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José Luis Viveros-Legorreta
Universidad Nacional Autónoma de México
Edgar Peláez-Rodríguez
Universidad Nacional Autónoma de México
Raúl Cueva del Castillo
rcueva@ecologia.unam.mx

Universidad Nacional Autónoma de México

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Inter-population differences in size and sexual size dimorphism in the clam *Donax variabilis*

José Luis Viveros-Legorreta[1], Edgar Peláez-Rodríguez[1] and *Raúl Cueva del Castillo[2]

Universidad Nacional Autónoma de México; Facultad de Estudios Superiores Iztacala,

[1]Carrera de Biología, [2]UBIPRO; Lab de Ecología.

***Corresponding author: rcueva@ecologia.unam.mx**

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Abstract

Sexual size dimorphism (SSD) is well-documented in species with internal fertilization but less so in those with external fertilization. In such species, In such species, SSD may primarily be shaped by fecundity selection. This study highlights notable variations in SSD and body size of *D. variabilis* across three beaches along the Veracruz coast, Mexico. While female fecundity was consistent across all beaches, resource allocation towards egg production and female size varied. One beach featured larger females showing a strong relationship between shell length and fecundity, with a hyperallometric link between shell length and width, indicating intense fecundity-driven selection. Additionally, in this beach we found a strong male-biased sex ratio and the males were larger and exhibited also a hyperallometric pattern, pointing to stronger sexual selection pressures. These findings suggest that the interplay between fecundity selection and sexual selection may explain the interpopulation divergence in *D. variabilis* populations.

Introduction

Sexual size dimorphism (SSD) varies greatly across animal groups, with males being larger in birds and mammals (male-biased SSD) and females larger in species like fish, snakes, turtles and many species of invertebrates (female-biased SSD) (Ghiselin 1974; Reiss 1989; Fairbairn et al 2007, 2013). These striking differences in male and female morphology have inspired various hypotheses to explain the evolutionary and ecological drivers of SSD (Jehl and Murray 1986; Hedrick and Temeles 1979; Shine 1989; Andersson 1994; Fairbairn 2007; Székely et al 2007). Three major hypotheses have received the greatest attention: sexual selection, fecundity selection and niche divergence between sexes (Shine 1989; Fairbairn 1997; Blanckenhorn 2005). The sexual selection hypothesis suggests that evolutionary pressures have likely shaped the differences in body size observed between males and females. In many animal species, male body size is strongly linked to reproductive success, especially in polygynous systems, where larger males often secure more mating opportunities (Andersson 1994). On the other hand, natural selection frequently favors sexual size dimorphism leaning toward larger females, as larger female body size is associated with increased egg size and larger clutch size (Ridley 1983; Ridley and Thompson 1979; Roff 1991, 1992; Honek 1993). Additionally, variations in body size between sexes could result from adaptations to different ecological roles, with such evolutionary differences potentially arising from resource competition (Darwin 1871; Shine 1989; Stephens and Wiens 2009).

The research on sexual size dimorphism (SSD) has primarily focused on vertebrates and invertebrates with internal fertilization (Blanckenhorn 2005; Fairbairn et al 2007; Fairbairn 2013), leaving species with external fertilization relatively underexplored (Levitan 2010). Sexual size and shape dimorphism is rare among externally fertilizing

marine invertebrates (Levitan 1998). Darwin (1871) suggested that the lack of SSD in this group of animals is because they do not experience sexual selection. Even when excluding sexual selection as a driving force behind the evolution of SSD, other selective pressures can account for its evolution. (Hedrick and Temeles 1979; Shine 1989; Arnold 1994). In organisms such as most bivalves, corals, lampshells, and sea lilies, the relative importance of sexual selection and fecundity selection can differ markedly from that observed in internally fertilizing species (Levitan 2010). With gametes released into the water, fertilization depends on gamete compatibility, sperm competition, or selective sperm choice rather than male size-related reproductive success. While larger males might produce more sperm due to larger gonads, their advantage is constrained by the need for precise sperm–egg synchronization and by the rapid dilution of gametes in the water column. (Levitan 1991, 1998; Styan and Butler 2003; Beekman et al 2016). Perhaps for these reasons, size does not predict the amount or rate of induced sperm release (Styan and Butler 2003). Instead, fecundity selection is likely to play a more prominent role, favoring larger females due to their higher egg production and reproductive output. Natural selection favors larger females, as larger size increases egg production and reproductive success in vertebrates and invertebrates (Ridley 1983; Roff 1991, 1992), including bivalve mollusks (Haag and Staton 2003). In addition, males and females may be subject to different selective pressures if they occupy distinct ecological niches or rely on different food resources (Shine 1989).

In this study we explored variations in body size and SSD in the gonochoristic bivalve *Donax variabilis* across three Gulf of Mexico beaches near the Naval Academy in Anton Lizardo, Veracruz, Mexico. The species is distributed along the coast of the Gulf of Mexico from Florida to the Yucatan Peninsula (Abbott 1989), inhabiting surf-washed zones on open-ocean sandy beaches between tide lines (Turner and Belding 1957). The

reproductive cycle spans approximately eight months, with spawning and recruitment peaks occurring in early spring. Fertilization occurs after spawning, which is induced by chemical and physical environmental stimuli. The fertilized eggs pass through various larval stages before developing into mature shellfish. Spawning activity and lower recruitment densities continue through summer and autumn, and the life span of the species is less than one year (Cobb et al 2011). In the genus, sex is genetically determined at fertilization without sex chromosomes (Cornet and Soulard 1990), and the ratio of males to females varies significantly between populations of the species of the genus, from nearly equal proportions of 1:1 (Kandeel et al 2017) to places where there is a noticeable male bias (Ben-Haddad et al 2025).

We hypothesized that environmental differences between beaches influence body size and SSD. Specifically, dissolved oxygen below critical thresholds, reduced pH, salinity outside species-specific tolerance ranges and strongly reducing (negative ORP) conditions all impose energetic costs on bivalves that are expected to constrain growth (Waldbusser et al 2011; Lamers et al 2013; Thomas et al 2019; Podbielski et al 2022; Kotsyuba and Dyachuk 2023), whereas lower temperatures are associated with slower growth but larger adult body size in marine ectotherms (Jones et al 2004; Moss et al 2016; Verberk et al 2021).We predicted that favorable conditions with abundant resources would support larger individuals. Furthermore, such environments could enhance SSD bias toward females, as females in these places could allocate more resources to egg production. Of course, other parameters, such as intra- and interspecific competition or predation, could contribute to interpopulation differentiation. Nonetheless, the primary emphasis was on investigating how abiotic factors influence this differentiation.

Differences between males and females in *D. variabilis* were analyzed by examining static allometry (Pélabon et al 2014; Stillwell et al 2016) between adult shells' length and width. Phenotypic divergences between males and females frequently arise as each of them plays different reproductive roles and experiences a sex-specific combination of selection forces acting upon their traits. However, when the selective pressures acted on the same trait in both sexes is similar, it is likely to yield comparable allometric slopes (Voje 2016; but see Bonduriansky 2007), reducing the level of SSD.

Material and Methods

Collection of D. variabilis

Specimens of *D. variabilis* clams were collected during the months of March, May, July, and November of 2022 from three beaches in Veracruz, Mexico: Playa Vicente Fox in Boca del Río (Beach 1), as well as El Jiote (Beach 2) and Barrancas (Beach 3) in Antón Lizardo municipality (Fig. 1). At each beach, five 10-meter-long transects were established, distributed perpendicularly to the shoreline, from the supralittoral to the infralittoral zone, with a spacing of 20 meters between transects to ensure representative coverage of the study area. The organisms were collected manually using two techniques: (1) sieving sand with a 1 mm mesh, which allowed capturing the organisms as they migrated with the waves, and (2) direct extraction, digging into the sand up to a depth of 10 cm at points where siphons were observed on the surface, indicating the presence of buried clams. The sampling effort was 30 minutes per transect, with two people working simultaneously to ensure homogeneity in the collection. Both sampling methods targeted individuals migrating with the current, while direct extraction targeted larger, buried ones. We

acknowledge that the sampling method could influence the total number of individuals captured. However, our goal was to analyze potential differences between beaches in terms of body size and sexual dimorphism of clams.

The collected clams were placed in PVC containers for transport and later fixed with 5% formalin for preservation. Each sample was meticulously labeled with detailed information, including the collection site, date, sample number, and transect of origin, ensuring the highest level of accuracy and traceability of the data for subsequent laboratory analysis.

Shell length and width for each organism were measured using a digital caliper (±0.01 mm accuracy, Mitutoyo Corp., Tokyo, Japan). Length referred to the maximum anteroposterior distance, while width represented the greatest lateral distance between the valves.

On each of the four sampling occasions, environmental parameters were measured at four points per beach, yielding 16 measurements per beach and 48 in total, using a Hanna HI 9828 multiparameter probe; a portable multiparameter instrument designed for field measurements of lakes, rivers, and marine environments. (Akinnigbagbe et al., 2025). The parameters measured included dissolved oxygen (ppm and %), pH, oxidation-reduction potential (ORP), temperature (°C), total dissolved solids (TDS, ppt), and salts (ppt). The probe also measured conductivity, but it was not separately analyzed because it was used by the instrument to calculate salinity. Bivalves experience significant physiological disturbances and altered energetic budgets when dissolved oxygen falls below critical thresholds (~2.0 mg/L or ~30% saturation; Kotsyuba and Dyachuk 2023; Thomas et al 2019), at reduced pH levels (< 7.6 in marine settings) significantly increase net shell dissolution rates and elevate metabolic maintenance costs (Waldbusser et al 2011),

exposure to salinity outside species-specific tolerance ranges leads to mantle retraction, reduced feeding, and osmotic cell damage (Podbielski et al 2022), and negative ORP at the sediment-water interface indicate anoxic microbial decomposition; under strongly reducing conditions, which inhibits aerobic respiration in benthic macrofauna (Lamers et al 2013).

Individuals sex identification

The sex of the collected clams was identified by examining their gonads, their shells were carefully opened by cutting the adductor muscles and removing the mantle. Under a Motic SMZ168 stereomicroscope (10x–40x), sex was identified based on the color and texture of the gonads: male gonads appeared creamy with small granules, while female gonads showed a pinkish hue with larger granules. For ambiguous cases, particularly in smaller specimens, gonad smears were prepared to detect gametes.

Oocyte counting and fecundity analysis

Oocyte counts were performed only on females with a shell length of ≥6 mm, as smaller organism posed challenges due to technical handling limitations. Females in advanced reproductive stages were identified by fully developed gonads with mature oocytes visible under stereomicroscopy. Gonadal tissue from these females was subsequently processed using histological techniques to quantify and characterize oocytes.

The tissues were fixed in 10% formalin for 24 hours and then embedded in paraffin. Histological sections of 12 µm thickness were made using a Leica RM2125 RTS microtome, following standardized protocols for bivalve tissues. Sample preparation began with deparaffinization of the histological sections in xylene, followed by rehydration in

absolute alcohol. Staining was conducted using the Diff-Quick kit, involving sequential application of fixative solution I and solution II, each for five minutes. A glacial acetic acid rinse was employed to enhance dye fixation, succeeded by a brief exposure to absolute alcohol to preserve coloration before rapid drying and mounting in Canada balsam. For oocyte analysis, histological sections were examined under a Nikon YS2-T compound microscope at 100x magnification. A systematic selection of representative oocytes was performed, and their diameters were measured with a micrometric eyepiece, ensuring precision and reliability in the assessment of reproductive structures.

Oocyte quantification was based on three non-consecutive histological sections per individual, since counting all oocytes within the gonad is impractical due to their high abundance and overlapping developmental stages. To prevent double counting, only vitellogenic oocytes sectioned through the nucleus were considered, and their diameters were measured with a micrometric eyepiece.

Statistical analyses.

Differences in environmental parameters between beaches

Since *D. variabilis* exhibits indeterminate growth and has a life cycle shorter than a year, only individuals with differentiated gonads were included in the analyses. This approach ensures that the samples represent cohorts that are relatively homogeneous in age, allowing for more accurate and consistent results.

We performed univariate ANOVAs to assess differences in each environmental parameter between beaches. Deviations from a 1:1 sex ratio at each beach were evaluated using a chi-square goodness-of-fit test. With the exception of major axis regressions, all statistical analyses were conducted using JMP 10 software (JMP 10, SAS Institute, Cary, NC).

Sexual size dimorphism

For sexual size dimorphism, just individuals with well differentiated gonads were included in the analyses. The data collected over the duration of the study were combined prior to conducting statistical analyses, and the morphometric variables were log10 transformed to meet residual homoscedasticity and the normality assumption of the parametric test and to fit the allometric relationships between the variables.

We conducted a multivariate analysis of variance (MANOVA) to assess potential differences across populations, sexes, and the magnitude of sexual dimorphism (interaction Population*Sex) across shell length and width. Univariate ANOVAs were carried out to further explore these findings. Because multiple comparisons were performed, P values were corrected using the Bonferroni method to reduce the experiment-wise error rate (Sokal and Rohlf 1995).

Male and female allometric relationships

Major axis (MA) regressions were used to analyze the relationship between shell width and length in males and females for the three populations. MA regression was chosen for its suitability in summarizing relationships between variables on logarithmic scales and providing accurate slope estimates, as it accounts for residual variation in both variables

(Warton et al 2006). Allometric slopes were compared between sexes using a Wald test. Analyses were conducted in SMATR v. 2.0 (Warton et al 2006). As males and females often experience distinct evolutionary pressures, deviations from isometry, such as hyperallometry (slope > 1) or hypoallometry (slope < 1), can reflect adaptive scaling patterns in one sex (Petrie 1988, 1992; Eberhard et al 1998; Frankino et al 2009). Such divergences may result from the interplay of natural and sexual selection (Cornwallis and Uller 2010; Maan and Seehausen 2011; Scales and Butler 2016).

Female body size and fecundity

An ANCOVA was conducted to assess differences in fecundity (number of oocytes) among females' shell length from three beaches. The analysis included beach as the independent variable, log females shell length as a covariate, and the interaction between them. Because multiple comparisons were performed, P values were corrected using the Bonferroni method (Sokal and Rohlf 1995).

Results

Differences in environmental parameters between beaches

We did not detect differences in dissolved oxygen ($F_{(2, 45)} = 0.471$, $P = 0.628$), percent of oxygen ($F_{(2, 45)} = 0.13$, $P = 0.875$), in pH ($F_{(2, 45)} = 0.185$, $P = 0.832$), and total of dissolved solids ($F_{(2, 45)} = 3.03$, $P = 0.058$) levels of the three beaches. However, there were differences between them in temperature ($F_{(2, 45)} = 4.110$, $P = 0.023$), salts ($F_{(2, 45)} = 9.671$, $P < 0.001$), and oxidation-reduction potential ($F_{(2, 45)} = 45.701$, $P < 0.001$). Beach 2 recorded

the highest water temperature (27.56±0.52°C), and oxidation-reduction potential (130.04 ± 4.19mV), while Beach 1 had the highest salinity (32.26 ± 0.35 mg/l), (Table 1).

A total of 2343 individuals of *D. variabilis* were collected across the three beaches. In Beach 1 were collected 334 males and 114 females, Sex Ratio (M/F): 2.93, (X^2 = 108.036; DF = 1, P < 0.0001); in Beach 2 were collected 54 males and 139 females, Sex Ratio (M/F): 6.86 (X^2 = 607.708, DF = 1; P < 0.0001); and in Beach 3 were collected 672 males and 130 females, Sex Ratio (M/F): 5.17 (X^2 = 366.289, DF = 1; P < 0.0001).

Body Size and Sexual Size dimorphism

The MANOVA indicated that there are body size differences between populations (Wilks' Lambda = 0.854$F_{(4, 4672)}$ = 95.430, P< 0.0001), sexes ($F_{(2, 2336)}$ = 322.537, P< 0.0001), and the magnitude of sexual size dimorphism between populations (Wilks' Lambda = 0.924, $F_{(4, 4672)}$ = 46.570, P < 0.0001).

Shell length: Univariate ANOVA revealed significant differences in shell length among populations ($F_{(2, 2342)}$ = 11.486, P < 0.0001; 0.01 after Bonferroni Correction (BC)), females were larger than males ($F_{(1,2342)}$ = 566.326, P < 0.0001; 0.0083 after BC), and the degree of sexual size dimorphism varied between populations (Population × Sex interaction: $F_{(2, 2342)}$ = 4.204, P = 0.015; 0.016 after BC). However, just beaches 1 and 2 exhibited significant differences between them (Population * Sex Interaction: $F_{(1, 1537)}$ = 9.600, P = 0.002; 0.0125 after BC), Beaches 1 and 3: (Population * Sex Interaction: $F_{(1, 1246)}$ = 1.286, P = 0.257), Beaches 2 and 3:(Population * Sex Interaction: $F_{(1, 1891)}$ = 2.912, P = 0.088; Fig 2a).

Shell width: Univariate ANOVAs revealed no significant differences among populations ($F_{(2, 2342)} = 9.971$, $P = 0.378$), significant differences between sexes ($F_{(1, 2342)} = 639.319$, $P < 0.0001$; 0.0083 after BC), the shell width was larger in females. Moreover, the degree of SSD varied between populations (Population × Sex interaction: $F_{(2, 2342)} = 18.762$, $P < 0.0001$; 0.0166 after BC). Differences in the shell width between Beaches 1 and 2 ($F_{(1, 1537)} = 34.563$, $P = P < 0.0001$, 0.01 after BC), and between 2 and 3 were significant ($F_{(1, 1891)} = 21.909$, $P < 0.0001$; 0.025 after BC; Fig 2b). However, the magnitude of SSD was not significant between Beaches 1 and 3 ($F_{(1, 1246)} = 1.285$, $P = 0.257$).

Male and female allometric relationships

The study reveals significant deviations from isometry in the shell length-to-width relationship for males and females across the three beaches. On beaches 1 and 3, both males and females displayed significant hypoallometric ratios, whereas at beach 2, both showed hyperallometry (Table 2), and their slopes differ significatively between them, whereas in Beaches 1 and 3 there were not significant differences between the slopes of males and females (Table 3).

Female body size and fecundity

The ANCOVA results indicated that the average fecundity was similar among the three populations, despite of in Beach 3 we found the smallest female of the study ($F_{(2, 377)} = 0.236$; $P = 0.789$). In addition, we found a strong positive relationship between shell length

279 and fecundity, suggesting that larger body size is associated with higher fecundity ($F_{(1, 377)}$
280 = 57.88; P < 0.0001; 0.01 after BC Fig. 4). However, the body size-fecundity relationships
281 differed between the three populations ($F_{(2, 377)}$ = 7.897; P = 0.0004, 0.0125 after BC).
282 Beach 3 and beach 2 exhibited respectively the weaker (β = 95.501, $F_{(1, 128)}$ = 7.755, P =
283 0.006; 0.025 after BC) and the stronger (β = 286.26, $F_{(1, 137)}$ = 62.046, < 0.0001; 0.0083
284 after BC; Fig. 4c) link between body size and fecundity, while population 1 shows an
285 intermedium relationship (β = 195.95, $F_{(1, 112)}$ = 13.829, P = 0.003; 0.0166 after BC).
286 Interestingly
287
288 **Discussion**
289 Sexual dimorphism is rare among externally fertilizing marine invertebrates. However, this
290 study reveals significant differences in body size and SSD in *D. variabilis* across three
291 beaches along the coast of Veracruz, Mexico. A proportion of body size could be explained
292 by differences in environmental conditions. Even though clam sizes differed across the
293 three beaches, we found that the largest clams would be growing in suboptimal conditions
294 at Beach 2, which had the warmest water, lower salinity and high oxidation-reduction
295 potential (ORP) (Table 1). In *D. variabilis* and other marine ectotherms cold temperatures
296 favor slower growing and a longer development time (Jones et al 2004; Moss et al 2016;
297 Verberk et al 2021), while in this and the other beaches ORP are far from the optimal
298 conditions; +150 to +250 mV (Turner and Belding 1957; Castagna and Chanley 1973),
299 although they are in the range of the optimal salinity (20-36 ppt; Chanley 1969; Ansell
300 1983), and temperature (20°C to 28°C) for their aerobic metabolism, feeding, and shell
301 calcification (Chanley 1969; Wilson 1999; Joyner-Matos et al 2006).

While environmental factors may partly explain the variations among *D. variabilis* populations, the divergence between them could result from the interplay between natural and sexual selection. In these and many ectothermic species, females are generally larger, correlating their size with fecundity and indicating stronger fecundity selection compared to sexual selection in males (Fairbairn 2013). Despite consistent female fecundity across three beaches, there are differences in resource allocation for egg production. Beach 2 females display the highest size-fecundity correlation, are larger, and have a hyperallometric shell ratio, suggesting substantial fecundity selection pressures. In contrast, females from the other beaches are smaller and show a hypoallometric ratio. The discrepancy in the intensity of fecundity selection between the populations could explain the highest SSD bias toward females in Beach 2. On the other hand, Beach 2 males are comparable in size to Beach 1 males but exhibit a hyperallometric relationship with a steeper slope compared to both their female peers and males from Beaches 1 and 3, which show a hypoallometric ratio. Additionally, the sex ratio on Beach 2 is highly biased toward males. Together, hyperallometry and an extreme sex ratio bias toward males suggest more intense sexual selection on Beach 2 males, surpassing both the fecundity selection on their females and the sexual selection exerted on males from the other beaches, which also have a similar hypoallometric trend to the females from their populations. Interestingly, the similar allometric ratios of males from Beaches 1 and 3 with their female counterparts may suggest that the intensity of fecundity and sexual selection has been similar in these populations.

In species such as most clams, where both sperm and eggs are released into the water for external fertilization, local sperm and egg availability post-release determines the nature and intensity of sexual selection (Levitan 2010). Sexual selection arises through gamete compatibility, sperm competition, or sperm choice (Beekman et al 2016), involving

simultaneous competition by both sexes. While larger body size doesn't directly enhance access to gametes, it often correlates with larger gonads and higher gamete output. Males producing more sperm during synchronized spawning events may gain a competitive edge in fertilizing eggs.

Together, our results suggest that environmental differences plus the interplay between fecundity selection and sexual selection may explain the interpopulation divergence in *D. variabilis*. However, we must point out that although natural selection is generally thought to favor the evolution of positive allometric traits (Bonduriansky and Day 2003; Bonduriansky 2007), also it can be a structural consequence of organismal architecture and development (Gould and Lewontin 1979). Experimental studies are needed to evaluate the intensity of natural and sexual selection on these populations. We are currently assessing the size of the gonads in males and examining their relationship with body size, in addition to estimating the production of their gametes as a function of their body size.

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Declarations

Conflicts of interest: None declared.

Ethics approval

The specimens used in this study were collected under the permit PPD/DGOPA-001/25 issuedby Secretaría de Agricultura y Desarrollo Rural; Comisión Nacional de Acuacultura y Pesca.

The authors of this work have agreed to their participation in it and have given their consent for its publication.

Data availability Data will be available under reasonable request.

Authors' contributions: RCC originally formulated the idea, JLV-L, EP-Rand RCC developed methodology, JLV-L and EPR conducted fieldwork, JLVL EPR and RCC performed statistical analyses, JLVL EPR and RCC wrote the manuscript.

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512 Figure legends

513 Fig. 1. Geographic location of the three beaches studied in the coast of Veracruz, Mexico.

514

515 Figure 2. Differences for the shell length (a) and width (b) of males and females of *D.*
516 *variabilis* from three Veracruz beaches, Mexico. In both traits the magnitude of
517 sexual dimorphism (interaction Population* Sex) resulted significant.

518

519 Figure 3. Allometric Major Axis regressions of shell width on shell length in males and
520 females of *D. variabilis* across three beaches of Veracruz, Mexico. a): Beach 1, b):
521 Beach 2, and c): Beach 3. Dashed lines with open triangles denote the values and
522 slopes for males, while solid lines with filled circles correspond to the values and
523 slopes for females.

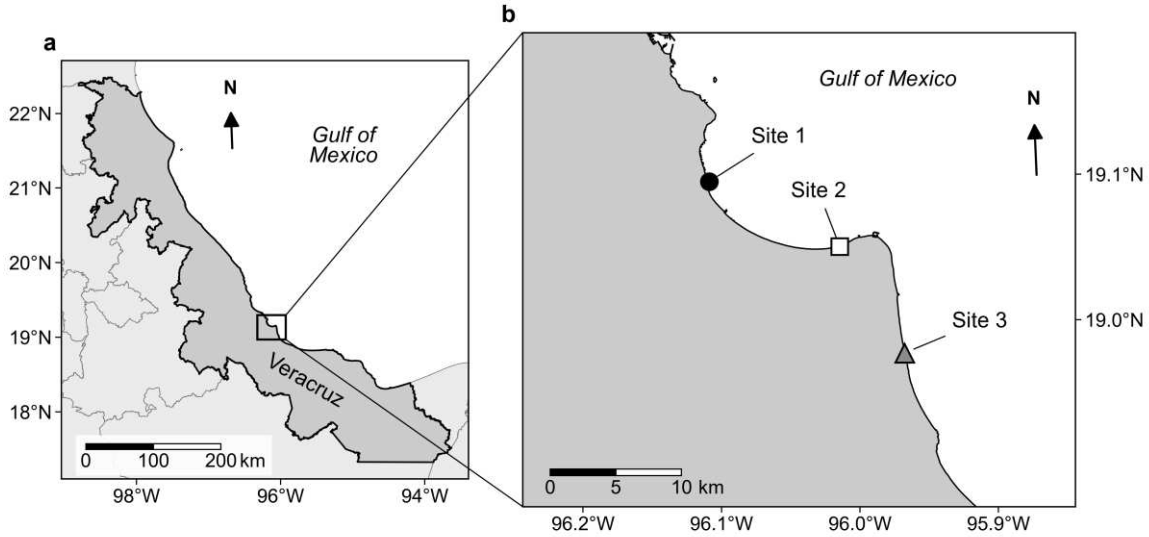
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525 Figure 4. Relationship between shell length and oocyte count in females of *D. variabilis*
526 across three beaches of Veracruz, Mexico. a): Beach 1, b): Beach 2, c): Beach 3.

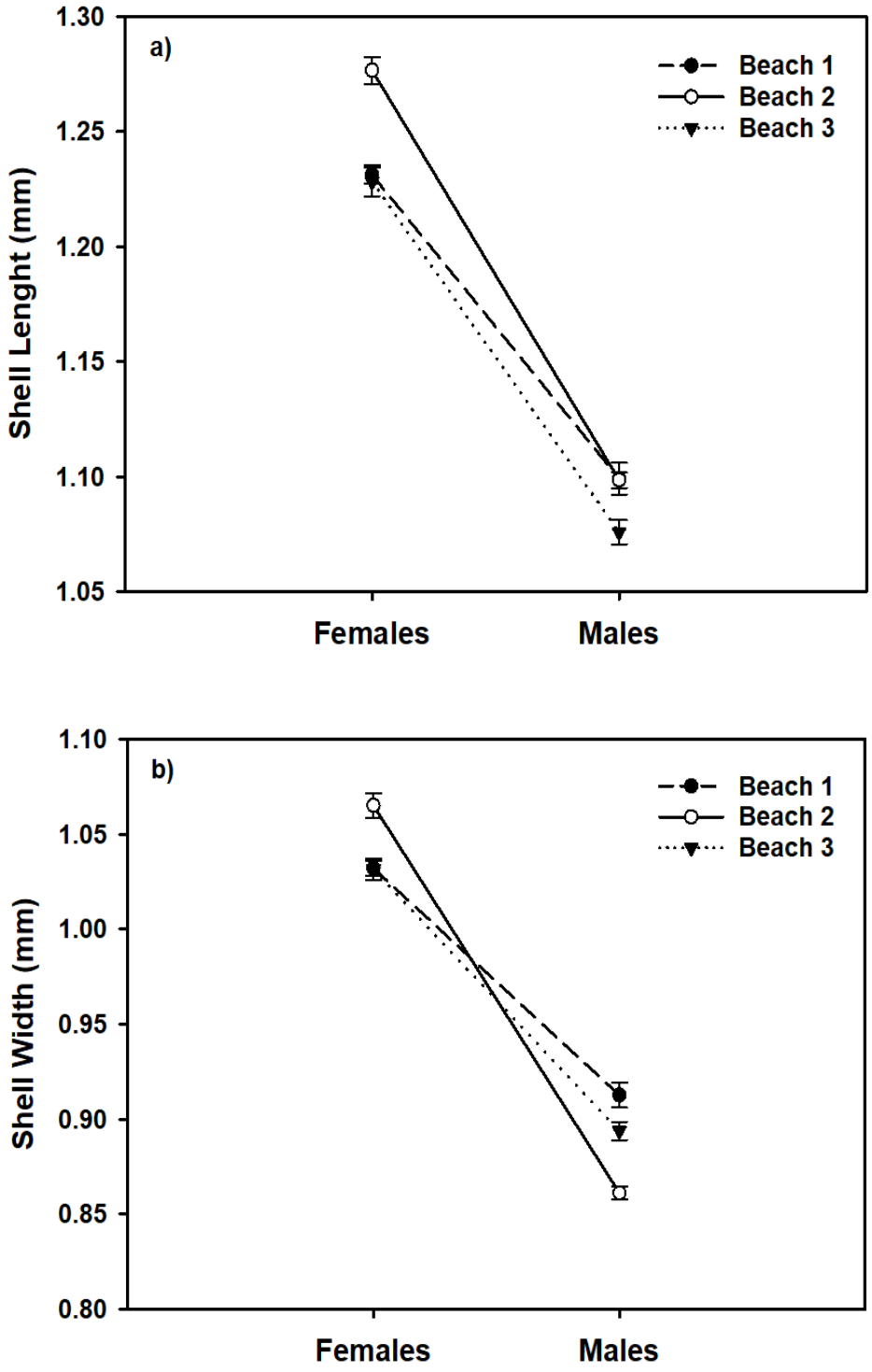
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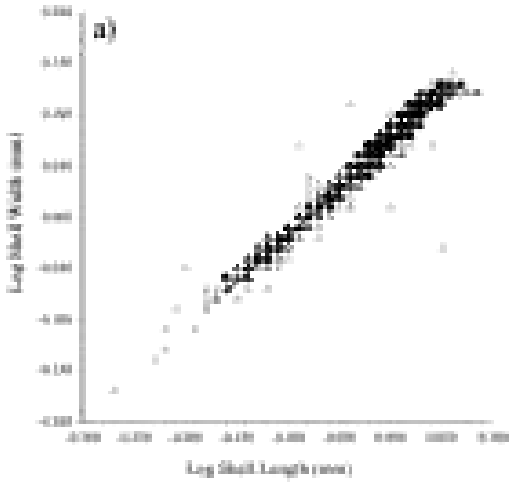
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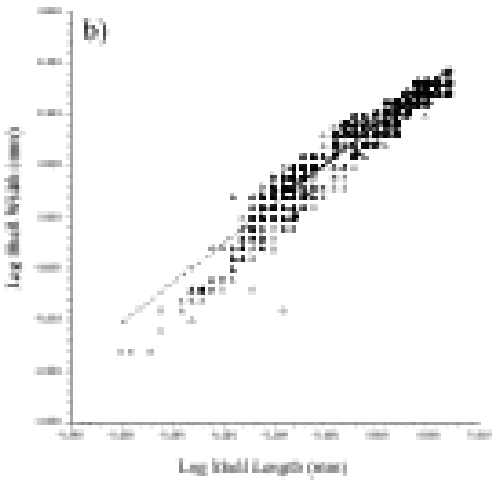
Fig 1

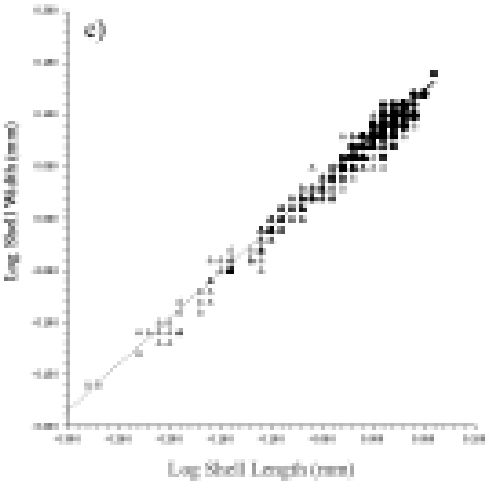

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b
N
Gulf of Mexico
Veracruz
22°N
21°N
20°N
19°N
18°N
0 100 200 km
98°W
96°W
94°W
Gulf of Mexico
Site 1
Site 2
Site 3
N
19.1°N
19.0°N
0 5 10 km
96.2°W
96.1°W
96.0°W
95.9°W

551 Figure 2
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a)
Log Shell Width (mm)
Log Shell Length (mm)


b)
Log Shell Width (mm)
Log Shell Length (mm)


c)
Log Shell Width (mm)
Log Shell Length (mm)

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559 Figure 4
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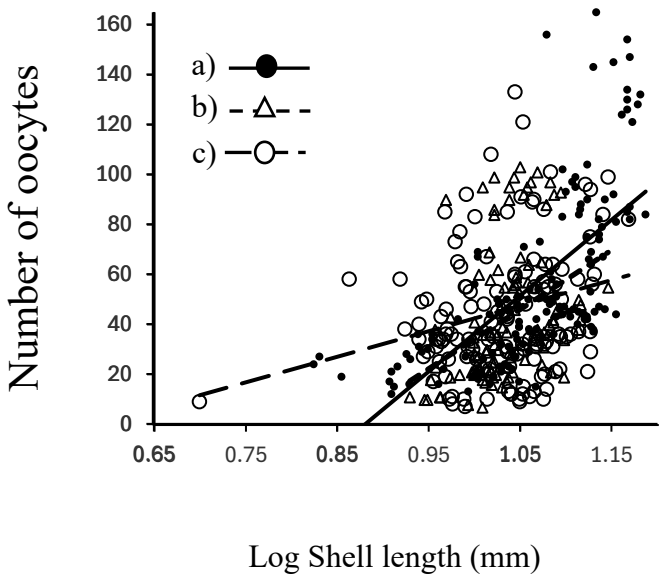


Table 1. Average environmental parameters measured in three beaches of the coast of Veracruz, Mexico. DO (ppm): Dissolved Oxygen (parts per million), DO%: Percent of Dissolved Oxigen, pH: hydrogen ion concentration, ºC: Degrees Celsius, TDS (ppm): Total of Dissolved Solids (parts per million), Salts: (mg/l); ORP: Oxidation -Reduction potential (mV). Accordingly, to Tukey's HSD test, **s**hared letters indicated no significant differences between beaches according to Holm-Sidak method.

Parameter	**Beach 1**	**Beach 2**	**Beach 3**
DO	3.08±0.45[a]	2.84±0.41[a]	3.49±0.55[a]
DO%	46.53±6.85[a]	45.43±6.79[a]	50.40±7.69[a]
pH	7.67±0.13[a]	7.60±0.12[a]	7.71±0.15[a]
ºC	25.80±0.37[a,c]	27.56±0.52[b,c]	26.41±0.40[c]
TDS ppt	24.54±0.27[a]	22.74±0.53[a]	24.38±0.79[a]
Salts	32.26±0.35[a]	28.94±0.72[b]	30.79±0.45[c]
ORP	69.85±10.16[a]	130.04±4.19[b]	43.23±2.97[c]

Table 2. Allometric relationships between the shell length and width of males and females of *D. variabilis* at three beaches in the coast of Veracruz; Mexico. Their significant deviations from isometry (H_0 β =1), sample size for males and females of each beach (n), variance explained (R^2), Slopes, Upper and Lower Confidence Intervals (CI), and F, and P values are shown. * Significant differences between males and females (P= 0.001).

Beach	Sex	n	R^2	Slope	Low CI	UppCI	H_0 β	F	P
1	Male	334	0.97	0.930	0.914	0.948	1	58.185	< 0.0001
1	Female	114	0.82	0.908	0.830	0.993	1	4.578	0.035
2	Male*	954	0.48	1.514	1.418	1.620	1	159.714	< 0.0001
2	Female*	139	0.86	1.147	1.070	1.231	1	15.378	< 0.0001
3	Male	672	0.96	0.894	0.880	0.908	1	203.457	< 0.0001
3	Female	130	0.92	0.889	0.846	0.935	1	21.606	< 0.0001

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Table 3. P values of post-hoc multiple comparison of allometric slopes of males (M) and females (F) of *D. variabilis* at three beaches in the coast of Veracruz, Mexico

Beach-Sex	1 M	1 F	2 M	2 F	3 M	3 F
1 M	1	0.577	**0.001**	**0.001**	**0.002**	0.092
1 F		1	**0.001**	**0.001**	0.748	0.677
2 M			1	**0.001**	**0.001**	**0.001**
2 F				1	**0.001**	**0.001**
3 M					1	0.830
3 F						1

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