

Gametogenic cycle of the tropical vermetids *Eualetes tulipa* and *Dendropoma corrodens* (Mollusca: Caenogastropoda: Vermetidae)

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The population density, annual gametogenic cycle, and size at sexual maturity of two vermetid species, Eualetes tulipa (invasive) and Dendropoma corrodens was studied in the southern Caribbean at the Planta Centro power plant intake channel and at Punta Mayorquina, Morrocoy National Park, respectively. Minimal size of mature females of E. tulipa and D. corrodens was 5.0 and 0.7 cm in body length respectively. The gonad represented between 44 to 47% of total body length, while it represented between 15 to 18% of total body volume (for both males and females). Gonad volume was significantly related to total body volume. A significant quadratic relationship between the number of capsules and female body volume was obtained for E. tulipa, indicating that females measuring 6–10 cm in body length are those that have the highest number of egg capsules. Females larger than 11 cm have a lower fecundity in terms of brooded capsules. Gonad sections of males and females of both species showed that several gametogenesis stages are simultaneous within the same individual. In the female, germ cells, oogoniums and primary oocytes with yolk granules were observed simultaneously. In the male, the spermatogonium, primary and secondary spermatocytes, spermatids and sperm cells were also observed within one gonad. The same pattern was observed throughout the year, showing no maturation/resting period or annual cycle. The area covered by D. corrodens was about 700 m² with a mean density of 13,000 ind/m². The area covered by the invasive E. tulipa was at least about 1000 m², with a density of 27,000 to 29,000 ind/m² in the first 3 m of depth. A follow up of the E. tulipa population showed that its density is increasing in time and has doubled in 20 years. This population increment could be related to an increase in organic suspended matter as evidenced in the region.

Keywords: Vermetidae, Gastropoda, gonad, gametogenesis, reproduction, Caribbean, invasive species

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INTRODUCTION

Gastropods of the family Vermetidae are characterized by the production of eggs contained in egg capsules which are brooded by the female freely in the mantle cavity (Morton, 1965; Hadfield *et al.*, 1972; Hughes, 1979; Miloslavich & Penchaszadeh, 1992) or attached to the internal side of the shell (Morton, 1951, 1965; Hadfield *et al.*, 1972; Hughes & Lewis, 1974; Bandel, 1975, 1976; Hughes, 1979). The spawn is composed of several individual egg capsules, spherical or elongated with a fragile membrane, their size and number depending on the species. Also, the production of eggs (1 to 400 eggs per capsule), egg diameter (100 to 480 µm), hatching mode (veliger larva or crawling juvenile) and the presence of nurse eggs are species specific (see Miloslavich & Penchaszadeh, 1992 for a review of some reproductive traits in Vermetidae). Most of the literature dealing with reproductive aspects in Vermetidae is focused then in describing egg capsules, embryos, and hatching modes, and studies on the gametogenesis are very rare. With few exceptions (Hodgson

& Eckelbarger, 2000), who studied the oogenesis in six species of patellid limpets from South Africa, gametogenic studies in gastropods are mostly limited to commercial species such as the *Buccinum undatum* Linnaeus, 1758 from east Canada (Martel *et al.*, 1986a, b; Gendron, 1992; Himmelman & Hamel, 1993), *Zidona dufresnei* (Donovan, 1823) from Argentina (Giménez & Penchaszadeh, 2002, 2003), or to imposex indicators such as *Reishia (Thais) clavigera* (Kuster, 1858) from the west coast of Korea (Lee, 1999) and *Nassarius reticulatus* Linnaeus, 1758 from the north-west of Portugal (Barroso & Moreira, 1998). In vermetids, the annual reproductive cycle has been studied in *Vermetus triquetrus* Bivona, 1832, *Dendropoma petraeum* (Monterosato, 1848) and *Serpulorbis arenaria* Linnaeus, 1767, three species from the coast of Spain (Calvo *et al.*, 1998; Calvo, 1999; Calvo & Templado, 2004a, b). Strathmann & Strathmann (2006) discussed the importance of the reproductive mode of a Hawaiian vermetid gastropod in achieving a high potential for invasion and establishment following maritime transport or natural rafting, and predicts a widespread distribution and invasion to warm waters.

Eualetes tulipa Chenu, 1843, was originally described as *Vermetus tulipa* by Chenu (1842–1853), however, no locality was specified for this material. Keen (1971) renamed this

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species as *Eualetes tulipa* and reported it for Panama Bay in the Pacific Ocean. Another report in the Panama area is at the Veraguas Province (Vega & González, 2002). In Hawaii, *E. tulipa* has been reported as a nonindigenous, introduced species in 32 localities including Kane'ohē Bay, Hilo Harbor, O'ahu Waikiki and O'ahu Honolulu, mainly attached to artificial substrates such as concrete pilings and walls, metal and wood substrates but also in natural environments such as reefs and mangrove roots (Coles & Eldredge, 2002; Coles *et al.*, 2002; Schlucker, 2003; Ray, 2005; database from the Bishop Museum, Hawaii). Specimens are large (about 5 cm in shell length) and tend to be solitary rather than colonial.

In the Venezuelan Caribbean, this species has been observed since at least 1986 (reported as *Vermetus* sp.) colonizing only on artificial substrates (Miloslavich & Penchaszadeh, 1992). Inventories of marine species in the Caribbean are far from being complete and Venezuela is not an exception. This lack of knowledge has the consequence that sometimes, when a new report is made on a species, it is difficult to accurately validate its status either as endemic or as introduced. The most complete local mollusc guide of the southern Caribbean is that of Díaz & Puyana (1994) which reports 5 species of Vermetidae belonging to the genera *Serpulorbis*, *Petalococonchus* and *Dendropoma*. For Venezuela, Tello (1975) reports 12 vermetid species within the genera *Petalococonchus*, *Serpulorbis* and *Siphonium*, which are mostly taxonomically out-dated names and none of which is *E. tulipa* or any of its synonyms. The problem of vermetid taxonomy is particularly complicated (Keen, 1961; Bieler, 1996), and few genera are defined by valid autapomorphic characters (Gould, 1994). Schiaparelli *et al.* (2006) when discussing the use of vermetid reef formations as sea level indicators stated that the complex taxonomy of this family has on occasions led researchers to mix different species, even belonging to different genera under a single name. With the development of recent molecular techniques, the confused vermetid taxonomy may be closer to a solution. In this aspect, Calvo *et al.* (2009) using mitochondrial genes cytochrome *c* oxidase subunit I (COI) and 16S rRNA (16S) sequences of *Dendropoma petraeum* from 18 different localities in the Mediterranean, discovered that there are at least 4 different cryptic species in what they called the *Dendropoma petraeum* complex. These genetic differences were also supported with reproductive information about intracapsular larval development and protoconch morphology. Reproductive studies on *E. tulipa* in the southern Caribbean point out that this species reproduces throughout the year. Females brood up to 54 egg capsules containing about 290 eggs each, of which around 190 develop to a veliger larva that settles within 24 hours of hatching (Miloslavich & Penchaszadeh, 1992).

Dendropoma corrodens D'Orbigny, 1842, is also a tropical species, originally described as *Vermetus corrodens* by D'Orbigny (1841) from material collected in Cuba and Martinique. This species is known in Cuba, east Florida, Bermuda, the Caribbean coasts of Colombia and Venezuela, and mid-Atlantic islands (Oliver, 1975; Miloslavich & Penchaszadeh, 1992; Díaz & Puyana, 1994). Specimens are small (about 1 cm) and live in colonies, encrusted to coral substrates. Females brood up to 8 egg capsules containing about 8 eggs of which 6 to 7 develop to a crawling juvenile that settles very close to the female (Miloslavich & Penchaszadeh, 1992).

The objective of this work is to study the annual reproductive cycle of *E. tulipa* and *D. corrodens*, their size at sexual maturity and gametogenesis, as well as the relationship between female size and fecundity. We will also estimate the density and area covered by populations of both species in their localities, and since *E. tulipa* has been reported as an invasive species, we will follow population density in time.

MATERIALS AND METHODS

Site description and population estimation

Specimens of *E. tulipa* (Figure 1) are found attached to the walls of the intake-cooling channel of the powerplant Planta Centro, in the central-west coast of Venezuela (10°30'6"N 68°9'36"W) Estado Carabobo. The channel is approximately 500 m long with a depth of 2 to 5 m, however, the zone that was accessible for our research consisted of the first 100 m. The channel has two parallel metal walls separated by a distance of 40 m. These walls are structured in columns of about 30–35 cm in width, being one column projected to the front (35 cm) and one column projected to the back (30 cm) with a 35 cm escalated space between both. Due to the functioning of the power plant, the channel has a constant flow of water from the sea to the engines that varies in speed depending on how many chimneys are working. The speed of the current in the channel varies between 0.13 and 0.23 m/second (Miloslavich, 1987) and the amount of suspended organic matter varies between 1.7 and 21.8 mg/l



Fig. 1. Adult individual of *Eualetes tulipa* after being extracted from the wall of the power plant channel at 0.5 m depth. Tube aperture is ~1 cm in diameter, and the shell is covered by epifauna.

(Miloslavich & Penchaszadeh, 1992). In 1986, all adult specimens from a total of 25 columns (5% of the total area) were counted up from the surface to the bottom (5 m). In 2006 and 2009, the same recount was made down to 3 m deep since the deeper two metres of the channel had been filled with fine sediment.

Specimens of *D. corrodens* colonize coralline rocks at Punta Mayorquina (10°53'45N 68°13'48"W), Morrocoy National Park, Estado Falcón, Venezuela, at a maximum of 1 m depth. The area has a mixture of coralline rocks, patches of the seagrass *Thalassia testudinum* and small colonies of the coral *Siderastrea siderea*. Water conditions at this locality are more oligotrophic and the amount of suspended organic matter varies between 0.2 and 7.5 mg/l (Miloslavich & Penchaszadeh, 1992). The total area colonized by *D. corrodens* was determined by bathymetry using as a reference the sea level at the coast and during high tide. The total number of juveniles (newly fixed to the rock) and adult specimens (embedded in the substrate) was counted in a total of 36 randomly selected rocks.

Gametogenesis

To study the gametogenic cycle of both species, between 10 and 14 adult specimens of *E. tulipa* were collected monthly from January to December 1986 from the walls of the channel, between 0 and 1 m depth. Coral rocks colonized by *D. corrodens* were collected from January to December 1986 at Punta Mayorquina at a maximum of 0.6 m depth.

The animals were extracted from the shell, left in Petri dishes with seawater until relaxation and measured. The soft body of the animal has a cylinder shape and the gonad can be distinguished from the digestive gland by its colour. In both species, female gonads are yellow, while male gonads are orange. The digestive gland is dark brown. We measured total length and diameter of the soft body (BL and BD) and of the gonad (GL and GD). The gonadosomatic index (GSI) was calculated by the relationship between total body–gonad volume with the following equation: $GSI (volume) = (GV/BV) \times 100$.

The volume of the gonad (GV) was calculated as the volume of a cylinder:

$GV = (\pi \times GD/2)^2 \times GL$, in which GD = gonad diameter and GL = gonad length. The body volume (BV) was calculated similarly as: $BV = (\pi \times BD/2)^2 \times BL$, in which BD = body diameter.

The total number of egg capsules brooded by each female was counted and the relationship between this number and female size, gonad size and GSI was determined.

After measuring the gonads, they were fixed in formalin (8% in seawater) for histological procedures using standard paraffin techniques and haematoxylin–eosin staining. Histological sections were observed and the diameter of the spermatogonium, oocytes and yolk granules was measured. Observations of live and preserved material were done with a ZEISS dissecting microscope and ZEISS microscope; measurements were taken with an ocular micrometer. Results are reported as mean \pm standard deviation.

Voucher adult material of *E. tulipa* (entire individuals fixed in formalin and preserved in ethanol) is deposited in the American Museum of Natural History, New York (catalogue number 232098).

RESULTS

Population density

In 1986, adults of *E. tulipa* were distributed along the depth gradient, however, a significant difference between the first 3 m (mean: 28.1 ± 8.2 ind/m², range: 10.4 to 48.9 ind/m²) and the last 2 m (mean: 14 ind/m², range: 3.0 to 35.5 ind/m²) was found (Kruskal–Wallis, $P < 0.001$ followed by Scheffé multiple comparisons). In 2006 and 2009, again no significant differences ($P < 0.001$) in density were found between the first 3 m. In 2006, the mean density was 52.7 ± 15.6 ind/m² (range: 22.1 to 100 ind/m²) and in 2009, the mean density was 71.6 ± 20.6 ind/m² (range: 46.2 to 121.5 ind/m²) (Figure 2). The last two deeper metres were no longer present since the bottom of the channel in this sector had been filled up with naturally deposited fine sediments. The total area covered by *E. tulipa* was at least 1000 m², but if the presence of adults was to be confirmed in the more restricted zones of the channel, the total area of colonization by this species could be at present of up to 3000 m² (2 walls of 500 m in length each with 3 m in depth).

The total area covered by substrate colonized by *D. corrodens* was 706 m² at a depth of between 0.3 and 0.6 m during high tide. The mean density was 13,000 ind/m² of which 46.2% were adults and 53.5% were recently fixed juveniles.

Gonadosomatic index

Brooding females of *E. tulipa* measured between 5.0 and 14.0 cm in total body length and between 0.4 and 1 cm in width, each female brooded between 1 and 54 egg capsules at different developmental stages (19.3 ± 13.3 , $N = 49$). The gonad represented about 47% and 44% of the total length of the female and male body respectively, while it represented about 13% of the total body volume of both sexes (Table 1). Brooding females of *D. corrodens* measured between 7.0 and 13.0 mm in total body length and between 1.2 and 1.7 mm in

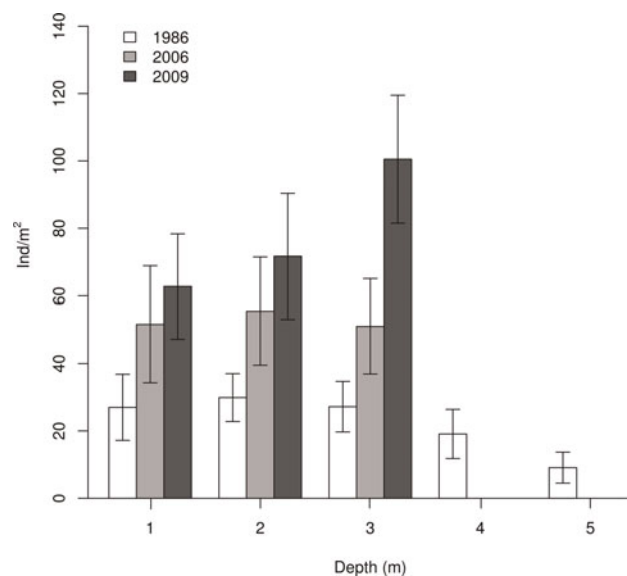


Fig. 2. Density of *Eualetes tulipa* at Plantacentro in 1986, 2006 and 2009. Bars represent mean density, vertical bars represent standard deviation.

Table 1. Body and gonad size of mature females and males of *Eualetes tulipa* (Et) and *Dendropoma corrodens* (Dc) (values represent mean \pm standard deviation, numbers in parentheses represent minimal and maximal values, and N is the number of observations).

	<i>Eualetes tulipa</i> (Females)	<i>Eualetes tulipa</i> (Males)	<i>Dendropoma corrodens</i> (Females)	<i>Dendropoma corrodens</i> (Males)
Total body length	8.58 \pm 1.87	8.22 \pm 1.63	10.1 \pm 1.5	9.0 \pm 1.9
Et (cm)	(5–14)	(5–12.5)	(7.3–13.2)	(3.8–13.5)
Dc (mm)	N = 49	N = 53	N = 25	N = 94
Body width	0.74 \pm 0.13	0.75 \pm 0.12	1.3 \pm 0.13	1.3 \pm 0.2
Et (cm)	(0.4–1.0)	(0.4–1.1)	(1.2–1.7)	(0.7–1.8)
Dc (mm)	N = 49	N = 53	N = 23	N = 93
Body volume	1.95 \pm 0.91	1.87 \pm 0.72	7.0 \pm 2.0	6.1 \pm 2.7
Et (cm ³)	(0.31–4.49)	(0.47–4.04)	(4.4–13.6)	(0.7–13.2)
Dc (mm ³)	N = 49	N = 53	N = 23	N = 93
Gonad length	4.29 \pm 1.41	3.72 \pm 1.13	4.5 \pm 1.0	4.1 \pm 1.1
Et (cm)	(1.5–7.8)	(1.4–6.0)	(2.0–6.5)	(1.5–6.5)
Dc (mm)	N = 40	N = 44	N = 22	N = 82
Gonad width	0.40 \pm 0.08	0.40 \pm 0.07	0.8 \pm 0.1	0.8 \pm 0.1
Et (cm)	(0.2–0.6)	(0.2–0.5)	(0.5–1.0)	(0.3–1.1)
Dc (mm)	N = 40	N = 44	N = 22	N = 75
Gonad volume	0.28 \pm 0.15	0.25 \pm 0.11	1.0 \pm 0.5	1.0 \pm 0.5
Et (cm ³)	(0.05–0.77)	(0.04–0.54)	(0.25–2.0)	(0.1–2.4)
Dc (mm ³)	N = 40	N = 44	N = 22	N = 75
GSI-volume (%)	13.2 \pm 6.0	12.6 \pm 5.3	15.3 \pm 6.1	17.6 \pm 5.7
	(3.7–36.6)	(2.8–24.4)	(4.6–27.8)	(7.4–34.5)
	N = 40	N = 44	N = 22	N = 75

width, each female brooded between 1 and 8 egg capsules, also at different developmental stages (3.4 ± 1.9 , $N = 25$). The gonad represented about 45% and 46% of the total length of the female and male body respectively, while it represented about 15% and 18% of the total body volume of both sexes (Table 1). Gonad volume of both females and males was correlated to total body volume for both species (Table 2). No significant differences were found in the GSI-volume throughout the year at a monthly basis (Figure 3). The gonad size is significantly related to body size for both species and sexes ($r > 0.5$ and $P < 0.01$). No significant linear correlation was observed between female size, gonad size or GSI with the number of capsules brooded per female (Pearson correlation $r < 0.2$ and $P > 0.05$ for all cases). The number of egg capsules brooded per females is very variable (Figures 4 & 5), however, a significant quadratic relationship between the log transformed number of capsules and female body volume was obtained for *E. tulipa* ($R^2 = 0.40$, $P < 0.00001$), indicating that females with a body volume of 1.25–3.25 cm³ (within a range of 6–10 cm in body length) are those that have the highest number of egg capsules (around 25 capsules/female). Females larger than 14 cm had a lower fecundity, and they brooded around 12 capsules/female (Figure 6).

Table 2. Correlations between gonad volume and total body volume in males and females of *Eualetes tulipa* and *Dendropoma corrodens*.

	Males	Females
<i>E. tulipa</i>	$r = 0.5294$ $P = 0.0002$	$r = 0.6705$ $P < 0.0001$
<i>D. corrodens</i>	$r = 0.7795$ $P < 0.0001$	$r = 0.5023$ $P = 0.0172$

Histological sections of the gonads

Gonad sections of males and females of both species showed that several gametogenesis stages are simultaneous within the same gonad (Figures 7 & 8). This pattern was observed throughout the year, showing no annual cycle with particular periods of gonad maturation or resting.

In the females of both species, germ cells, oogoniums and primary oocytes were observed simultaneously throughout the year. Yolk granules were abundant in female gonads of both species. In *E. tulipa*, the mean diameter of these granules was $8.1 \pm 2.2 \mu\text{m}$ (range: 2.5–15.0, $N = 151$) and the oocyte diameter before vitellogenesis (without the yolk granules) was $28.0 \pm 11.1 \mu\text{m}$ (range: 12.5–75 μm , $N = 64$). The yolk granules that surrounded the oocytes peripherally measured $4.4 \pm 0.9 \mu\text{m}$ (range: 2–7 μm , $N = 106$); these peripheral oocytes are also significantly smaller than the rest (Mann–Whitney U , $P < 0.001$). In *D. corrodens*, the mean diameter of yolk granules was $8.1 \pm 2.2 \mu\text{m}$ (range: 2.5–12.5, $N = 150$) and the oocyte diameter without the yolk granules was $49.3 \pm 10.6 \mu\text{m}$ (range: 25–75 μm , $N = 21$). The yolk granules that surrounded the oocytes peripherally measured $4.0 \pm 0.9 \mu\text{m}$ (range: 2–6 μm , $N = 35$); these peripheral oocytes are significantly smaller than the rest (Mann–Whitney U , $P < 0.001$). After vitellogenesis is completed, the eggs of *E. tulipa* measured $240 \pm 14 \mu\text{m}$ in diameter, while the eggs of *D. corrodens* measured $276 \pm 25 \mu\text{m}$ in diameter.

In the males of both species, the spermatogonium, primary and secondary spermatocytes, spermatids and sperm cells were also observed within one gonad and throughout the year. The diameter of the spermatogonia of *D. corrodens* varied between 4 and 6 μm (4.9 ± 0.37 , $N = 105$), and between 4 and 7 μm (4.8 ± 0.5 , $N = 91$) for *E. tulipa*.

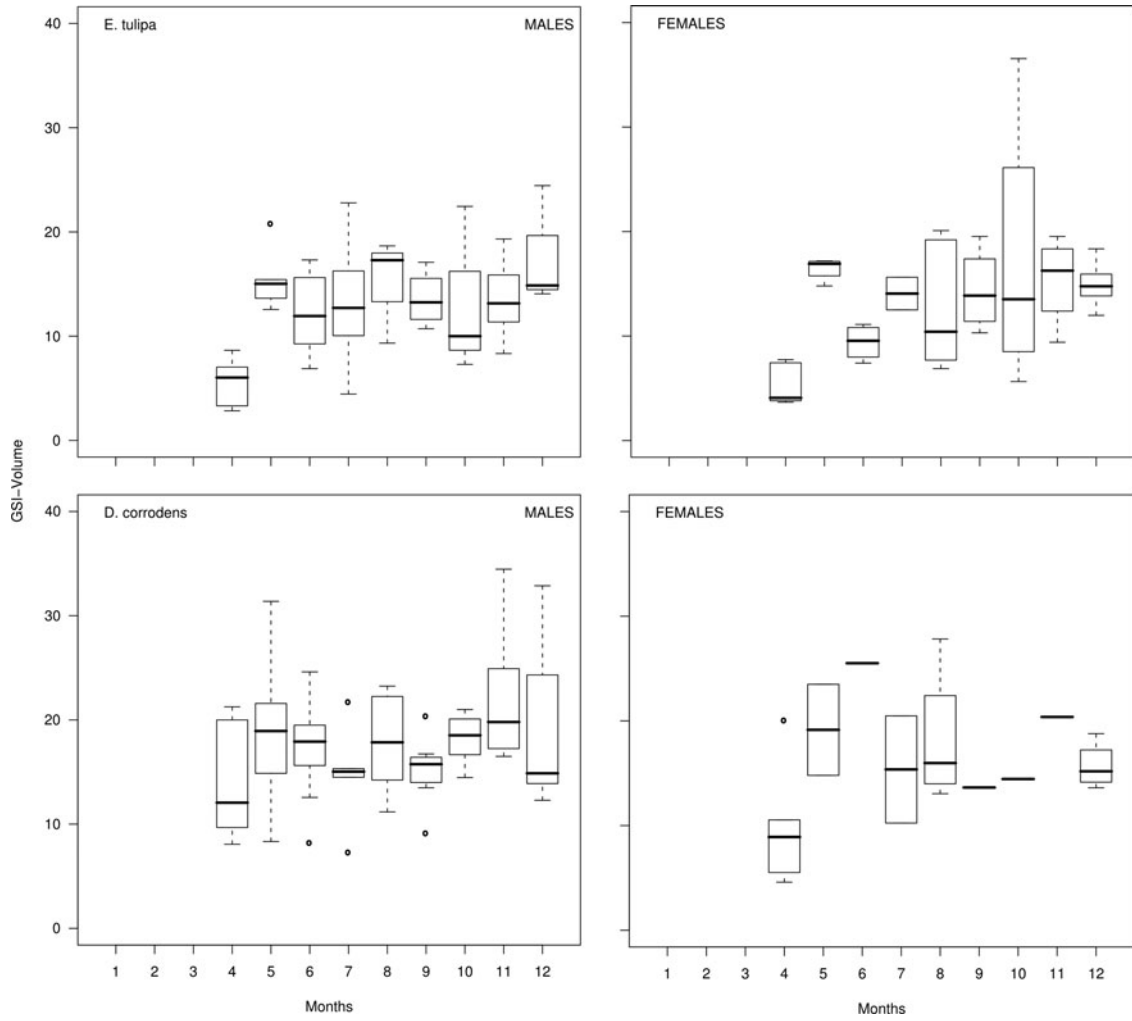


Fig. 3. Gonadosomatic index-volume for females and males of *Eualetes tulipa* and *Dendropoma corrodens* throughout the year at a monthly basis.

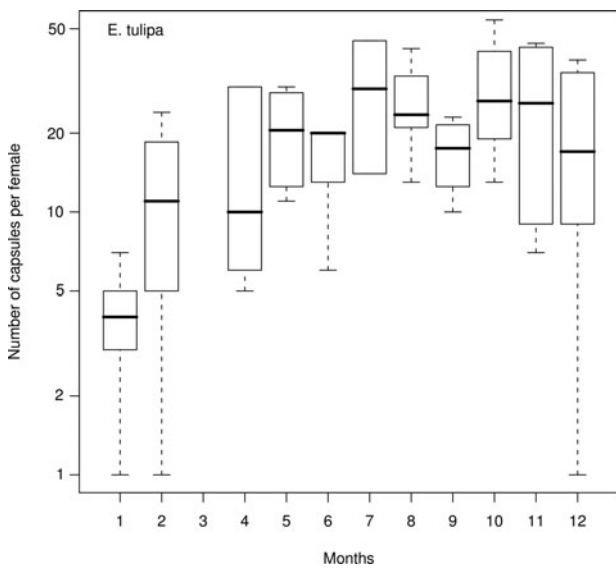


Fig. 4. Number of capsules brooded per female of *Eualetes tulipa* throughout the year at a monthly basis.

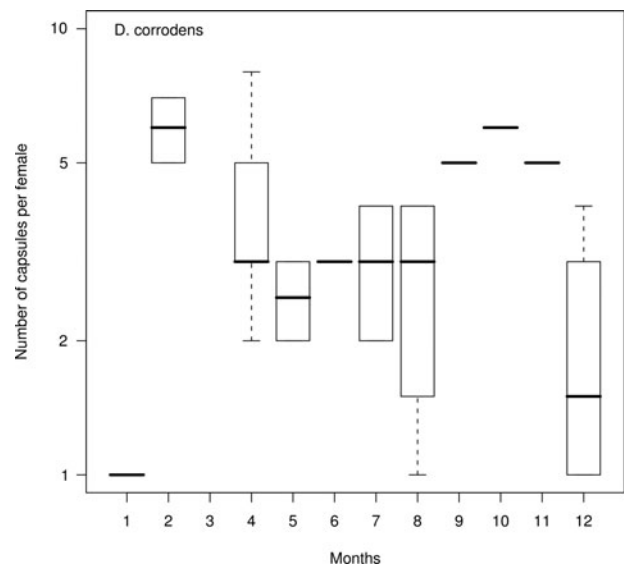


Fig. 5. Number of capsules brooded per female of *Dendropoma corrodens* throughout the year at a monthly basis.

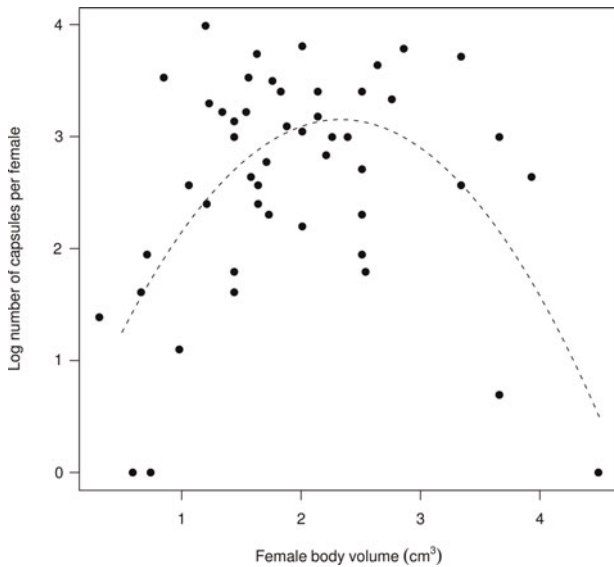


Fig. 6. Quadratic relationship between the number of capsules brooded per female (log transformed) and female size (body volume) for *E. tulipa* ($R^2 = 0.3917$, $P < 0.0001$).

DISCUSSION

The high density of individuals of both species found at the two sites is clear evidence of the reproductive success of both species. *Eualetes tulipa*, assumed to be restricted to the tropical Pacific near Panama bay, has been observed in Hawaii since 1972 where they are found mostly in harbours or embayments (Coles & Eldredge, 2002). In Venezuela, besides the population at the Planta Centro power plant

channel (built in the 1940s), two more populations have been detected, also on artificial substrates. The first is at the old El Palito refinery dock (built in the 1950s) and the second is on the remains of the vessel 'Sesostris', a ship sunk since World War II at Isla Larga. Both sites are also located on the central coast of Venezuela and have the singularity, as does the power plant channel, that the access to the public is restricted, therefore, the vermetid populations live practically undisturbed by humans. The fact that *E. tulipa* is probably an invasive species is supported by all the recent literature confirming its presence in previously uncolonized habitats (Coles *et al.*, 2002; Coles & Eldredge, 2002; Schlucker, 2003; Ray, 2005). We observed that the population in the power plant has more than duplicated in the last 20 years, which not only indicates reproductive success but also the ability to compete for space with the rest of the fouling community composed mainly by sponges, hydrozoans, barnacles, anemones, bryozoans and tunicates (Losada *et al.*, 1988). Also, an increase in the biomass of *E. tulipa*, an epifaunal suspension feeding species (it extends mucous threads to collect suspended organic matter), could be related to an increase in organic suspended matter, or to nutrient supply (Birkeland, 1977, 1987), which has been reported in several localities along the Venezuelan coast mostly due to human activities including discharges of untreated wastewater and sediments from both upstream and coastal sources (Penchaszadeh *et al.*, 2000). Chlorophyll concentration maps can be used as a proxy for water enrichment and the algorithms for chlorophyll determination are known to be affected by coloured dissolved organic matter CDOM (Odriozola *et al.*, 2007). In coastal zones, rivers usually supply large amounts of dissolved matter, which serve as a nutrient source for the ocean. In the last years, such supply of sediments and

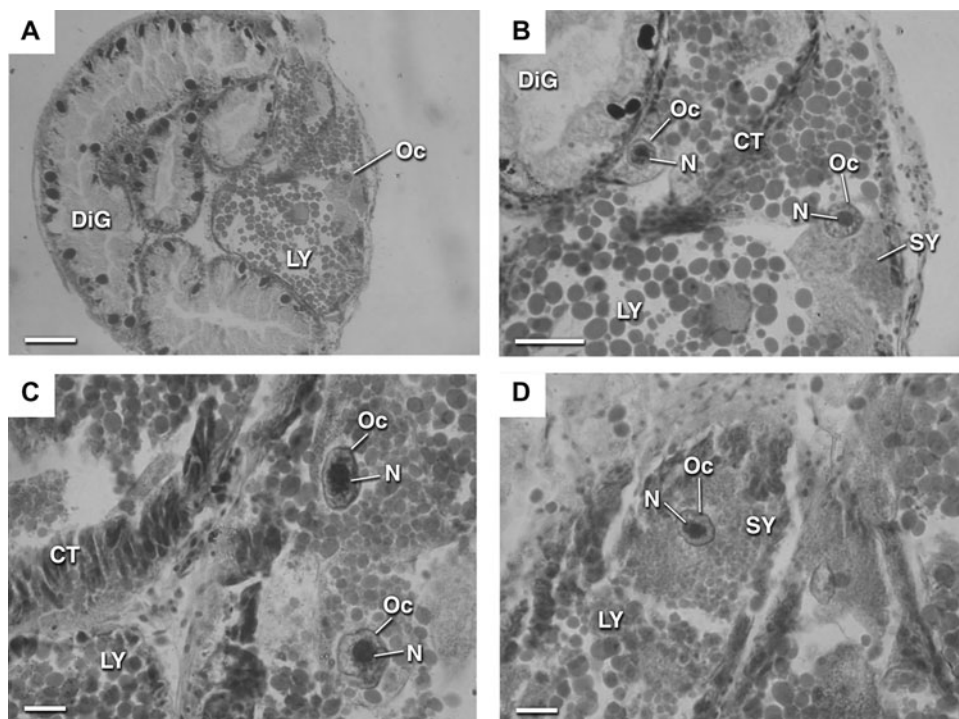


Fig. 7. Histological section of the female gonads of *Dendropoma corrodens* (A and B), and *Eualetes tulipa* (C and D). CT, connective tissue; DiG, digestive gland; LY, large yolk; N, nucleus; Oc, oocyte; SY, small yolk. Scale bars: (A) 200 μm , (B) 100 μm , (C) and (D) 30 μm .

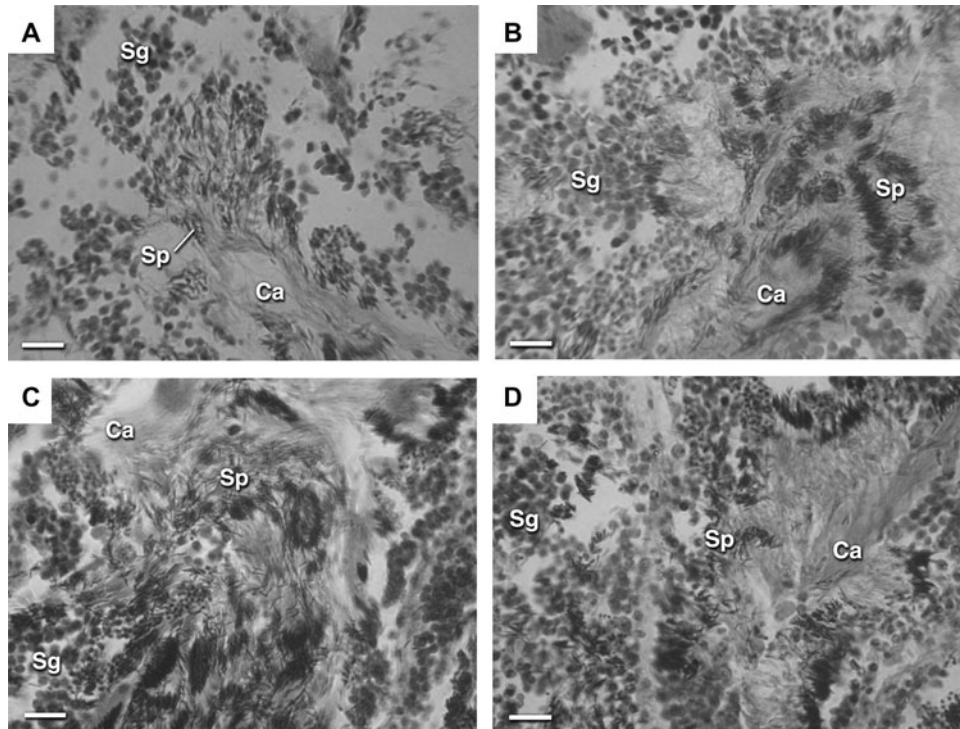


Fig. 8. Histological section of the male gonads of *Dendropoma corrodens* (A and B), and *Eualetes tulipa* (C and D). Ca, cavity after sperm shed; DiG, digestive gland; Sg, spermatogonia; Sp, spermatozoans. Scale bar: 30 μm .

dissolved elements from watersheds has increased due to deforestation, erosion, and coastal development, activities that have increased significantly in the area in the last years (Chollet & Bone, 2007). The measure over the time of both CDOM and photosynthetic pigment concentration provides a synoptic evidence of water enrichment and therefore, food availability for suspension feeders. Temporal changes in the

chlorophyll-*a* (Chl*a*) concentration for the last ten years (calculated from Sea-viewing Wide Field-of-view Sensor or SeaWiFS images; Figure 9) evidenced elevated Chl*a* concentration product of several high precipitations events (e.g. 1999, documented by Chollet & Bone, 2007), that represent high inputs of nutrients and CDOM from the Tocuyo and Aroa Rivers. However, since late 2002, the general tendency

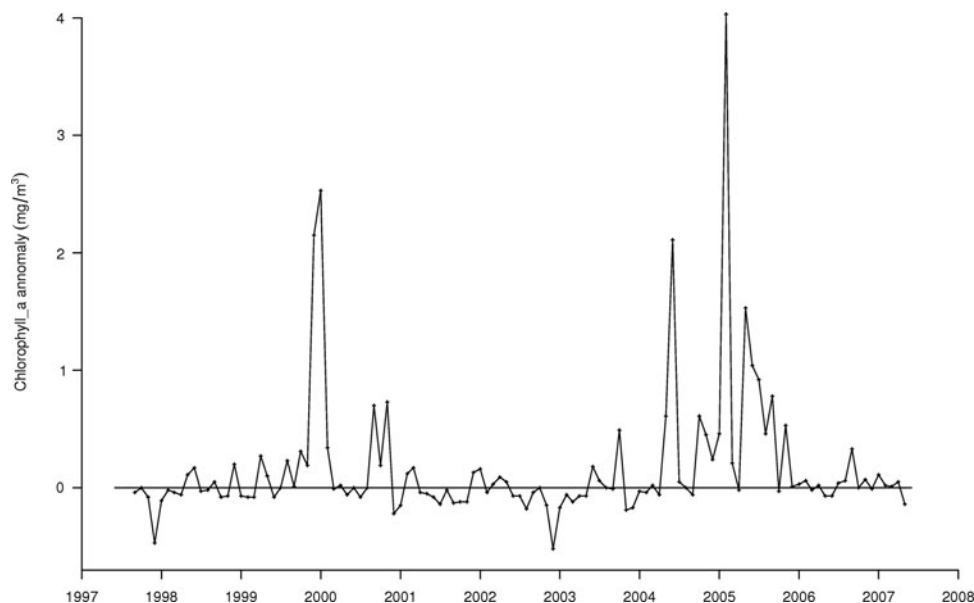


Fig. 9. Chlorophyll-*a* (Chl*a*) surface concentration anomalies calculated from Sea-viewing Wide Field-of-view Sensor (SeaWiFS) images in the region of Golfo Triste from 1997–2007. The observation window covered the Golfo Triste region, including the main river plumes (10.4–10.9N 67.8–68.5W), from where monthly average time series of Chl*a* surface concentration anomalies were extracted from SeaWiFS images. The images and data were acquired using the GES–DISC Interactive Online Visualization ANd aNalysis Infrastructure (Giovanni) as part of the NASA Goddard Earth Sciences (GES) Data and Information Services Center (DISC).

of the anomalies became positive and the variability larger, suggesting unusually high river inputs for the area. Ramírez-Llodra (2002) discussed the importance of food availability before and during gametogenesis for several invertebrate groups, mostly crustaceans, polychaetes, bivalves, and echinoderms. Since the reproductive capability of a species is determined by phylogenetic, morphological and physiological aspects, the gametogenic pattern of a species will not be altered by an increase in food supply, however, it can increase the number of eggs produced or improve their quality (Jaekle, 1995; Ramírez-Llodra, 2002).

No seasonal pattern was evident in the reproductive cycle of *E. tulipa* and *D. corrodens*, neither for males nor females. Both species reproduce throughout the year and the microscopic observation of the gonads showed that all stages of gametogenesis are simultaneous within the same animal. This gonadic activity supports the results of Miloslavich & Penchaszadeh (1992) who found females brooding egg capsules all year long. They also indicate that these species have a continuous spawning, which is confirmed by the fact that within the same female, egg capsules at different stages of development were found. Females brooding capsules with embryos at different stages of development has also been reported in *V. triquetrus*, in *D. petraeum* and in *Serpulorbis arenarius*, however, in these species, a clear seasonal reproductive time was observed, so the egg production during this season must be continuous and egg capsules were probably deposited over a long period (Calvo *et al.*, 1998; Calvo & Templado, 2004a, b). Several papers have reported that fecundity in marine gastropod species, measured as egg capsule and juvenile production, increases with female size (Ramírez-Llodra, 2002). Perron (1983) reported an age-specific fecundity increase with female size for *Conus pennaeus* Born, 1778 in Hawaii; Miloslavich & Dufresne (1994) reported for *Buccinum cyaneum* Bruguière, 1792 from the Saguenay fjord in Canada, that egg capsules deposited by larger females contained more eggs and embryos than those of smaller females; Miloslavich *et al.* (2003) reported that female size determined fecundity in terms of juvenile production for two species of *Crepidula* in the southern Caribbean, either by increasing the number of hatchlings per female at larger female sizes (*C. aphysioides* Reeve, 1859) or by producing larger capsules with more hatchlings (*C. navicula* Morch, 1877). Harding *et al.* (2007) reported for the invasive *Rapana venosa* Valenciennes, 1846 that whelk fecundity increased with female size (from 123 embryos/capsule in females measuring 45 mm to 3673 embryos/capsule in females measuring 163 mm). Such difference in the number of eggs per capsule was due to the fact that larger females produced much larger egg capsules (7 mm and 30–33 mm in capsule height for small and large whelks respectively). An interesting fact of our results was that females brood more capsules as they get larger, but seem to reach an optimal reproductive size, in which the number of brooded capsules is maximal, and then, at larger sizes, fecundity decreases. Ramírez-Llodra (2002) reviewed the relationship between fecundity and female size and age in decapod crustaceans, polychaetes, bivalves, and in some colonial groups such as corals. In bivalves, the growth rate increases in the first years and then declines, while the production of gametes increases throughout their lifetime. As a certain shell size is attained, the energy is allocated from growth to reproduction, being the inner volume of the shell the limit

for gamete production. Since vermetids brood the egg capsules either in the mantle cavity or attached to the internal walls of the shells, it can be expected that, as in bivalves, female and shell size will constrain the number of egg capsules that may be brooded at the same time. Despite that a decline in the reproductive capacity of older individuals is expected in any population, this has not been clearly reported in the literature for gastropods. Fertility reduction in mature and/or larger individuals has been reported in populations affected either by imposex due to tributyltin (TBT) pollution (Oehlman *et al.*, 1996; Ramón & Amor, 2002), or to parasitic castration due to gonadal infection mostly by trematodes (Tetrault *et al.*, 2000). In our study we did not determine the relationship between female size and offspring size. Following the offspring definition of Marshall & Keough (2008), for *E. tulipa* and *D. corrodens*, offspring size would be equivalent to hatching size as this is the stage in which the 'propagule' becomes independent from maternal nutrition. These authors state that within a range of species, larger mothers produce larger offspring, however, they also discuss that this might not reflect reality as the percentage of species where no relationships has been observed is probably underestimated, and many species have not been studied at all. In Calyptreids for instance, Chaparro *et al.* (1999) reported for *Crepidula dilatata* Lamarck, 1822, a positive correlation between female size and offspring size, while Miloslavich *et al.* (2003) reported no correlation between these same parameters for *C. aphysioides* and *C. navicula*, indicating that even at the family level, no generalizations can be made.

The histological study of the gonads of *E. tulipa* and *D. corrodens* confirm that both species are dioecious as reported by Miloslavich & Penchaszadeh (1992), since we never found specimens with male and female gonads simultaneously. Heller (1993) reviewed that about 99% of the prosobranch gastropods are dioecious, however, Calvo & Templado (2004b) presented evidence that *Serpulorbis arenaria*, a Mediterranean vermetid, is a sequential hermaphrodite with alternating sexuality. This is evidenced by a clearly male biased ratio (small individuals were males, while the largest were either males or females), and to the presence of a few males with a pallial slit and egg capsules, a character linked to females, indicating that they had been females before. As mentioned earlier, studies in gastropod gametogenesis and particularly in vermetids are very scarce, so reports of hermaphroditic species, either simultaneous or protandric could be very underestimated.

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REFERENCES

- Bandel K.** (1975) Das Embryonalgehäuse karibischer Meso- und Neogastropoden (Mollusca). *Akademie der Wissenschaften und der Literatur, Mainz* 1, 1–133.
- Bandel K.** (1976) Observations on spawn, embryonic development and ecology of some Caribbean lower mesogastropoda. *The Veliger* 18, 249–271.
- Barroso C.M. and Moreira M.H.** (1998) Reproductive cycle of *Nassarius reticulatus* in the Ria de Aveiro, Portugal: implications for imposex studies. *Journal of the Marine Biological Association of the United Kingdom* 78, 1233–1246.
- Bieler R.** (1996) Mörch's worm-snail taxa (Caenogastropoda: Vermetidae, Siliquariidae, Turritellidae). *American Malacological Bulletin* 13, 23–35.
- Birkeland C.** (1977) The importance of the rate of biomass accumulation in early successional stages of benthic communities to the survival of coral recruits. In Taylor D.L. (ed.) *Proceedings of the Third International Coral Reef Symposium, Miami, Florida, Volume 1: Biology*. Rosenstiel School of Marine and Atmospheric Research, pp. 15–21.
- Birkeland C.** (1987) Nutrient availability as a major determinant of differences among coastal hard-substratum communities in different regions of the tropics. In Birkeland C. (ed.) *Comparison between Atlantic and Pacific tropical marine coastal ecosystems: community structure, ecological processes, and productivity*. UNESCO Reports in Marine Science, 46, pp. 43–98.
- Calvo M.** (1999) *Biología reproductora de tres especies de la familia Vermetidae (mollusca: Gastropoda) de las costas mediterráneas de la Península Ibérica*. PhD thesis. Universidad Autónoma de Madrid, Madrid, Spain.
- Calvo M., Templado J. and Penchaszadeh P.E.** (1998) Reproductive biology of the gregarious Mediterranean vermetid gastropod *Dendropoma petraeum*. *Journal of the Marine Biological Association of the United Kingdom* 78, 525–549.
- Calvo M. and Templado J.** (2004a) Reproduction and development in a vermetid gastropod, *Vermetus triquetrus*. *Invertebrate Biology* 123, 289–303.
- Calvo M. and Templado J.** (2004b) Reproduction and sex reversal of the solitary vermetid gastropod *Serpulorbis arenarius*. *Marine Biology* 146, 963–973.
- Calvo M., Templado J., Oliverio M. and Machordom A.** (2009) Hidden Mediterranean biodiversity: molecular evidence for a cryptic species complex within the reef building vermetid gastropod *Dendropoma petraeum* (Mollusca: Caenogastropoda). *Biological Journal of the Linnean Society* 96, 898–912.
- Chaparro O.R., Oyarzun R.F., Vergara A.M. and Thompson R.J.** (1999) Energy investment in nurse eggs and egg capsules in *Crepidula dilatata* Lamarck (Gastropoda, Calyptraeidae) and its influence on the hatching size of the juvenile. *Journal of Experimental Marine Biology and Ecology* 232: 261–274.
- Chenu J.C.** (1842–1853) *Illustrations conchyliologiques ou description et figures de toutes les coquilles connues vivantes et fossiles, classées suivant le système de Lamarck modifié d'après les progrès de la science et comprenant les genres nouveaux et les espèces récemment découvertes*. Paris: A. Franck.
- Chollet I. and Bone D.** (2007) Effects of heavy rainfall on polychaetes: differential spatial patterns generated by a large-scale disturbance. *Journal of Experimental Marine Biology and Ecology* 340, 113–125.
- Coles S.L. and Eldredge L.G.** (2002) Nonindigenous species introductions on coral reefs: A need for information. *Pacific Science* 56, 191–209.
- Coles S.L., DeFelice R.C. and Eldredge L.G.** (2002) Nonindigenous marine species at Waikiki and Hawaii Kai, Ōahu, Hawaii. *Final Report prepared for the David and Lucile Packard Foundation and the State of Hawaii Department of Land and Natural Resources, Division of Aquatic Resources, Honolulu*. Bishop Museum Technical Report No. 25, 245 pp.
- D'Orbigny A.** (1841) *Mollusques—histoire physique, Politique et Naturelle de l'île de Cuba* 1. Paris: Arthus Bertrand, pp. 1–240.
- Díaz J.M. and Puyana M.** (1994) *Moluscos del Caribe Colombiano. Un catálogo ilustrado*. 1st edition. Colombia: Colciencias-Fundación Natura–INVEMAR.
- Gendron L.** (1992) Determination of the size at sexual maturity of the waved whelk *Buccinum undatum* Linnaeus, 1758, in the Gulf of St. Lawrence, as a basis for the establishment of a minimum catchable size. *Journal of Shellfish Research* 11, 1–7.
- Giménez J. and Penchaszadeh P.E.** (2002) Reproductive cycle of *Zidona dufresnei* (Caenogastropoda: Volutidae) from the southwestern Atlantic Ocean. *Marine Biology* 140, 755–761.
- Giménez J. and Penchaszadeh P.E.** (2003) Size at first sexual maturity in *Zidona dufresnei* (Caenogastropoda: Volutidae) of the south-western Atlantic Ocean (Mar del Plata, Argentina). *Journal of the Marine Biological Association of the United Kingdom* 83, 293–296.
- Gould S.J.** (1994) *Petalococonchus sculpturatus alaminatus*, a new Pliocene subspecies of Vermetid gastropods lacking its defining generic character, with comments on Vermetid systematics in general. *Journal of Paleontology* 68, 1025–1036.
- Hadfield M.G., Kay E.A., Gillete M.U. and Lloyd M.C.** (1972) The Vermetidae (Mollusca: Gastropoda) of the Hawaiian Islands. *International Journal on Life in Oceans and Coastal Waters* 12, 81–98.
- Harding J.M., Mann R. and Kiduff C.W.** (2007) The effects of female size on fecundity in a large marine gastropod *Rapana venosa* (Muricidae). *Journal of Shellfish Research* 26, 33–42.
- Heller J.** (1993) Hermaphroditism in molluscs. *Biological Journal of the Linnean Society* 48, 19–42.
- Himmelman J.H. and Hamel J.F.** (1993) Diet, behaviour and reproduction of the whelk *Buccinum undatum* in the northern Gulf of St. Lawrence, eastern Canada. *Marine Biology* 116, 423–430.
- Hodgson A.N. and Eckelbarger K.** (2000) Ultrastructure of the ovary and oogenesis in six species of patellid limpets (Gastropoda: Patellogastropoda) from South Africa. *Invertebrate Biology* 119, 265–277.
- Hughes R.N.** (1979) Notes on the reproductive strategies of the South African vermetid gastropods *Dendropoma corallinaceum* and *Serpulorbis natalensis*. *The Veliger* 21, 423–427.
- Hughes R.N. and Lewis A.H.** (1974) On the distribution, feeding and reproduction of the vermetid gastropod *Dendropoma maxima*. *Journal of Zoology*, London 172, 531–547.
- Jaekle W.B.** (1995) Variation in the size, energy content, and biochemical composition of invertebrate eggs: correlates to the mode of larval development. In McEdward L. (ed.) *Ecology of marine invertebrate larvae*. CRC Marine Science Series 6, pp. 49–77.
- Keen A.M.** (1961) A proposed reclassification of the gastropod family Vermetidae. *Bulletin of the British Museum (Natural History) (Zoology)* 7, 183–214, pls. 54–55.

- Keen M.** (1971) *Sea-shells of tropical west America (marine mollusks from Baja California to Peru)*. 2nd edition. Stanford: Stanford University Press.
- Lee J.H.** (1999) Gametogenesis and reproductive cycle of the rock shell, *Reishia (Thais) clavigera* (Neogastropoda: Muricidae), on the west coast of Korea. *Korean Journal of Biological Sciences* 3, 375–383.
- Losada F., Martín A., Feragotto W. and Alamo C.** (1988) Interacciones biológicas en el canal de toma de la planta termoeléctrica del centro en Punta Morón, Venezuela. *Ecotrópicos* 1, 55–70.
- Marshall D.J. and Keough M.J.** (2008) The evolutionary ecology of offspring size in marine invertebrates. *Advances in Marine Biology* 53, 1–60.
- Martel A., Larrivé D.H., Klein K.R. and Himmelman J.H.** (1986a) Reproductive cycle and seasonal feeding activity of the neogastropod *Buccinum undatum*. *Marine Biology* 74, 73–78.
- Martel A., Larrivé D.H., Klein K.R. and Himmelman J.H.** (1986b) Behaviour and timing of copulation and egg-laying in the neogastropod *Buccinum undatum* L. *Journal of Experimental Marine Biology and Ecology* 96, 27–42.
- Miloslavich P.** (1987) *Biología y estrategias reproductivas de Vermetus sp. y Dendropoma corrodens, dos especies pertenecientes a la familia Vermetidae (Gasterópodos, Prosobranquios)*. Licenciatura thesis. Universidad Simón Bolívar, Caracas, Venezuela.
- Miloslavich P. and Dufresne L.** (1994) Development and effect of female size on egg and juvenile production in the neogastropod *Buccinum cyaneum* from the Saguenay fjord. *Canadian Journal of Fisheries and Aquatic Sciences* 51, 2866–2872.
- Miloslavich P. and Penchaszadeh P.E.** (1992) Reproductive biology of *Vermetus* sp. and *Dendropoma corrodens* (Orbigny, 1842): two Venezuelan gastropods from the Southern Caribbean. *The Veliger* 35, 78–88.
- Miloslavich P., Penchaszadeh P.E. and Klein E.** (2003) Reproduction of *Crepidula navicula* Morch, 1877 and *Crepidula aphysioides* Reeve, 1859 (Caenogastropoda) from Morrocoy and La Restinga Lagoon, Venezuela. *The Nautilus* 117, 121–134.
- Morton J.E.** (1951) The structure and adaptations of the New Zealand Vermetidae. Part I. The genus *Serpulorbis*. *Transactions of the Royal Society of New Zealand* 79, 1–19.
- Morton J.E.** (1965) Form and function in the evolution of the Vermetidae. *Bulletin of the British Museum (Natural History)* 11, 585–630.
- Odriozola A.L., Varela R., Hu C., Astor Y., Lorenzoni L. and Müller-Karger F.E.** (2007) On the absorption of light in the Orinoco River plume. *Continental Shelf Research* 27, 1447–1464.
- Oehlmann J., Fioroni P., Stroben E. and Market B.** (1996) Tributyltin (TBT) effects on *Ocenebra aciculata* (Gastropoda: Muricidae): imposex development, sterilization, sex change and population decline. *The Science of the Total Environment* 188, 205–223.
- Oliver A.P.H.** (1975) *Hamlyn guide to shells of the world*. London: Hamlyn.
- Penchaszadeh P., León C.A., Alvarez H., Bone D., Castellano P., Castillo M.M., Díaz Y., García M.P., Lemus M., Losada F., Martín A., Miloslavich P., Paredes C., Pérez D., Sebastiani M., Steccoli D., Roa V. and Villamizar A.** (2000) The coastline of Venezuela. In Sheppard C. (ed.) *Seas at the millennium. An environmental evaluation*. University of Warwick, UK, Volume I: Europe, The Americas and West Africa, pp. 643–661.
- Perron F.** (1983) Growth, fecundity and mortality of *Conus pennaceus* in Hawaii. *Ecology* 64, 53–62.
- Ray G.L.** (2005) *Invasive marine and estuarine animals of Hawaii and other Pacific Islands*. Aquatic Nuisance Species Research Program, ERDC/TN ANSRP-05-3, 19 pp.
- Ramírez-Llodra E.** (2002) Fecundity and life-history strategies in marine invertebrates. *Advances in Marine Biology* 43, 87–170.
- Ramón M. and Amor M.J.** (2002) Reproductive cycle of *Bolinus brandaris* and penis and genital duct size variations in a population affected by imposex. *Journal of the Marine Biological Association of the United Kingdom* 82, 435–442.
- Schiaparelli S., Albertelli G. and Cattaneo-Vietti R.** (2006) Phenotypic plasticity of Vermetidae suspension feeding: a potential bias in their use as biological sea-level indicators. *Marine Ecology* 27, 44–53.
- Schlucker A.** (2003) *State of Hawaii Aquatic Invasive Species Management Plan*. The Department of Land and Natural Resources, Division of Aquatic Resources, The Nature Conservancy of Hawaii, 205 pp.
- Strathmann M.F. and Strathmann R.R.** (2006) A Vermetid gastropod with complex intracapsular cannibalism of nurse eggs and sibling larvae and a high potential for invasion. *Pacific Science* 60, 97–108.
- Tello J.** (1975) *Catálogo de la fauna venezolana VIII. Mollusca*. Caracas, Venezuela: Arte.
- Tetrault F., Himmelman J.H. and Measures L.** (2000) Impact of a castrating trematode, *Neophasis* sp., on the common whelk *Buccinum undatum* in the northern Gulf of St. Lawrence. *Biological Bulletin. Marine Biological Laboratory, Woods Hole* 198, 261–271.

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Vega A.J. and González A. (2002) Moluscos del Pacífico Veraguense. *Tecnociencia* 4, 1–45.

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