

# Resource allocation trade-offs in the sea urchin *Tripneustes gratilla* under relative storminess and wave exposure

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**ABSTRACT:** Increased temperature and storminess will render nearshore tropical organisms more vulnerable to climate change. The sea urchin *Tripneustes gratilla* lives in a wide range of habitats and can survive even in harsh conditions, but the mechanisms underlying its resilience are not well understood. We investigated the somatic and reproductive phenotypic traits of adults ( $66.78 \pm 0.22$  mm test diameter) from seagrass- and seaweed-dominated sites during 2 monsoon seasons in a location frequently disturbed by strong cyclones and northeasterly winds in the northeastern Philippines (NE Phil). These were compared with seagrass sites in a less exposed location in the northwest portion of the islands (NW Phil). Populations from NE Phil had significantly thicker and heavier body walls, but significantly smaller Aristotle's lanterns, guts and gonads regardless of season compared to those from NW Phil. Moreover, the body walls in individuals from the seaweed-dominated sites were thicker and heavier. Allocation of the body wall to body weight was 62.2 and 53.7% in the NE Phil seaweed and seagrass sites, respectively, both of which were significantly greater than those from the NW Phil seagrass sites (44.74%). In contrast to other studies, plasticity in the Aristotle's lantern was not related to food availability. The differences in the Aristotle's lantern, gonad and body wall weights indicate a trade-off in resource allocation for feeding (growth) and reproduction in favor of maintenance. Comparative analysis of multiple phenotypic traits of *T. gratilla* populations provides substantive empirical field evidence and insights into the resilience of tropical species to increased storminess and wave exposure.

**KEY WORDS:** Resource allocation · Trade-offs · Phenotypic plasticity · *Tripneustes gratilla* · Storminess · Monsoon winds · Seaweed-, Seagrass-habitats

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## 1. INTRODUCTION

Climate-driven changes in coastal environments have been predicted to include enhanced impacts of temperature- and wind-induced forces (e.g. waves, surges and tropical cyclones). These hydrodynamic forces have been shown to influence the distribution and survival of many intertidal and benthic organisms (e.g. Koehl 1984, 1999, Denny & Gaines 1990, Denny 1995, Bell & Gosline 1997, Blanchette et al. 2008). Tropical cyclones exemplify one of the greatest risks to the survival of these intertidal organisms,

by enhancing wave action that can quickly injure, dislodge or kill individuals. The northeastern Philippines, within the western north Pacific basin, is a region subject to the most intense tropical cyclones (Cinco et al. 2016, Takagi & Esteban 2016, NASA's Earth Observatory <https://earthobservatory.nasa.gov/images/7079/historic-tropical-cyclone-tracks>) and the highest average wind velocity worldwide (Hsiang & Narita 2012). Hence, the impact of frequent exposure to tropical cyclones and monsoon wind-induced wave actions on intertidal organisms can be more intense here compared to other areas worldwide.

Understanding the responses of subtidal invertebrates in this region is particularly important considering that knowledge about the effect of natural disturbance on shallow water benthic invertebrates in tropical marine ecosystems is currently lacking (Przeslawski et al. 2008), as is information regarding their vulnerability to climate change.

Wave exposure and water flow have been shown to affect the structural strength, dislodgement resistance, morphology, growth and survival of both sessile and mobile organisms. For example, based on model simulations, the dislodgement probability of the mussel *Mytilus californianus*, a dominant space competitor, was found to be directly related to the force due to wave velocity and height (Denny 1995). Further, byssal thread thickness and attachment strength were positively related to hydrodynamic forces (Bell & Gosline 1997). In the mussels *M. edulis* and *M. trossulus*, attachment strength increased during a season of stronger wave action (Hunt & Scheibling 2001, Carrington 2002 for *M. edulis*). For mobile organisms, the movement of sea urchins has also been shown to be inhibited by increased water flow in laboratory experiments (e.g. *Centrostephanus coronatus*, Lissner 1980; *Colobocentrotus atratus*, *Echinometra mathaei*, *Strongylocentrotus purpuratus*, Denny & Gaylord 1996; *S. nudus*, Kawamata 1998, 2010; *S. droebachiensis*, Siddon & Witman 2003, Lauzon-Guay & Scheibling 2007). Variations in hydrodynamic forces also influence movement and feeding behavior of finfishes (e.g. Fulton & Bellwood 2005, Finelli et al. 2009). However, unlike finfishes, many benthic invertebrates do not have the ability to move quickly, escape and avoid the stress associated with the impact of exposure to storminess and surge. Their ability to forage for food in turbulent, disturbed habitats is limited, and could consequently limit their reproduction (Denny & Gaines 1990). Based on the reduced movement of sea urchins exposed to high water flow that has been recorded in various laboratory studies, it has been inferred that high wave action would inhibit foraging behavior. This could translate to lower food intake, processing capacity and gonad production. Wave disturbance was also shown to decrease the gonad and reproductive condition in the temperate sea urchin *Evechinus chloroticus* held in experimental sea cages (James 2006), and decrease the gonad index of *Paracentrotus lividus* sampled from the field (Gianguzza et al. 2013). Empirical data from natural populations in habitats frequently exposed to tropical cyclones and strong waves associated with high wind velocities (e.g. monsoon winds) is very limited. Such informa-

tion would provide a better understanding of the adaptive mechanisms underlying the life history characteristics of resilient species. Furthermore, analysis of the resource allocation pattern of a species is important in understanding its life history traits and strategies, since the 3 basic component processes (growth, maintenance and reproduction) compete for the limited energy resources obtained by an organism (Gadgil & Bossert 1970). Maintenance allocation, according to Ebert (1982), includes all morphological, physiological and behavioral characteristics that increase the probability of survival.

The echinoid *Tripneustes gratilla* is common in the tropical Indo-west Pacific region. It is known to be a ruderal strategist, characterized by fast growth rates, high respiration rates, high feeding capacity and reproductive effort and short life span (Shimabukuro 1991, Lawrence & Bazhin 1998, Lawrence & Agatsuma 2001, 2007, 2013). This species is usually found in shallow seagrass and seaweed habitats that have high potential primary productivity and food availability, and that are exposed to various natural and human-induced disturbances (Lawrence 2013). Populations of *T. gratilla* are found in a wide range of protected as well as high-energy habitats (Ebert 1982, Ogden et al. 1989, Lawrence & Agatsuma 2007, 2013). We have found them in rocky shores with *Sargassum* sp. frequently swept by extremely strong wave action. They can also survive in very low food and starved conditions in sea cages for relatively long periods (i.e. >180 d; Bangi 2001). Although the biology and ecology of the genus *Tripneustes* has been extensively studied (reviewed in Lawrence & Agatsuma 2001, 2007, 2013), the adaptations of populations in habitats which are frequently exposed to tropical cyclones, strong monsoon winds and associated wave action have not been investigated.

*T. gratilla* is a commercially important fishery species that is harvested for its gonads (Talaue-McManus & Kesner 1995, Juinio-Meñez et al. 1998, 2008a, Andrew et al. 2002). It is also ecologically important in recycling nutrients and in the reduction of algal overgrowth in seagrass and reef ecosystems (e.g. Koike et al. 1987, Mukai et al. 1989, Klumpp et al. 1993, Dy & Yap 2000, Valentine & Duffy 2006, Stimson et al. 2007). Thus, it is an excellent key model organism with which to gain insights on climate change adaptations and potential changes in its ecosystems and fisheries.

This study determined whether there are significant variations in somatic (body wall, Aristotle's lantern, gut) and reproductive (gonad) phenotypic traits of *T. gratilla* populations. These traits were compared among populations in seagrass- and seaweed-domi-

nated sites with different exposures to storminess and northeasterly monsoon winds (i.e. frequency of tropical cyclones or storminess, associated surge and wave action) during 2 monsoon seasons. In addition, potential trade-offs in relative allocation between somatic and reproductive traits, as affected by relative exposure to storminess, were investigated.

## 2. MATERIALS AND METHODS

### 2.1. Study locations and sites

The study sites that were frequently exposed to tropical cyclones and northeasterly winds were located within the seagrass- and seaweed-dominated beds of Palau Island, San Vicente, in the northeastern Philippines (hereafter, NE Phil) ( $18^{\circ}33'24.22''$  N,  $122^{\circ}08'25.40''$  E; Fig. 1A). NE Phil is generally wet throughout the year, with a distinct cold, windy season especially during the NE monsoon season (November to March hereafter NEMon), and a hot season during the easterly monsoon (April to June hereafter EasterlyMon). Wind-induced waves and surge peak from January to February (Cinco et al. 2016, Philippine Atmospheric Geophysical and Astronomical Service Administration, [www1.pagasa.dost.gov.ph](http://www1.pagasa.dost.gov.ph)) in the NEMon season, with very high associated wind stress (C. Repollo unpubl. data). NE Phil is located within the frequent path of the most powerful tropical cyclones worldwide based on track data from 1951 to 2013 and 1851 to 2006 (Takagi & Esteban 2016, NASA's Earth Observatory <https://earthobservatory.nasa.gov/images/7079/historic-tropical-cyclone-tracks>).

This same region was also found to have the highest average wind velocity, recorded at over  $30 \text{ m s}^{-1}$  (Hsiang & Narita 2012). Incorporating the frequency and wind velocity of powerful tropical cyclones into the Wave Exposure Model's (WEMo) relative exposure index (REI), NE Phil has a much higher REI compared to other sites in the archipelago (Villanoy et al. 2013).

In contrast, Santiago Island, Bolinao, which is located in the northwestern Philippines (hereafter, NW Phil) ( $16^{\circ}24'00.82''$  N,  $119^{\circ}55'59.81''$  E; Fig. 1B) is far less exposed to tropical cyclones and the impact of NEMon winds. There are 2 pronounced seasons in this region: the dry season from November to April, and a wet season during the rest of the year.

We used 'relative exposure to storminess and monsoon winds' ('Re') to infer the effect of wave action. A total of 8 study sites from the 2 locations were categorized with respect to Re as follows: (1) frequently exposed NE Phil, seaweed-dominated (SW-S1, SW-S2 and SW-S3; Fig. 1A); (2) frequently exposed NE Phil, seagrass-dominated (SG-S1, SG-S2 and SG-S3; Fig. 1A); and (3) less exposed NW Phil, seagrass-dominated (SG-S4 and SG-S5; Fig. 1B). In NW Phil, sufficient numbers of *Tripneustes gratilla* adults were found in only 2 seagrass-dominated sites. The collection of samples and determination of environmental conditions were conducted on 22–23 January 2016 in NW Phil and 28–29 January 2016 in NE Phil to represent the NEMon season, and on 22–23 April 2016 in the NE Phil and 28–29 April 2016 in the NW Phil for the EasterlyMon season. This was based on the peak of the NEMon winds (also the usual end of typhoon season) and the extremes of the cold and hot seasons.

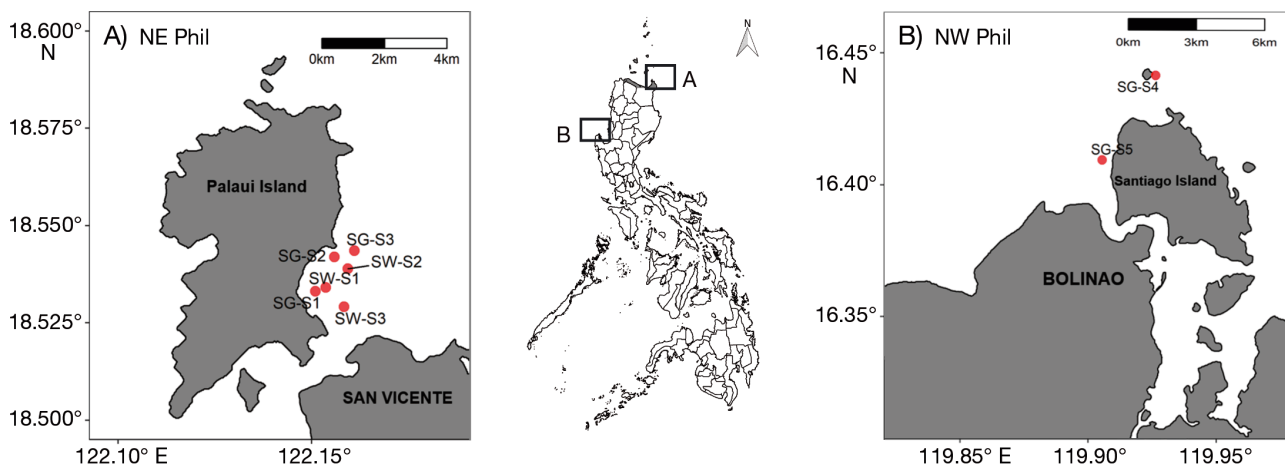


Fig. 1. Study locations in (A) northeastern (NE Phil; San Vicente) and (B) northwestern (NW Phil; Bolinao) Philippines; middle map shows their relative locations (black rectangles). Red dots: sampling locations at seagrass-dominated sites in NE Phil (SG-S1, -S2 and -S3) and NW Phil (SG-S4 and -S5); seaweed-dominated sites (SW-S1, -S2 and -S3) are only in NE Phil

## 2.2. Environmental physical parameters

Thermologgers (HOBO Water Temp Prodata logger; Onset Computer Corporation) were installed in each location to record sea surface temperature (SST) over a year. During each sampling period at each site, mean depth was determined from measurements during high and low tides within the sampling period, using a depth gauge and an expandable ruler >1 m long for relatively shallow sites. Relative water movement was also determined at each site using the 'diffusion factor (DF) technique' or clod card method (Doty 1971, Jokiel 1978), with dissolution rate of plaster of Paris being directly proportional to water current velocity. A total of 3 replicate cards were deployed over two 24 h periods at each site during each sampling period. Clod cards were calibrated by immersing 3 cards in 125 l of seawater (35 ppt salinity), replicated 3 times. The diffusion rate (DR) was determined as the plaster weight loss (in g) over a 24 h period, while DF was calculated as the ratio of weight loss in the experimental plaster block (field) to the weight loss of a control setup in calm water during calibration (Doty 1971).

## 2.3. Quantitative characterization of macrophyte habitats and food abundance

The main criteria for determining the study sites within each location were the presence and abundance of both the adult *T. gratilla* (of similar sizes), and their known macrophytic food/habitat: seagrass or seaweeds. The average above-ground biomass (fresh or dry weight;  $\text{g m}^{-2}$ ) of seagrass and seaweeds and the density of seagrass shoots (shoot number  $\text{m}^{-2}$ ) were determined at each site using a standard quadrat method (English et al. 1997, Kendrick & Lavery 2001). Up to 15 quadrats were laid within the three  $100 \times 3\text{--}4$  m belt transects used in each site every sampling season. Representative samples were collected within the total quadrats assessed per site, and brought to the laboratory for sorting and identification to species level (Fortes 1986, 2014 for seagrasses; Trono 1997, 2004 for seaweeds). Samples were rinsed with fresh water, and epiphytes were gently scraped off prior to drying at  $60^\circ\text{C}$  to constant weight. To determine dry biomass, cleaned seagrass and seaweed samples were weighed fresh using an electronic weighing scale prior to drying at  $60^\circ\text{C}$  to a constant weight.

## 2.4. *T. gratilla* somatic and reproductive parameters

Average density at each site (ind.  $100 \text{ m}^{-2}$ ) was determined from the same transects used in the characterization of macrophyte habitats during the same sampling seasons. All *T. gratilla* found within each belt transect were counted and measured. A Vernier caliper was used to measure the test diameter (TD) and test height of each individual to the nearest 0.5 mm. The density per site was relatively low (2.1 to  $9.1 \text{ ind. } 100 \text{ m}^{-2}$ ).

Since an urchin's ability to obtain food is affected by its size (Lawrence 1990), and the total body weight as well as that of the internal components (e.g. gonads) have been found to be directly related to TD (e.g. Tuason & Gomez 1979, Juinio-Meñez et al. 2008b), 30 to 35 adult *T. gratilla* ranging in size from 62 to 70 mm TD were collected from each site and sampling season for laboratory analyses. Sampling for the phenotypic variables was limited to only 10 females of similar test size per site, considering the difference in the sex ratio, gonad size and quality between males and females.

Each urchin was measured and blot-dried with a dry cloth and tissue paper before weighing (to the nearest 0.1 g) to determine total fresh body weight (BW). The gut of each sample was removed from the test, and contents were carefully removed and set aside, weighed (to the nearest 0.1 g) prior to preservation in buffered 5% formalin and seawater for further analysis. Since the BW of the sea urchins varied between sites ( $F_{2,157} = 4.979$ ,  $p = 0.008$ ), the amount of food in the gut was evaluated using the repletion index (RI), which is an indicator of consumption rate. RI was calculated and modified from Väitölä et al. (2003) as:  $\text{RI} = \text{FW}_{\text{dt}} / \text{BW} \times 100$ , where  $\text{FW}_{\text{dt}}$  is the fresh weight of the digestive tract contents (in g), and BW as the total fresh weight of the body (in g).

Each sample was further dissected to determine the weight of the main body components: body wall (test and spines), Aristotle's lantern (teeth and jaw pyramid), gonad, and gut or digestive tract. The gonadosomatic index (GSI), which is the ratio of gonad fresh weight ( $\text{FW}_{\text{g}}$ ) to total body fresh weight (BW) (in g), multiplied by 100 was calculated as:  $\text{GSI} = \text{FW}_{\text{g}} / \text{BW} \times 100$ . The body wall, Aristotle's lantern and gut indices (BI, ALI and GI, respectively) were similarly determined as the ratio of the respective weights and BW of the sea urchin, multiplied by 100.

The thickness of the test (TTh, internal to external width) was measured using a digital caliper to the nearest 0.01 mm (modified from Byrne et al. 2014).

Five or more pieces of dried tests containing the ambitus were collected from each individual sample ( $n = 10 \text{ ind. site}^{-1} \text{ season}^{-1}$ ).

## 2.5. Data and statistical analyses

Data were checked for normal distribution and homogeneity of variance using Kolmogorov-Smirnov and Levene's tests. Appropriate data transformations and normalization were made prior to univariate and multivariate analyses of variance. However, some variables were heterogenous even after transformation. A permutational analysis of variance (PERMANOVA), which is considered a nonparametric to semi-parametric test (Anderson 2001a,b, 2005) was used to test differences in the multiple response variables (somatic and reproductive phenotypic traits) in *T. gratilla* with respect to Re. In the analyses, Re was a fixed factor, with 3 levels (frequently exposed NE Phil, seaweed-dominated sites; frequently exposed NE Phil, seagrass-dominated sites; and less exposed NW Phil, seagrass sites). 'Site' (Si) was a random factor nested in Re, 3 levels (Sites 1, 2 and 3); and 'Season' (Se) as random factor, 2 levels (NEMon and EasterlyMon seasons) nested in Re and Si. Data were computed on a resemblance matrix based on a normalized Euclidean distance dissimilarity index on square root-transformed and untransformed data. Similar tests were done to determine significant differences in the environmental variables (i.e. biological: seaweeds and seagrass dry above-ground biomass, *T. gratilla* density; and physical: DR, depth) with respect to Re. Differences in mean SST and seagrass shoot density between the frequently exposed and less exposed locations were tested using Student's *t*-tests. All *p*-values in PERMANOVA were obtained using random 4999 permutations of residuals under a reduced model (Anderson 2001b, Anderson & ter Braak 2003). When possible permutations were not enough to get an appropriate exchangeability of units, a *p*-value was calculated using 4999 Monte Carlo draws from the theoretical asymptotic permutation distribution (Anderson 2001b, 2005). *A posteriori* pair-wise tests were done where appropriate. The univariate analyses used only 1 variable per analysis based on Euclidean distance index on square-root transformed or untransformed data. The potential effects of covariables, such as total BW and TD on the body component weights or test thickness were analyzed using permutational multivariate analysis of covariance (PERMANCOVA), with a similar process of computations as in PERMANOVA.

To compute and visualize the correlational structure of the multiple phenotypic variables, as well as their relationships with the environmental variables, vectors were overlaid representing the Pearson's correlation of each variable with the ordination axes (principal coordinates analysis [PCO], and canonical analysis of principal coordinates [CAP]; Anderson & Willis 2003). In addition to CAP, scatter plots, regression analyses and a distance-based linear model (distLM; McArdle & Anderson 2001) were used to determine the type of relationships of the phenotypic traits, and how these traits related to (environmental) predictor variables. Prior to analysis of environmental variables and their relationships with the phenotypic response variables, data were analyzed by principal component analysis (PCA). Calculations were based on normalized Euclidean distance on log-transformed data. All multivariate and univariate analyses were done using the software Primer 6 (based on Clarke & Warwick 2001) with add-on package PRIMER-e and PERMANOVA+ software (Clarke & Gorley 2006).

## 3. RESULTS

### 3.1. Physical environmental parameters

Mean SSTs differed significantly between the frequently exposed sites in NE Phil and the less exposed NW Phil sites (2-tailed  $t_{760} = -25.302$ ,  $p = 0.001$ ). Mean SSTs in NE Phil were consistently lower ( $24.84 \pm 0.06^\circ\text{C}$ , NEMon;  $28.04 \pm 0.12^\circ\text{C}$ , EasterlyMon) compared to NW Phil ( $27.15 \pm 0.11^\circ\text{C}$ , NEMon;  $30.92 \pm 0.07^\circ\text{C}$ , EasterlyMon) (Fig. S1 in the Supplement at [www.int-res.com/articles/suppl/m608p165\\_supp.pdf](http://www.int-res.com/articles/suppl/m608p165_supp.pdf)).

The overall mean DR as a measure of relative water movement also differed significantly across relative exposures in NE and NW Phil, and between sampling seasons. No significant difference was observed across sites ( $p > 0.05$ ) in NE and NW Phil. In addition to being frequently exposed to storms, the sites in NE Phil were also frequently exposed to stronger water turbulence (mean DR =  $30.11 \pm 0.43$  to  $36.45 \pm 0.72 \text{ g d}^{-1}$ ) within the 2 sampling seasons, compared to the sites in NW Phil ( $11.80 \pm 0.29$  to  $18.74 \pm 0.25 \text{ g d}^{-1}$ ). Within NE Phil, the seaweed-dominated sites were more exposed to significantly stronger turbulence (overall mean DR =  $39.35 \pm 0.48 \text{ g d}^{-1}$  and  $27.92 \pm 0.60 \text{ g d}^{-1}$  during NEMon and EasterlyMon, respectively) compared to the seagrass-dominated sites (mean DR =  $27.28 \pm 0.41 \text{ g d}^{-1}$ , NEMon;  $16.83 \pm 0.13 \text{ g d}^{-1}$ , EasterlyMon). Significantly higher mean DR values

were observed during NEMon compared to EasterlyMon, both in NE and NW Phil. The seaweed-dominated sites with the strongest water turbulence also had the lowest mean depth. The greater depth of the seagrass sites within the frequently exposed NE Phil region likely buffered the effect of strong water turbulence, in particular during EasterlyMon (Fig. S2 in the Supplement). As with DR, mean depth differed significantly across locations in NE and NW Phil, and seasons, but not across sites.

### 3.2. Macrophyte habitats and food resources

The above-ground dry (Fig. 2A) and fresh biomass varied significantly across relative exposures in NE and NW Phil and between seasons, but not across sites. Within the frequently exposed NE Phil, the mean dry and fresh biomasses of seaweed were significantly higher by 4 to 5 orders of magnitude compared to the seagrass. Comparing the seagrass-dominated sites, the mean fresh and dry biomasses were significantly higher in NW Phil sites, in particular during EasterlyMon, compared to those in NE Phil. On the other hand, overall mean shoot density (Fig. 2B) was significantly higher in the NE Phil sites (by 2 orders of magnitude) compared to those in the NW Phil sites (2-tailed  $t_{11} = 2.450$ ,  $p = 0.032$ ), particularly during the NEMon (2-tailed  $t_5 = 3.449$ ,  $p = 0.018$ ).

*Sargassum* spp. comprised the bulk of the biomass in the seaweed-dominated sites (85.01 to 97.18% during NEMon and 62.44 to 86.14% during EasterlyMon). Other relatively abundant species included *Gracilaria edulis* during NEMon and *Laurencia flex-*

*ilis* and *Mastophora rosea* during EasterlyMon. In the seagrass-dominated sites, the bulk of the biomass was comprised of *Thalassia hemprichii* (90.64 to 94.25% during NEMon and 59.6 to 73.06% during EasterlyMon in NW Phil sites, but only 37.54 to 51.03% in NE Phil).

### 3.3. *Tripneustes gratilla* density and gut content weight

The mean density of *Tripneustes gratilla* was relatively low: 2.1 ind. 100 m<sup>-2</sup> during a southwest monsoon season (SWMon) in NW Phil, but up to 9.1 ind. 100 m<sup>-2</sup> during summer in NE Phil (Fig. 3A). There was no significant variation across relative exposures and sites but there was variation with sampling seasons. Notably, recruits (<30 mm TD) were found in most of the sampling sites and seasons. Recruits comprised up to 28.6% of the density in NE and NW Phil sites. Just like the adults, these individuals were found covered with fronds of *Sargassum* spp. during NEMon, with *M. rosea* during summer, and with *Padina minor* during late EasterlyMon/summer to SWMon.

The gut content fullness of all urchins was about 60% on average (but up to almost 100% full). The relative consumption rates (based on RI) of *T. gratilla* adults are shown in Fig. 3B. RIs were significantly higher in the samples from the less exposed NW Phil sites ( $23 \pm 1.25$ , but up to 33) compared to those from the frequently exposed NE Phil ( $19.27 \pm 0.64$  and  $19.51 \pm 0.79$ ), particularly during the NE monsoon season when the difference in water turbulence was highest.

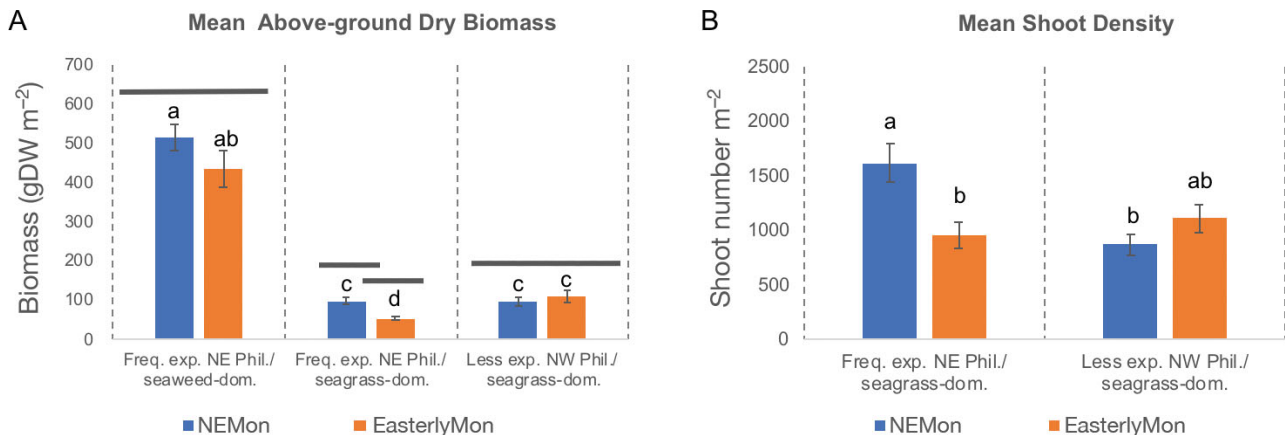


Fig. 2. Overall mean ( $\pm$ SE) (A) above-ground seaweed/seagrass dry biomass and (B) seagrass shoot density measured with respect to relative exposure to storminess and surge (NEMon: NE monsoon season; EasterlyMon: easterly/summer monsoon season) in the northeastern (NE Phil) and northwestern (NW Phil) study sites in the Philippines. Means with the same bar levels (across relative exposure) or the same letters (between seasons) are not significantly different from each other (PERMANOVA, pairwise post hoc tests, paired  $t$ -test for seagrass shoot density,  $p < 0.05$ , 2-tailed)

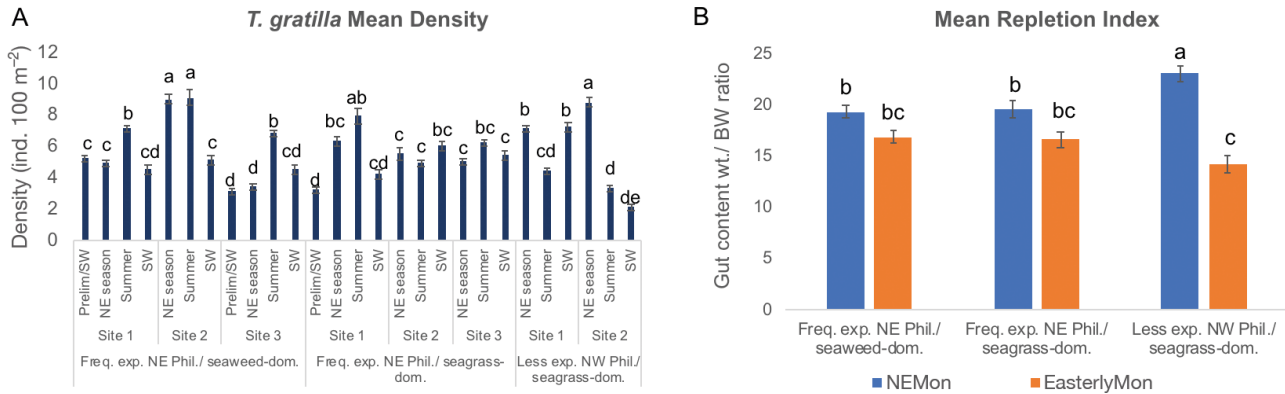


Fig. 3. Mean ( $\pm$  SE) (A) density and (B) consumption rates (based on repletion index) of similarly-sized adult *Tripneustes gratilla* with respect to their relative exposure to storminess and surge. Means with the same letters (between seasons) are not significantly different from each other (PERMANOVA, pairwise post hoc tests); see Fig. 1 for study site locations

### 3.4. Somatic and reproductive phenotypic traits

The different phenotypic traits of *T. gratilla* populations with respect to relative exposure to storminess are shown in Fig. 4A,B. Overall, both multivariate and univariate analyses on the somatic (TTh; BI, or weight; ALI, or weight; GI, or weight) and reproductive (GSI, or weight) traits showed that the effect of Re was highly significant and more than the effect of sampling season, as indicated by the pseudo-*F* values (Tables 1A–E & 2).

The TTh of urchins from the frequently exposed NE Phil were significantly thicker ( $1.85 \pm 0.03$  mm for seaweed-dominated sites;  $1.58 \pm 0.03$  mm for seagrass sites) compared to those in the seagrass sites at the less exposed NW Phil location ( $1.35 \pm 0.025$  mm; Fig. 4, Tables 1A & 2). Notably, within NE Phil, TTh of individuals from the seaweed-dominated sites were also significantly thicker, regardless of season. PERMANOVA still showed the significant effect of Re, after removing the effect of TD or total BW as a covariate.

The BI (Fig. 4A) from the frequently exposed NE Phil was also significantly higher ( $49.16 \pm 0.64$  for seaweed-dominated sites;  $45.13 \pm 0.58$  for seagrass sites) compared to those from the less exposed NW Phil ( $39.56 \pm 0.63$ ; Tables 1B & 2), regardless of season. Likewise, the BI and body wall weight (Fig. 4B) of samples from the seaweed-dominated sites were also significantly higher compared to those in the seagrass sites, regardless of season. PERMANOVA showed the significant effect of BW as a covariate of body wall weight (Table 1B). However, removal of the covariate still showed the significant effect of Re on the variations in body wall weight.

Conversely, for Aristotle's lantern and gut components, the mean indices (ALI, GI) and weights in the

frequently exposed NE Phil sites were significantly lower compared to the less exposed NW Phil (Fig. 4A,B Tables 1C,D & 2). However, the values from the seaweed-dominated sites were not significantly different from those from the seagrass sites, regardless of season. Notably, the overall mean ALI of individuals from the frequently exposed locations ( $7.80 \pm 0.53$  in the seaweed-dominated sites to  $7.92 \pm 0.39$  in the seagrass sites) were only a little over half of that in the less-exposed location ( $13.680 \pm 0.65$ ). On the other hand, overall mean GI of individuals from the frequently exposed locations were  $3.33 \pm 0.15$  in the seaweed-dominated sites to  $3.17 \pm 0.09$  in the seagrass sites, while  $4.69 \pm 0.16$  in the less exposed location. For the lantern weight, the effect of the covariate, BW, was not significant based on PERMANOVA. Gut weight, on the other hand, was affected by BW (Table 1D), however removal of BW as covariate showed the significance of Re on gut weight to be marginal. Overall, results further support the significant effect of Re on the somatic components, considering that the individuals were of the same sizes (in terms of TD) across locations and sites ( $F_{2,157} = 1.091$ ,  $p = 0.339$ ).

Similar to ALI and GI, the mean GSI of *T. gratilla* in the frequently exposed NE Phil were significantly lower ( $13.03 \pm 0.46$  from the seaweed-dominated sites;  $12.55 \pm 0.39$  from seagrass sites) compared to those from the less exposed sites ( $19.76 \pm 0.53$ ), regardless of season (Fig. 4A,B, Tables 1E & 2). Within the frequently exposed location, as with ALI and GI, the GSI of samples from the seaweed-dominated sites were not significantly different from those from the seagrass sites. Gonad weight was also affected by BW (Table 1E), however, removal of the BW covariate still showed the significant effect of Re on gonad weight.

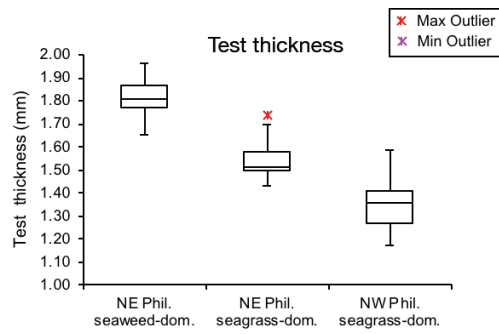
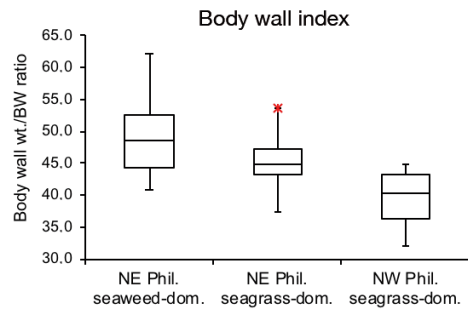
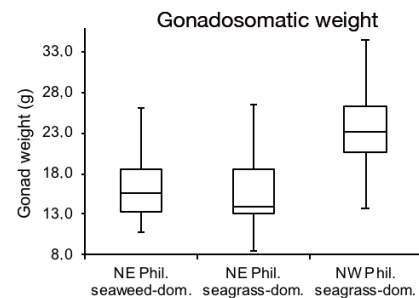
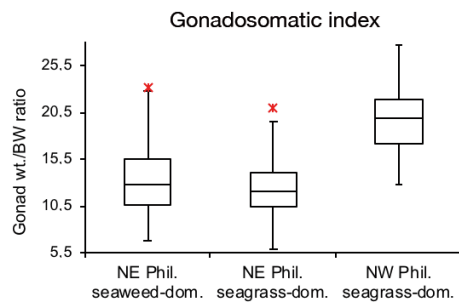
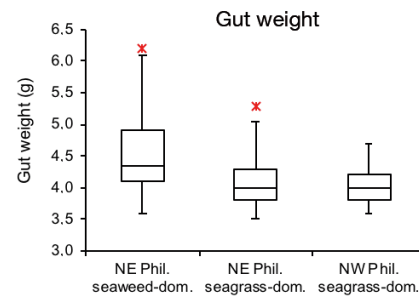
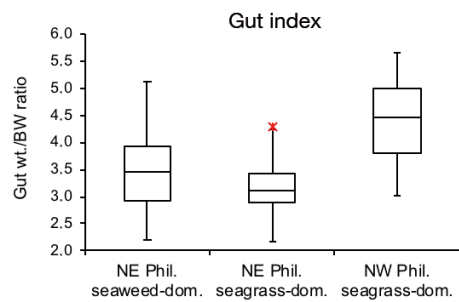
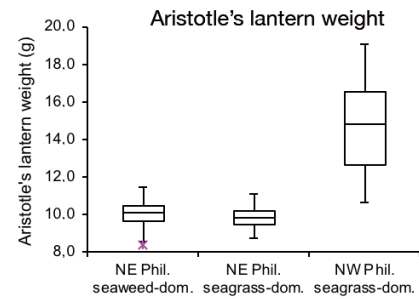
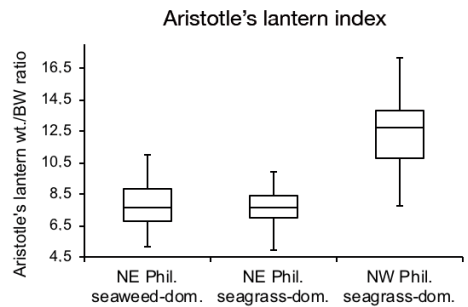
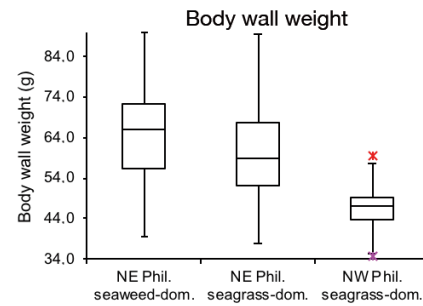


Fig. 4. Boxplots of test thickness and (A) % weight or ratio to total body weight, and (B) mean weight, for body wall, Aristotle's lantern, gut and gonad in similarly sized adult *Tripneustes gratilla* sampled with respect to their relative exposure to storminess and surge. See Fig. 1 for study site locations

(A) % weight ratio to total body weight



(B) Mean weight





Based on the overall percentages of the weight of main body components relative to the total weight of these components, *T. gratilla* allocates most of its resources to its body wall (Fig. 5). The body wall in individuals from the frequently exposed NE Phil sites comprised 66.16% (seagrass sites) to 67.41% (seaweed sites) of their total body weight compared to only 52.86% in the less exposed NW Phil sites.

The second greatest allocation was to the gonads which comprised 17.50 to 18.38% in NE Phil sites, but as high as 26.27% in the NW Phil sites. Notably, the allocation in the less exposed NW Phil sites for gonad, gut and Aristotle's lantern was highest, while for body wall it was the lowest. The lower allocation for gonad, gut and Aristotle's lantern in the frequently exposed NE Phil sites was due to the lower mass of these com-

Table 1. PERMANOVA results testing for differences in *Tripneustes gratilla* test thickness (A), and weight, and weight to total body weight ratio for body wall (B), Aristotle's lantern (C), gut (D), and gonad (E), with respect to relative exposure to storminess and surge (Re), a fixed factor; site (Si), a random factor nested in Re; and season (Se), a random factor nested in Si and Re. Separate tables for the analysis of body weight (BW) and test diameter (TD) as covariables, are shown in (A). Separate tables for the analysis of BW as a covariable, and for actual body component weight after removing the covariable, are shown in (B–E). Euclidian distance index on untransformed data was used for the resemblance matrix in (A–E). Significant p-values are indicated in **bold**, obtained using 4999 permutations or through Monte Carlo (MC) where appropriate. Pair-wise tests, specifically for the main factor of interest, Re, were done as an *a posteriori* check for significant effect. Asterisk (\*) indicates that p(permutation) and p(MC) values are marginally significant with respect to Re for gut weight (D) after the removal of BW as a covariable

<b>(A) Test thickness</b>								
Source	Test thickness (without covariable TD or BW)							
	df	MS	Pseudo-F	p				
Re – Relative exposure to surge/storminess	2	16.49	59.997	<b>0.0006</b>				
Si (Re) – Site	5	0.29	1.809	0.2122				
Se (Si (Re)) – Season	8	0.16	6.275	<b>0.0002</b>				
Res	1084	0.03						
Total	1099							
Source	Test thickness (with TD as covariable)				Test thickness (with BW as covariable)			
	df	MS	Pseudo-F	p	MS	Pseudo-F	p	
TD or BW Test diameter or total body weight (as covariable)	1	0.0191	0.758	0.376	0.0197	0.078	0.7816	
Re – Relative exposure to surge/storminess	2	16.272	59.204	<b>0.0004</b>	15.95	58.612	<b>0.0002</b>	
Si (Re) – Site	5	0.289	1.796	0.2208	0.289	1.823	0.2138	
Se (Si (Re)) – Season	8	0.1596	6.350	<b>0.0002</b>	0.157	6.238	<b>0.0002</b>	
Res	1083	0.0251			0.025			
Total	1099							
<b>(B) Body wall</b>								
Source	Body wall index (without covariable BW)							
	df	MS	Pseudo-F	p				
Re – Relative exposure to surge/storminess	2	1106.30	46.638	<b>0.0006</b>				
Si (Re) – Site	5	23.72	0.177	0.9638				
Se (Si (Re)) – Season	8	133.69	13.262	<b>0.0002</b>				
Res	144	10.08						
Total	159							
Source	Body wall weight (with covariable BW)				Body wall weight (covariable BW removed)			
	df	MS	Pseudo-F	p	df	MS	Pseudo-F	p
BW – Total body weight (as covariable)	1	18556	507.92	<b>0.0002</b>				
Re – Relative exposure to surge/storminess	2	1670.30	34.448	<b>0.0014</b>	2	3809	22.242	<b>0.0046</b>
Si (Re) – Site	5	48.70	0.20	0.958	5	171.25	0.347	0.8688
Se (Si (Re)) – Season	8	249.62	19.09	<b>0.0002</b>	8	493.56	5.232	<b>0.0002</b>
Res	143	13.08			144	94.33		
Total	159				159			

Continued on next page

Table 1 (continued)

<b>(C) Aristotle's lantern</b>									
Aristotle's lantern index (without covariable BW)									
Source	df	MS	Pseudo- <i>F</i>	p					
Re – Relative exposure to surge/storminess	2	332.0	137.23	<b>0.0002</b>					
Si (Re) – Site	5	2.4	0.19	0.9612					
Se (Si (Re)) – Season	8	13.0	6.30	<b>0.0002</b>					
Res	144	2.1							
Total	159								
Aristotle's lantern weight									
(with covariable BW)					(covariable BW removed)				
Source	df	MS	Pseudo- <i>F</i>	p	df	MS	Pseudo- <i>F</i>	p	
BW – Total body weight (as covariable)	1	3.66	1.43	0.2532					
Re – Relative exposure to surge/storminess	2	334.09	62.78	<b>0.0006</b>	2	326.01	51.071	<b>0.0006</b>	
Si (Re) – Site	5	5.37	0.30	0.9054	5	6.38	0.34	0.8722	
Se (Si (Re)) – Season	8	17.95	31.27	<b>0.0002</b>	8	18.64	29.41	<b>0.0002</b>	
Res	143	0.57			144	0.63			
Total	159				159				
<b>(D) Gut</b>									
Gut index (without covariable BW)									
Source	df	MS	Pseudo- <i>F</i>	p					
Re – Relative exposure to surge/storminess	2	19.156	23.174	<b>0.0048</b>					
Si (Re) – Site	5	0.827	0.518	0.7474					
Se (Si (Re)) – Season	8	1.595	4.313	<b>0.0002</b>					
Res	144	0.370							
Total	159								
Gut weight									
(with covariable BW)					(covariable BW removed)				
Source	df	MS	Pseudo- <i>F</i>	p	df	MS	Pseudo- <i>F</i>	p(perm)	p(MC)
BW – Total body weight (as covariable)	1	5.18	19.39	<b>0.0002</b>					
Re – Relative exposure to surge/storminess	2	3.11	4.61	0.0732	2	3.845	5.80	<b>0.0544*</b>	<b>0.0524*</b>
Si (Re) – Site	5	0.68	0.43	0.8186	5	0.662	0.38	0.865	0.8534
Se (Si (Re)) – Season	8	1.60	23.16	<b>0.0002</b>	8	1.740	19.89	<b>0.0002</b>	<b>0.0002</b>
Res	143	0.07			144	0.088			
Total	159				159				
<b>(E) Gonad</b>									
Gonad index (without covariable BW)									
Source	df	MS	Pseudo- <i>F</i>	p					
Re – Relative exposure to surge/storminess	2	731.99	47.469	<b>0.0004</b>					
Si (Re) – Site	5	15.42	0.253	0.927					
Se (Si (Re)) – Season	8	60.85	7.569	<b>0.0002</b>					
Res	144	8.04							
Total	159								
Gonad weight									
(with covariable BW)					(covariable BW removed)				
Source	df	MS	Pseudo- <i>F</i>	p	df	MS	Pseudo- <i>F</i>	p	
BW – Total body weight (as covariable)	1	213.77	8.444	<b>0.008</b>					
Re – Relative exposure to surge/storminess	2	863.32	29.231	<b>0.0018</b>	2	674.56	14.167	<b>0.0102</b>	
Si (Re) – Site	5	29.61	0.198	0.9536	5	47.62	0.262	0.9218	
Se (Si (Re)) – Season	8	149.2	12.334	<b>0.0002</b>	8	182.03	13.316	<b>0.0002</b>	
Res	143	12.10			144	13.67			
Total	159				159				

Table 2. Multivariate PERMANOVA results testing for differences in *Tripneustes gratilla* multiple phenotypic traits (body components % weight and test thickness), and body component weights, after removing the effect of the covariable, BW (body weight), with respect to relative exposure to surge or storminess (Re), a fixed factor; Site (Si), a random factor nested in Re; and Season (Se), a random factor nested in Si and Re. Euclidean distance index on square-root transformed data was used for the resemblance matrix. Significant p-values are indicated in **bold**, obtained using 4999 permutations. Pair-wise tests for Re were done as an *a posteriori* check for significant effect

Source	Main body components % weight and test thickness				Main body component weights (after removing the covariable, BW)			
	df	MS	Pseudo- <i>F</i>	p	df	MS	Pseudo- <i>F</i>	p
Re – Relative exposure to surge/storminess	2	28.417	49.88	<b>0.0002</b>	2	4865.00	11.870	<b>0.0006</b>
Si (Re) – Site	5	0.569	0.25	0.9976	5	409.86	0.426	0.9134
Se (Si (Re)) – Season	8	2.310	6.85	<b>0.0002</b>	8	961.22	6.840	<b>0.0002</b>
Res	144	0.337			143	140.52		
Total	159				159			

ponents compared to those from the NW Phil sites, despite similarities in TD size of samples.

### 3.5. Effect of body weight

The TD did not differ significantly among sampled individuals ( $F_{2,157} = 1.091$ ,  $p = 0.339$ ); however, the mean total BW was significantly different ( $F_{2,157} = 4.979$ ,  $p = 0.008$ ). PERMANCOVA on the corresponding weights of the body components showed the significant effect of BW as a covariate (except for Aristotle's lantern), however, after removing BW from the analyses, the main factor, Re, for all component weights (Tables 1B–E & 2) remained significant.

Overall, the mean body component weights (body wall, Aristotle's lantern, gut, gonad), with or without the BW covariate, also showed the significant effect of Re. Body wall weights were significantly higher in the frequently exposed location (i.e.  $64.69 \pm 1.62$  g for seaweed sites;  $59.67 \pm 1.49$  g for seagrass sites) compared to those from the less exposed location ( $47.06 \pm 0.86$  g). Likewise, within the frequently exposed location, mean body wall weights of samples from the seaweed sites were significantly higher compared to those from the seagrass sites, regardless of season. The distLM and regression analysis showed that body wall weight explained most (71.35%) of the variations observed in the total BW. With greater allocation for body wall, the Aristotle's lantern and gonad components of individuals from the frequently exposed location had significantly lower mean weights compared to those from the less exposed sites, again regardless of season. Within the frequently exposed location, mean weights for these components from samples in the seaweed sites did not significantly differ from those from the seagrass sites.

### 3.6. Multivariate analysis of the relationships of phenotypic traits

The structure and significance of the relationships of phenotypic traits as an index and weight are shown in Fig. 6A,B, respectively, and in Table 2. CAP discriminant power is indicated by the significant ( $p = 0.0002$ ) and very high allocation success (95.62 to 96.25%) and canonical correlation coefficient ( $\delta^2 = 0.9012$  to 0.9013) for the first PCO ordination, explaining 48.6 to 66.1% of the variations in *T. gratilla* phenotypic traits (Tables S1a & S2a). The second PCO axis still showed a relatively high percentage (22.0 to 26.1%) of variations explained by the significant difference in TTh and BI or body wall weight of samples between the seaweed-dominated sites and the seagrass sites within the frequently exposed NE Phil.

% allocation of resources to main body components

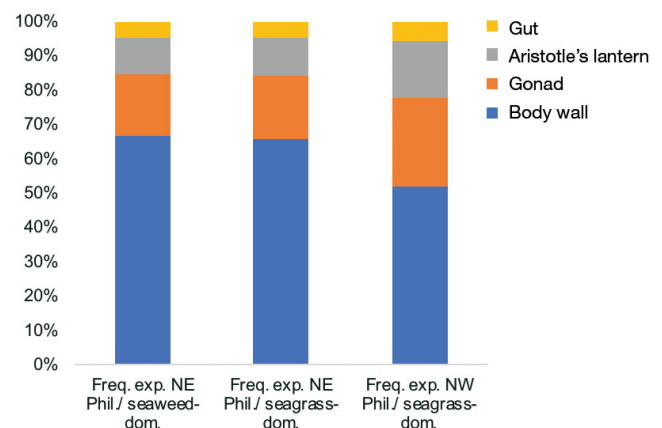
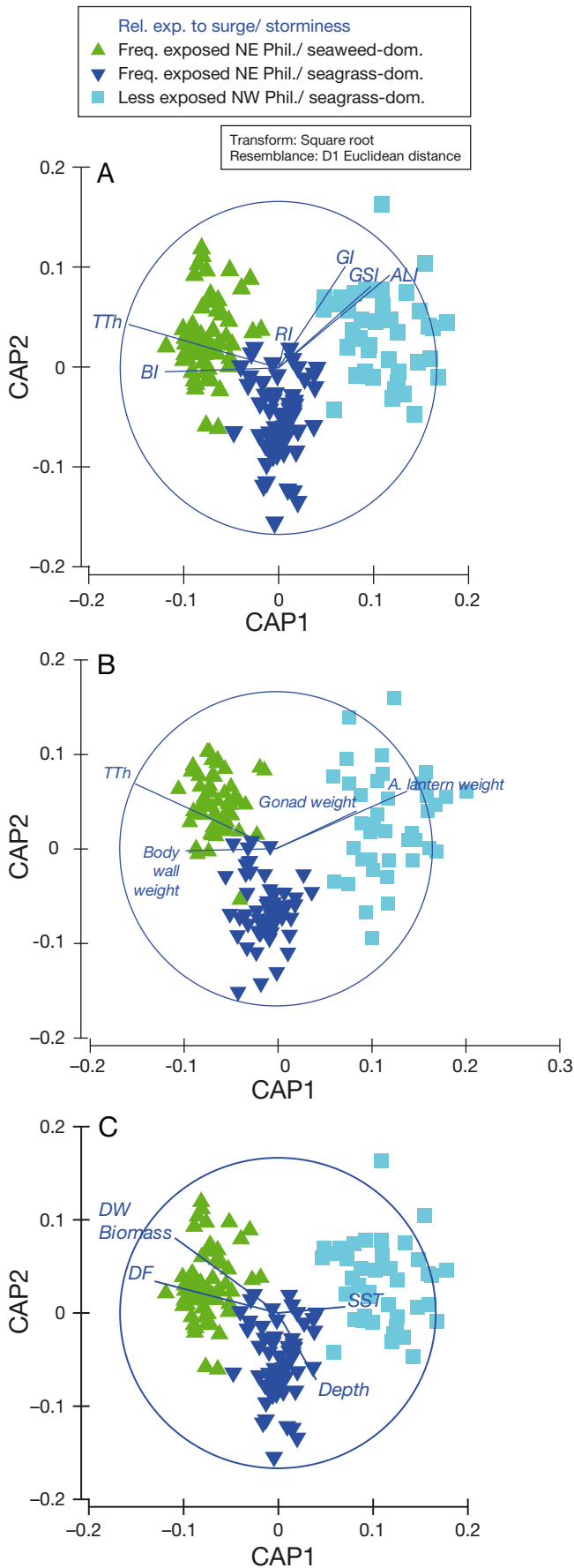


Fig. 5. Overall relative percentage allocation of resources to main body components in similarly-sized adult *Tripneustes gratilla* sampled with respect to their relative exposure to storminess and surge. See Fig. 1 for study site locations



Based on CAP results (Fig. 6A,B, Tables S1b & S2b), the phenotypic traits significantly ( $p = 0.0002$ ) associated with the less exposed NW Phil sites (with positive correlations) are higher GSI and gonad weight, higher ALI and Aristotle's lantern weight, higher GI and RI and lower TTh, BI and body wall weight. On the other hand, the phenotypic traits associated with the frequently exposed NE Phil sites (negative correlations) are higher TTh, higher BI and body wall weight and lower GSI, gonad weight, ALI, Aristotle's lantern weight, lower GI and RI. In the second CAP ordination, higher TTh and BI, body wall weight were significantly associated with the seaweed-dominated frequently exposed sites.

In addition to CAP, regression and distLM plus corresponding PERMANOVA also showed significant relationships of the phenotypic variables (Fig. S3b, Table S3). For example, body wall weight or BI and TTh were significantly positively related ( $p = 0.0002$ ). On the other hand, both these variables were significantly negatively related to Aristotle's lantern, gonad and gut ( $p = 0.0002$ , in terms of weight or as an index).

Overall, the first CAP ordination showed almost completely opposing variables, indicating trade-offs (Fig. 6A,B) between phenotypic traits as a response to the effect of Re. Although the direction of the vectors representing the correlations of the traits were not 180 degrees opposite to each other, as would be expected for a complete trade-off (in resource allocation) with respect to relative exposure to storminess,

Fig. 6. Constrained canonical analysis of principal coordinates (CAP) plots showing (A,B) trade-offs between *Tripneustes gratilla* phenotypic traits as a response to relative exposure to storminess and related factors, and (C) environmental variables with respect to relative exposure to storminess and related factors. The first canonical axis explained (A) 66.1%, (B) 48.6%, and (C) 66.1% of the variation, while the second axis explained (A) 22.2%, (B) 26.1%, and (C) 22.2% (as shown in Tables S1a & S2a in the Supplement). Vectors show correlations with original variables along the first 2 CAP axes, indicating the differences in (A,B) responses of different phenotypic traits with respect to 'relative exposure to storminess' and related factors, and in (C) the influence of environmental variables, mainly diffusion factor (DF), sea surface temperature (SST), mean site depth and seaweed/seagrass dry weight (DW) biomass, in explaining variations observed in the phenotypic traits. The phenotypic traits in (A) are TTh (test thickness), BI (body wall index), GSI (gonad index), ALI (Aristotle's lantern index), GI (gut index), and RI (repletion index); and in (B) are TTh (test thickness), body wall weight, Aristotle's lantern weight, and gonad weight. Details of CAP analyses are shown (A,B) in Tables S1a, S1b, S2a & S2b and (C) Tables S1a & S1b in the Supplement

the traits that explained the most of this observed pattern were related mainly to maintenance (TTh, BI, body wall weight) on one side (frequently exposed NE Phil), and reproduction (GSI, gonad weight) which is related to feeding and food processing (Aristotle's lantern weight, ALI, GI, RI) on the opposite side (less exposed NW Phil).

### 3.7. Relationship of phenotypic traits variation and environmental factors

The structure and significance of the relationships between phenotypic traits and environmental variables are shown in Fig. 6C and Table 3. Based on CAP results, the environmental variables (Fig. 6C,

Table S1b) with significant ( $p = 0.0002$ ) correlations with the less exposed location include higher SST and mean depth, and higher DF and dry weight biomass with the frequently exposed location. Higher DF was related to higher TTh, BI and body wall weight (maintenance), while higher SST was associated with reproduction and feeding (ALI, GSI, GI, Aristotle's lantern and gonad weight). In addition to CAP, the separation of groups and degree of relationships of phenotypic variables and environmental parameters with respect to the effect of Re are shown in the distLM analysis (Table 3). The effect of *T. gratilla* density was not significant ( $p = 0.6124$ ). Sequential tests showed that majority of the physical factors (DF, SST, depth), and partly DW biomass, contributed significantly (about 40%) to the variations observed in *T.*

Table 3. Distance-based linear model (distLM) results examining the relationships between *Tripneustes gratilla*'s phenotypic traits and environmental factors determined with respect to 'relative exposure to storminess and surge' at 3 sites and during 2 seasons for each study location. Proportion of variance in phenotypic traits explained by environmental variables in stepwise sequential tests following Akaike's information criterion corrected (AICc). SST: sea surface temperature; DF: diffusion factor; Depth: mean site depth; FW and DW biomass: seaweed/seagrass fresh and dry weight biomass, respectively. Prop.: proportion of variance explained by each variable; Cumul.: cumulative proportion of variance explained by multiple variables; res.df: residual degrees of freedom

Marginal tests on environmental variables							
Variable	SS (trace)	Pseudo- <i>F</i>	<i>p</i>	Prop.			
1. SST	1259.2	24.60	0.0002	0.1347			
2. DF	2208.4	48.88	0.0002	0.2362			
3. Depth	184.3	3.18	0.0444	0.0197			
4. FW biomass	924.6	17.35	0.0002	0.0989			
5. DW biomass	1117.3	21.45	0.0002	0.1195			
6. <i>T. gratilla</i> density	31.2	0.53	0.6124	0.0033			
Sequential tests result							
Variable	AIC <sub>c</sub>	SS (trace)	Pseudo- <i>F</i>	<i>p</i>	Prop.	Cumul.	res.df
+DF	611.76	2208.4	48.88	0.0002	0.2363	0.2363	158
+SST	591.85	916.6	23.13	0.0002	0.0981	0.3344	157
+Depth	581.85	453.1	12.25	0.0002	0.0485	0.3828	156
+DW biomass	579.37	164.0	4.54	0.0062	0.0175	0.4004	155
Best solution							
	AIC <sub>c</sub>	R <sup>2</sup>	RSS	No. Vars	Selections		
	579.37	0.4004	5604.4	4	1–3, 5		
Percentage of multivariate variation in biological variables (phenotypic traits) fitted to the variation in environmental variables by individual axes, based on distLM dbrDA analysis							
Axis	% explained variation out of fitted model		% explained variation out of total variation				
	Individual	Cumulative	Individual	Cumulative			
1	77.05	77.05	30.85	30.85			
2	22.06	99.11	8.83	39.68			
3	0.85	99.96	0.34	40.02			
4	0.04	100	0.02	40.04			

*gratilla* phenotypic traits, as shown in Fig. 6C. Notably, out of the total 40%, DF explained most of the variations in the first PCO or CAP ordination (23.63%), while SST only explained 9.81%.

## 4. DISCUSSION

### 4.1. Effects of relative storminess and wave exposure

This study has shown that sea urchins *Tripneustes gratilla* from high exposure sites in NE Phil have a significantly thicker, heavier body wall, but with significantly smaller gonad, Aristotle's lantern and gut compared to those from a similar habitat type in a less exposed location in NW Phil, regardless of season. Within the same exposed location, individuals had significantly thicker, heavier body walls and better gonad quality (H. G. P. Bangi & M. A. Juinio-Meñez, unpubl. data) in the seaweed-dominated sites, where wave exposure and water flow were significantly higher compared to those in the seagrass-dominated sites. Results of univariate and multivariate analyses clearly showed significant variability in the phenotypic traits of this species under relative exposure to storminess and northeasterly monsoon winds. Furthermore, after removing the effect of total BW as a covariate, the component weights and body wall thickness were still significantly different despite similarities in TD size. Taken together, this shows that the significant variations observed in phenotypic traits of the *T. gratilla* populations at the 8 study sites are significantly related to their relative wave exposure. To our knowledge, this comparative study is the first empirical test of the effect of relative exposure to storminess and associated hydrodynamism on the phenotypic traits of a tropical sea urchin. Previous studies have only documented the occurrence of this species in protected as well as in high-wave energy habitats (e.g. Ebert 1982, Ogden et al. 1989, Lawrence & Agatsuma 2007, 2013).

### 4.2. Implications of effects on body wall

The influence of storminess and relative wave exposure is most evident in the thicker and heavier body wall in the NE Phil populations. Body wall robustness (thickness and weight) indicates an adaptation to mechanical stress or the pressure of potential breakage or mortality in relation to chronic strong hydrodynamism. Based on the regression esti-

mates of survivorship probability of 17 Indo-West Pacific sea urchin species, Ebert (1982) showed that survivorship increased with increased relative size of the body wall and with increased protection from surf. According to Ebert's (1982) model, the same species that can survive over a wide range of surf conditions will tend to have the longest life span in the calmest water. Interestingly, it was noted that *T. gratilla* was atypical since it has a very short life span and very thin body wall, but was observed to recruit even in high-surf zones together with other species (e.g. *Colobocentrotus atratus*, *Heterocentrotus mamillatus*, *Echinometra mathaei*). These species have a massive body wall that comprises about 62% (on average) of their body weight, with slow growth and long life span (Ebert 1982, 1988). The body wall of *T. gratilla* in frequently exposed NE Phil sites comprised 40.76 to 62.2% of their total body weight compared to only 31.96 to 44.74% in the less exposed NW sites. Notably, the percentage of body wall weight in the NE sites was much greater than the average estimated for thin-body walled species (34.5%) and was similar to those of thick-body walled species (62%) in a comparative analysis of different echinoids conducted by Ebert (1982, 1988). Considering that *T. gratilla* belongs to a group of echinoids with the thinnest body wall (Ebert 1982), its survival in high surf areas and even in harsh conditions during tropical cyclones indicates that phenotypic plasticity of the body wall provides protection from potential shell breakage due to strong wave action, and hence a greater probability of survival. The development of a thicker body wall also provides additional protection from possible predation or test crushing (e.g. Guidetti & Mori 2005, Asnaghi et al. 2013, Byrne et al. 2014).

The clear differences in the phenotypic traits among populations suggest trade-offs in different environments to maximize fitness. *T. gratilla* allocates more of its energy resources for protection and maintenance (up to 62.2% of total body weight or about 67% of main body component weights) than to feeding and production of gonads in the frequently exposed NE sites. In addition, individuals from these sites had a smaller Aristotle's lantern, gonad, gut (weight or relative weight) and exhibited relatively lower RI. Feeding in the frequently exposed sites poses a greater threat to survival, hence a greater need for protection. This was evident in the observation of some individuals being found lodged in rocks or coral head crevices during the NEMon sampling period. This sheltering behavior is atypical for *T. gratilla*, which is commonly found in open areas on

seagrass and macrophyte beds. These individuals had deformed tests (H. G. P. Bangi & M. A. Juinio-Meñez unpubl. data), indicating that they may have stayed in the crevices for a considerable period. It is likely that such sheltering constrained feeding. This is the first field test and validation of the predictions of existing resource allocation models for greater allocation of resources to maintenance and defense in order to increase survival probability (Gadgil & Bossert 1970, Ebert 1982).

#### 4.3. Relationship of feeding structure with consumption, gut and gonad size

The greater energy allocated for protection (body wall) in the frequently exposed NE Phil sites appears to be at the expense of feeding capacity and reproduction. CAP and distLM analyses showed that the GI and GSI and weights were significantly related to the feeding structure ALI and weight. Moreover, those with lower gut, gonad and Aristotle's lantern (weights or relative weights) also had lower consumption rates, despite an abundance of food in the NE Phil sites. The smaller size of the feeding structure (in terms of weight) in individuals from the NE sites was positively correlated with lower consumption rates (RIs), and more importantly, with smaller gut and gonad indices. In manipulative laboratory experiments on *Diadema setosum* (Ebert 1980) and *E. mathaei* (Black et al. 1984), the food intake capacity of urchins with relatively smaller lanterns was lower compared to urchins with larger lanterns.

Contrary to studies on other species, which reported that a larger lantern is a response to food limitation (Ebert 1980, Black et al. 1982, Lewis et al. 1990, Levitan 1991, Edwards & Ebert 1991, Ebert 1996, McShane et al. 1996, Fernandez & Boudouresque 1997), there was no indication that the larger Aristotle's lantern of *T. gratilla* in the NW sites was due to food limitation. The mean above-ground biomass of seagrass at the NW sites was significantly higher than at the NE sites. The lantern size was smaller in both the NE seagrass- and seaweed-dominated sites where food was also abundant. Moreover, none of the urchins had empty guts nor extremely low gut content (H. G. P. Bangi & M. A. Juinio-Meñez unpubl. data), indicating that food was readily available at the study sites in both locations. Even individuals collected during NE monsoon or after tropical cyclones, when macrophyte cover was reduced, did not have empty guts; the gut contents of all individuals was over 60% (up to almost 100%) full.

The larger feeding structure in the less exposed NW sites was positively correlated with higher consumption rates (repletion indices), larger gut (food processing) and larger gonad size (reproduction). The gut has a functional role in transferring nutrients to the gonads during the reproductive period and as a storage organ that allows echinoids to withstand periods of nutritional starvation (Klinger et al. 1988, Bishop & Watts 1994). This may explain the positive relationship between the size of the gonad and the relative size of the gut of *T. gratilla* in this study. Notably, the average gonad size of individuals sampled from the less exposed NW sites was about 20 to 26% of the body weight. This is much higher than those reported by Tuason & Gomez (1979) and our previous study of only up to 15% (Juinio-Meñez et al. 2008b). In comparison, average gonad sizes were significantly lower in individuals from both the seaweed-dominated sites (13%) and the seagrass site (12%) in the NE region. Results of this study suggest that this may be a consequence of reduced feeding capacity (i.e. smaller Aristotle's lantern) despite an abundance of food.

#### 4.4. Resilience of populations

The persistence of the *T. gratilla* population is indicated by the regular presence of recruits (<30 mm TD) and adults, even in the NE sites that are frequently hit by tropical cyclones. The overall density of the species in this region is also comparable to, or did not significantly differ from those in the less exposed NW sites, although density varied between different seasons and the mean density in general was relatively low (maximum of 9.1 ind. 100 m<sup>-2</sup>). If plasticity of the body wall in the frequently exposed location provides an adaptive advantage by increasing the probability of survival (Ebert 1982) of the population to the pre- or reproductive stage, the probability of future reproduction could be expected to be enhanced and favor maintenance of the population.

The sea urchins were also observed hiding in crevices or under patches of coral heads or rocks covered with seaweed or drift algae — even during night time, when they are expected to be active (Klumpp et al. 1993). The much higher biomass and relative rugosity of the dominant *Sargassum* spp. in the seaweed-dominated sites may offer more protection from strong wave action compared to the seagrass beds. Seagrass beds usually do not have rugose or heterogeneous canopy where sea urchins can attach

and protect themselves in the case of high water energy during tropical cyclones or to escape predation (Lawrence 2013).

## 5. CONCLUSIONS

*Tripneustes gratilla* is a resilient species that is capable of persisting in environments which are frequently disturbed and exposed to wind-induced wave action and storminess. Under these conditions, they allocate more of their resources to maintenance and protection by having a thicker and heavier body wall—a strategy to enhance their survival, hence maximize their fitness. Seeking shelter and protection may outweigh the benefits of optimizing grazing, given high food abundance. The plasticity of the body wall enhances survival, at the expense of feeding and reproduction (i.e. smaller Aristotle's lantern, gut and gonads). In the seaweed-dominated sites, access to higher quality food appears to offset the effect of relative exposure to storminess on reproduction. Conversely, sea urchins in the seagrass-dominated sites in the less exposed location allocated more resources to larger feeding structures, which translated to larger gonads and gut area for energy storage and absorption for the gonads. The findings of this study provide substantive insights on direct and indirect mechanisms underlying the resilience and ruderal characteristics of this species. To our knowledge, this is the first report of phenotypic plasticity in the body wall and Aristotle's lantern in the genus *Tripneustes*, although it has been predicted for ruderal species to have moderate plasticity but is more common in competitive species (Lawrence 1990, Lawrence & Bazhin 1998). Its resistance to starvation and ability to withstand long periods of low food ration in cages (>180 d; Bangi 2001) indicates that this is a very stress-tolerant species. Based on population genetic demographic analyses, its vulnerability to extinction is low (Casilagan et al. 2013). The different relative resource allocations to body wall robustness (thickness and weight), reproductive phenotypic traits (gonad size and quality) and feeding traits (Aristotle's lantern, gut) enables *T. gratilla* to persist in varying environmental conditions. Thus, predicted future climate-related impacts of temperature- and wind-induced hydrodynamic forces will most likely not have major impacts on *T. gratilla* populations. However, other stressors that can possibly interact with storminess or associated wave action, such as ocean acidification and predation pressure, should also be investigated.

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## LITERATURE CITED

- Anderson MJ (2001a) A new method for non-parametric multivariate analysis of variance. *Austral Ecol* 26:32–46
- ✦ Anderson MJ (2001b) Permutation tests for univariate or multivariate analysis of variance and regression. *Can J Fish Aquat Sci* 58:626–639
- Anderson MJ (2005) PERMANOVA: a FORTRAN computer program for permutational multivariate analysis of variance. Department of Statistics, University of Auckland
- ✦ Anderson MJ, ter Braak CJF (2003) Permutation tests for multi-factorial analysis of variance. *J Stat Comput Simul* 73:85–113
- ✦ Anderson MJ, Willis TJ (2003) Canonical analysis of principal coordinates: a useful method of constrained ordination for ecology. *Ecology* 84:511–525
- Andrew NL, Agatsuma Y, Ballesteros E, Bazhin AG and others (2002) Status and management of the world sea urchin fisheries. *Oceanogr Mar Biol Annu Rev* 40:343–425
- ✦ Asnaghi V, Chiantore M, Mangialajo L, Gazeau F, Francour P, Alliouane S, Gattuso JP (2013) Cascading effects of ocean acidification in a rocky subtidal community. *PLOS ONE* 8:e61978
- Bangi HGP (2001) The effect of adult nutrition on somatic and gonadal growth, egg quality and larval development of the sea urchin *Tripneustes gratilla* Linnaeus 1758 (Echinodermata: Echinoidea). MSc thesis, University of the Philippines Diliman, Quezon City
- ✦ Bell EC, Gosline JM (1997) Strategies for life in flow: tenacity, morphometry, and probability of dislodgment of two *Mytilus* species. *Mar Ecol Prog Ser* 159:197–208
- ✦ Bishop CD, Watts SA (1994) Two-stage recovery of gametogenic activity following starvation in *Lytechinus variegatus* Lamarck (Echinodermata: Echinoidea). *J Exp Mar Biol Ecol* 177:27–36
- ✦ Black R, Johnson MS, Trendall JT (1982) Relative size of Aristotle's lantern in *Echinometra mathaei* occurring at different densities. *Mar Biol* 71:101–106
- ✦ Black R, Codd C, Hebbert D, Vink S, Burt J (1984) The functional significance of the relative size of Aristotle's lantern in the sea urchin *Echinometra mathaei* (de Blainville). *J Exp Mar Biol Ecol* 77:81–97
- Blanchette CA, O'Donnell MJ, Stewart HL (2008) Waves as an ecological process. In: Jørgensen SE, Fath BD (eds) *Encyclopedia of ecology*, Vol 5: Ecological processes.



- Elsevier, Oxford, p 3764–3770
- ✦ Byrne M, Smith AM, West S, Collard M, Dubois P, Grabalandy A, Dworjanyan SA (2014) Warming influences  $Mg^{2+}$  content, while warming and acidification influence calcification and test strength of a sea urchin. *Environ Sci Technol* 48:12620–12627
- ✦ Carrington E (2002) Seasonal variation in the attachment strength of blue mussels: causes and consequences. *Limnol Oceanogr* 47:1723–1733
- ✦ Casilagan ILN, Juinio-Meñez MA, Crandall ED (2013) Genetic diversity, population structure, and demographic history of exploited sea urchin populations (*Tripneustes gratilla*) in the Philippines. *J Exp Mar Biol Ecol* 449: 284–293
- ✦ Cinco TA, de Guzman RG, Ortiz AMD, Delfino RJP and others (2016) Observed trends and impacts of tropical cyclones in the Philippines. *Int J Climatol* 36:4638–4650
- Clarke KR, Gorley RN (2006) PRIMER v6: user manual/tutorial. PRIMER-e, Plymouth
- Clarke KR, Warwick RM (2001) Change in marine communities: an approach to statistical analysis and interpretation, 2<sup>nd</sup> edn. PRIMER-e, Plymouth Marine Laboratory, Plymouth
- ✦ Denny MW (1995) Predicting physical disturbance: mechanistic approaches to the study of survivorship on wave-swept shores. *Ecol Monogr* 65:371–418
- ✦ Denny MW, Gaines S (1990) On the prediction of maximal intertidal wave forces. *Limnol Oceanogr* 35:1–15
- ✦ Denny M, Gaylord B (1996) Why the urchin lost its spines: hydrodynamic forces and survivorship in three echinoids. *J Exp Biol* 199:717–729
- ✦ Doty SM (1971) Measurement of water movement in reference to benthic algal growth. *Bot Mar* 14:32–35
- ✦ Dy DT, Yap HT (2000) Ammonium and phosphate excretion in three common echinoderms from Philippine coral reefs. *J Exp Mar Biol Ecol* 251:227–238
- Ebert TA (1980) Relative growth of sea urchin jaws: an example of plastic resource allocation. *Bull Mar Sci* 30: 467–474
- ✦ Ebert TA (1982) Longevity, life history and relative body wall size in sea urchins. *Ecol Monogr* 52:353–394
- ✦ Ebert TA (1988) Allometry, design and constraint of body components and of shape in sea urchins. *J Nat Hist* 22: 1407–1425
- Ebert TA (1996) Adaptive aspects of phenotypic plasticity in Echinoderms. *Oceanol Acta* 19:347–355
- Edwards PB, Ebert TA (1991) Plastic responses to limited food availability and spine damage in the sea urchin *Strongylocentrotus purpuratus*. *J Exp Mar Biol Ecol* 145: 205–220
- English S, Wilkinson C, Baker V (1997) Seagrass communities. In: Survey manual for tropical marine resources. ASEAN-Australia Marine Science Project. Australian Institute of Marine Science, Townsville, p 241–252
- ✦ Fernandez C, Boudouresque CF (1997) Phenotypic plasticity of *Paracentrotus lividus* (Echinodermata: Echinoidea) in a lagoonal environment. *Mar Ecol Prog Ser* 152:145–154
- ✦ Finelli CM, Clarke RD, Robinson HE, Buskey EJ (2009) Waterflow controls distribution and feeding behavior of two co-occurring coral reef fishes: I. Field measurements. *Coral Reefs* 28:461–473
- Fortes MD (1986) Taxonomy and ecology of Philippine seagrasses. PhD dissertation, University of the Philippines Diliman, Quezon City
- Fortes MD (2014) A review: biodiversity, distribution and conservation of Philippine seagrasses. *Philipp J Sci* 142: 95–111
- ✦ Fulton CJ, Bellwood DR (2005) Wave-induced water motion and the functional implications for coral reef fish assemblages. *Limnol Oceanogr* 50:255–264
- ✦ Gadgil M, Bossert WH (1970) Life historical consequences of natural selection. *Am Nat* 104:1–24
- ✦ Gianguzza P, Bonaviri C, Prato E, Fanelli G and others (2013) Hydrodynamism and its influence on the reproductive condition of the edible sea urchin *Paracentrotus lividus*. *Mar Environ Res* 85:29–33
- ✦ Guidetti P, Mori M (2005) Morpho-functional defenses of Mediterranean sea urchins, *Paracentrotus lividus* and *Arbacia lixula* against fish predators. *Mar Biol* 147: 797–802
- Hsiang SM, Narita D (2012) Adaptation to cyclone risk: evidence from the global cross-section. *Clim Change Econ* 3:1250011
- ✦ Hunt HL, Scheibling RE (2001) Predicting wave dislodgment of mussels: variation in attachment strength with body size, habitat, and season. *Mar Ecol Prog Ser* 213: 157–164
- ✦ James PJ (2006) The effects of wave and feeding disturbance on roe enhancement of the sea urchin *Evechinus chloroticus* held in sea-cages. *Aquaculture* 252:361–371
- ✦ Jokiel PL (1978) Effects of water motion on reef corals. *J Exp Mar Biol Ecol* 35:87–97
- Juinio-Meñez MA, Macawaris N, Bangi HGP (1998) Community-based sea urchin (*Tripneustes gratilla*) grow-out culture as a resource management tool. In: Jamieson GS, Campbell A (eds) Proceedings of the North Pacific symposium on invertebrate stock assessment and management. *Can Spec Publ Fish Aquat Sci* 125:393–399
- ✦ Juinio-Meñez MA, Bangi HGP, Malay MCD, Pastor D (2008a) Enhancing the recovery of depleted *Tripneustes gratilla* sea urchin stocks through grow out culture and restocking. *Rev Fish Sci* 16:35–40
- Juinio-Meñez MA, Bangi HGP, Malay MCD (2008b) Effect of type of feed, stocking density and grow out site on gonad index, growth and survivorship of cultured sea urchin *Tripneustes gratilla*. *Philipp Agric Sci* 91:439–449
- ✦ Kawamata S (1998) Effect of wave-induced oscillatory flow on grazing by a subtidal sea urchin *Strongylocentrotus nudus* (A. Agassiz). *J Exp Mar Biol Ecol* 224:31–34
- Kawamata S (2010) Inhibitory effects of wave action on destructive grazing by sea urchins: a review. *Bull Fish Res Ag* 32:96–102
- Kendrick GA, Lavery PS (2001) Assessing biomass, assemblage structure and productivity of algal epiphytes on seagrasses. In: Short FT, Coles RG (eds) Global seagrass research methods. Elsevier, Amsterdam, p 199–222
- ✦ Klinger TS, Watts SA, Forcucci D (1988) Effect of short-term feeding and starvation on storage and synthetic capacities of gut tissues of *Lytechinus variegatus* (Lamarck) (Echinodermata: Echinoidea). *J Exp Mar Biol Ecol* 117: 187–195
- ✦ Klumpp DW, Salita-Espinosa JT, Fortes MD (1993) Feeding ecology and trophic role of sea urchins in a tropical seagrass community. *Aquat Bot* 45:205–229
- ✦ Koehl MAR (1984) How do benthic organisms withstand moving water? *Am Zool* 24:57–70
- ✦ Koehl MAR (1999) Ecological biomechanics of benthic organisms: life history, mechanical design, and temporal patterns of mechanical stress. *J Exp Biol* 202:3469–3476
- ✦ Koike I, Mukai H, Nojima S (1987) The role of the sea urchin

- Tripneustes gratilla* (Linnaeus) in decomposition and nutrient cycling in a tropical seagrass bed. *Ecol Res* 2: 19–29
- ✦ Lauzon-Guay JS, Scheibling RE (2007) Seasonal variation in movement, aggregation and destructive grazing of the green sea urchin (*Strongylocentrotus droebachiensis*) in relation to wave action and sea temperature. *Mar Biol* 151:2109–2118
- Lawrence JM (1990) The effect of stress and disturbance in echinoderms. *Zool Sci* 7:17–28
- Lawrence JM (2013) Sea urchin life history strategies. In: Lawrence JM (ed) *Sea urchins: biology and ecology*, 3<sup>rd</sup> edn. Elsevier, Amsterdam, p 15–24
- Lawrence JM, Agatsuma Y (2001) Ecology of *Tripneustes*. In: Lawrence JM (ed) *Edible sea urchins: biology and ecology*, 1<sup>st</sup> edn. Elsevier, Amsterdam, p 395–413
- Lawrence JM, Agatsuma Y (2007) Ecology of *Tripneustes*. In: Lawrence JM (ed) *Edible sea urchins: biology and ecology*, 2<sup>nd</sup> edn. Academic Press, Amsterdam, p 499–520
- Lawrence JM, Agatsuma Y (2013) *Tripneustes*. In: Lawrence JM (ed) *Sea urchins: biology and ecology*, 3<sup>rd</sup> edn. Elsevier, Amsterdam, p 491–508
- Lawrence JM, Bazhin A (1998) Life-history strategies and the potential of sea urchins for aquaculture. *J Shellfish Res* 17:1515–1522
- ✦ Levitan DR (1991) Skeletal changes in the test and jaws of the sea urchin *Diadema antillarum* in response to food limitation. *Mar Biol* 111:431–435
- ✦ Lewis CA, Ebert TA, Boren ME (1990) Allocation of <sup>45</sup>calcium to body components of starved and fed sea urchins (*Strongylocentrotus purpuratus*). *Mar Biol* 105:213–222
- ✦ Lissner AL (1980) Some effects of turbulence on the activity of the sea urchin *Centrostephanus coronatus* Verrill. *J Exp Mar Biol Ecol* 48:185–193
- ✦ McArdle BH, Anderson MJ (2001) Fitting multivariate models to community data: a comment on distance-based redundancy analysis. *Ecology* 82:290–297
- ✦ McShane PE, Gerring PK, Anderson OA, Stewart RA (1996) Population differences in the reproductive biology of *Evechinus chloroticus* (Echinoidea: Echinometridae). *N Z J Mar Freshw Res* 30:333–339
- ✦ Mukai H, Koike I, Nishihira M, Nojima S (1989) Oxygen consumption and ammonium excretion of mega-sized benthic invertebrates in a tropical seagrass bed. *J Exp Mar Biol Ecol* 134:101–115
- Ogden NB, Ogden JC, Abbott IA (1989) Distribution, abundance and food of sea urchins in a leeward Hawaiian reef. *Bull Mar Sci* 45:539–549
- ✦ Przeslawski R, Ah Yong S, Byrne M, Woerheide G, Hutchings PAT (2008) Beyond corals and fish: the effects of climate change on noncoral benthic invertebrates of tropical reefs. *Glob Change Biol* 14:2773–2795
- Shimabukuro S (1991) *Tripneustes gratilla* (sea urchin). In: Shokita S, Kakazu K, Tomori A, Toma T (eds) *Aquaculture in tropical areas*. Midoro Shobo, Tokyo, p 313–328 (English edn prepared by M. Yamaguchi)
- ✦ Siddon CE, Witman JD (2003) Influence of chronic, low-level hydrodynamic forces on subtidal community structure. *Mar Ecol Prog Ser* 261:99–110
- ✦ Stimson J, Cunha T, Philippoff J (2007) Food preferences and related behavior of the browsing sea urchin *Tripneustes gratilla* (Linnaeus) and its potential for use as a biological control agent. *Mar Biol* 151:1761–1772
- ✦ Takagi H, Esteban M (2016) Statistics of tropical cyclones in the Philippines: unusual characteristics of 2013 typhoon Haiyan. *Nat Hazards* 80:211–222
- Talae-McManus LT, Kesner KP (1995) Valuation of a Philippine municipal sea urchin fishery and implications of its collapse. In: Junio-Meñez MAR, Newkirk GF (eds) *From the Fourth Annual Common Property Conference*, Manila, p 229–239
- Trono GC Jr (1997) Field guide and atlas of the seaweed resources of the Philippines. Bookmark, Manila
- Trono GC Jr (2004) Field guide and atlas of the seaweed resources of the Philippines, 2<sup>nd</sup> edn. Bookmark, Makati City
- Tuason AY, Gomez ED (1979) The reproductive biology of *Tripneustes gratilla* Linnaeus (Echinodermata: Echinoidea) with some notes on *Diadema setosum* Leske. *Proc Int Symp Mar Biogeogr Evol Southern Hemisphere* 2:707–716
- ✦ Vaïtilingon D, Rasolofonirina R, Jangoux M (2003) Feeding preferences, seasonal gut repletion indices and diel feeding patterns of the sea urchin *Tripneustes gratilla* (Echinodermata: Echinoidea) on a coastal habitat off Toliara (Madagascar). *Mar Biol* 143:451–458
- Valentine JF, Duffy JE (2006) The central role of grazing in seagrass ecology. In: Larkum AWD, Orth RJ, Duarte CM (eds) *Seagrasses: biology, ecology and conservation*. Springer, Dordrecht, p 463–501
- Villanoy CL, Salamante E, Cabrera O (2013) Exposure: waves and storm surges. In: Geronimo RC, Folloso NMG, Quibilan MCC, Samson MS, Tiquio MGJP (eds) *Vulnerability assessment tools for coastal ecosystems: a guidebook*. Marine Environment and Resources Foundation, Quezon City, p 44–55

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