 2021). Based on these criteria, two shelters were identified on this photo (white arrows).

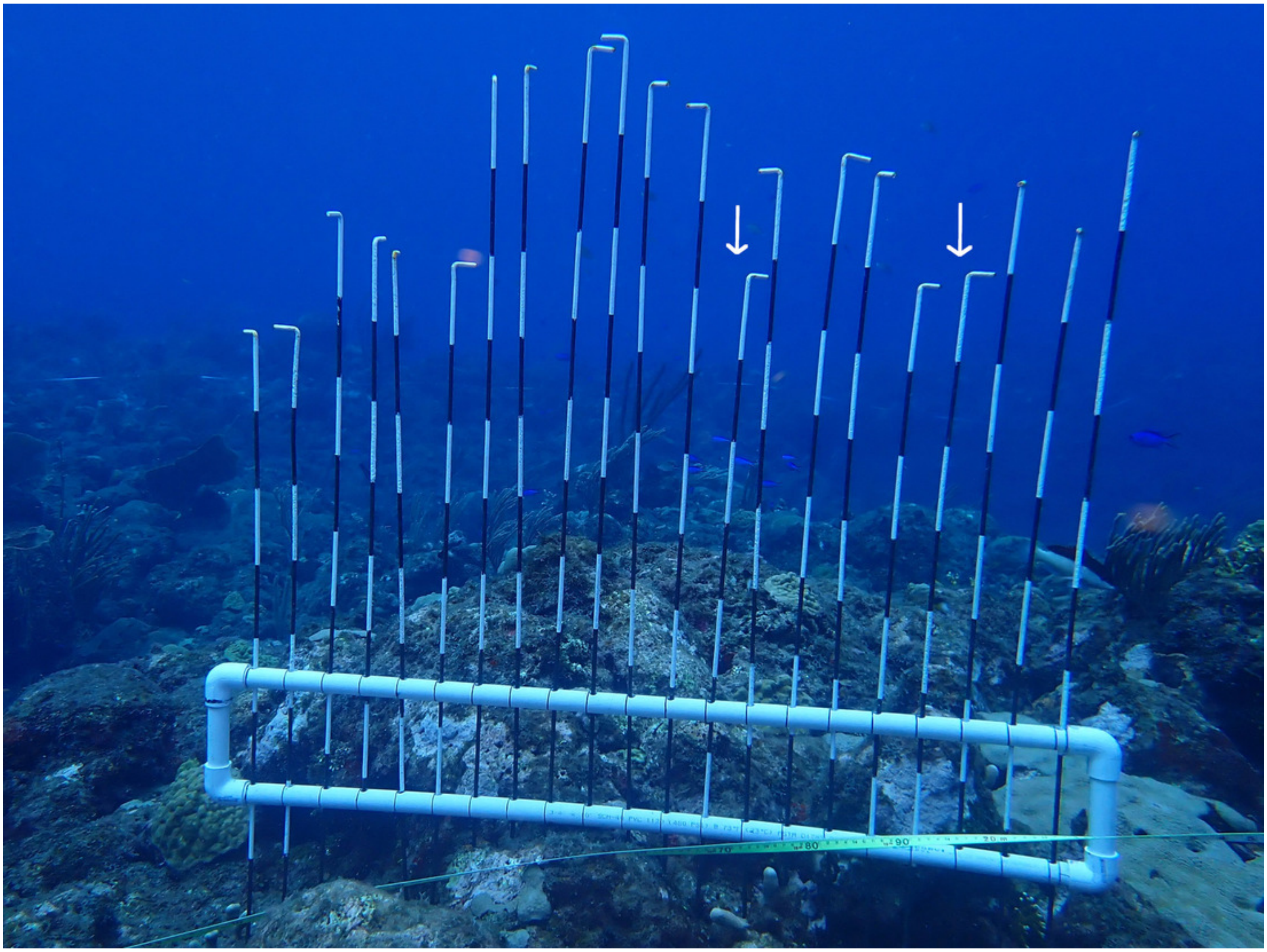
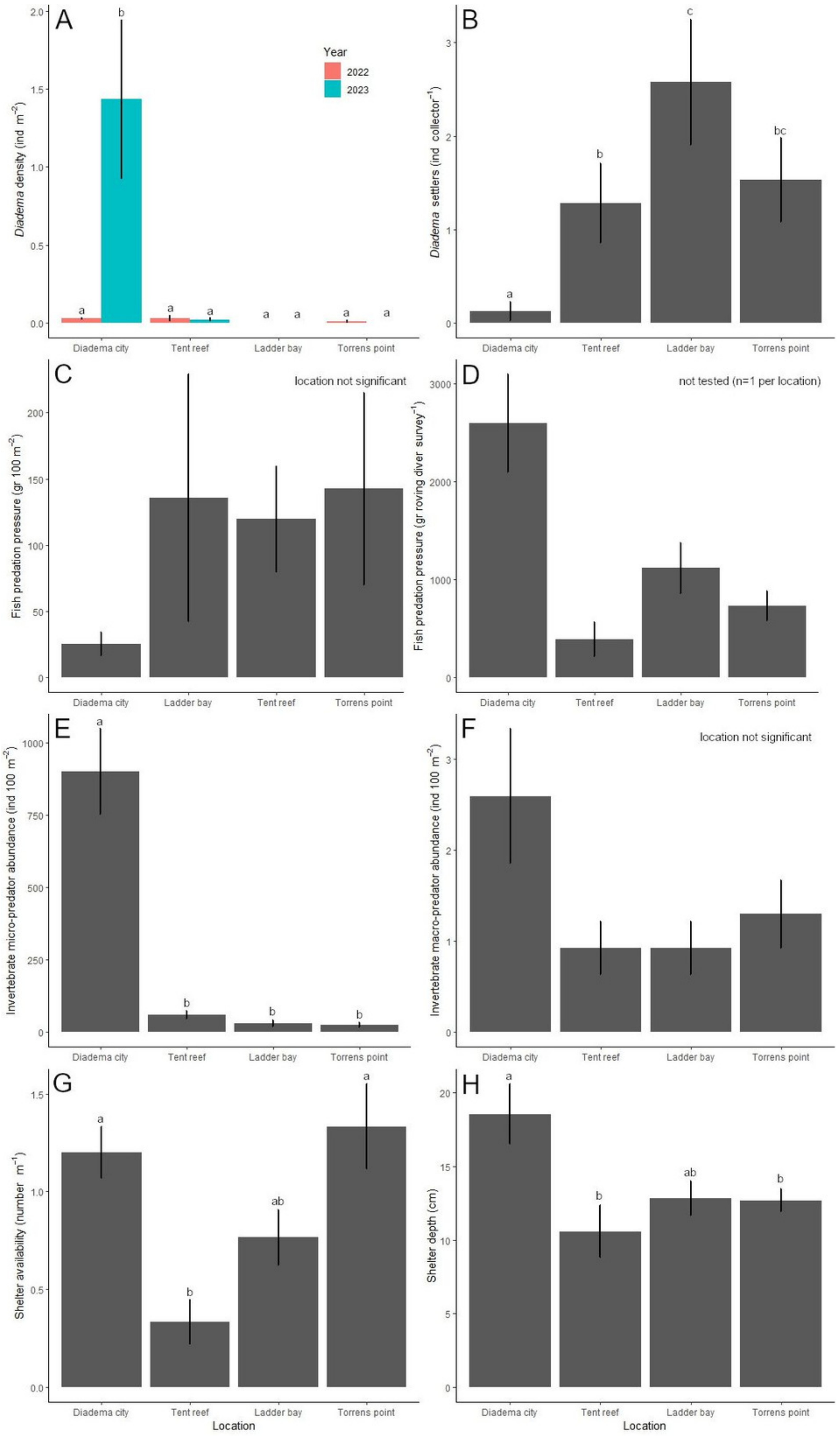


Figure 4

Results per location

Per location, the average (A) the *Diadema* density (ind m -2) in 2022 (right after the die-off) and 2023, (B) *Diadema* settlement (ind collector -1), (C) fish predation pressure (gr 100m -2), (D) fish predation pressure (gr roving diver survey -1), (E) invertebrate micro-predator abundance (ind 100m -2), (F) invertebrate macro-predator abundance (ind 100m -2), (G) shelter availability (number m -1), and (H) shelter depth (cm). Locations which do not share a common lowercase letter differ significantly from each other.


A
Year
2022
2023
Diadema density (ind m^{-2})
2.0
1.5
1.0
0.5
0.0
b
a
a
a
a
a
a
a
Diadema city
Tent reef
Ladder bay
Torrens point
B
Diadema settlers (ind collector^{-1})
3
2
1
0
c
b
bc
a
Diadema city
Tent reef
Ladder bay
Torrens point
C
location not significant
Fish predation pressure (gr 100 m^{-2})
200
150
100
50
0
Diadema city
Ladder bay
Tent reef
Torrens point
D
not tested (n=1 per location)
Fish predation pressure (gr roving diver survey^{-1})
3000
2000
1000
0
Diadema city
Tent reef
Ladder bay
Torrens point
E
Invertebrate micro-predator abundance (ind 100 m^{-2})
1000
750
500
250
0
a
b
b
b
Diadema city
Tent reef
Ladder bay
Torrens point
F
location not significant
Invertebrate macro-predator abundance (ind 100 m^{-2})
3
2
1
0
Diadema city
Tent reef
Ladder bay
Torrens point
G
Shelter availability (number m^{-1})
1.5
1.0
0.5
0.0
a
b
ab
a
Diadema city
Tent reef
Ladder bay
Torrens point
Location
H
Shelter depth (cm)
20
15
10
5
0
a
b
ab
b
Diadema city
Tent reef
Ladder bay
Torrens point
Location