TAXONOMIC AND ECOLOGICAL NOTES ON SOME *DIDEMNUM* SPECIES (ASCIDIACEA, DIDEMNIDAE) FROM SÃO SEBASTIÃO CHANNEL, SOUTH-EASTERN BRAZIL

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(With 5 figures and 1 plate)

RESUMO

Notas Taxonômicas e Ecológicas sobre Algumas Espécies de *Didemnum* (Ascidiacea, Didemnidae) do Canal de São Sebastião, Sudeste Brasileiro

Foram estudadas nove espécies coletadas em águas rasas do Canal de São Sebastião (Sudeste brasileiro). Uma das espécies coletadas é endêmica, duas têm uma distribuição geográfica restrita ao Atlântico ocidental e as espécies restantes podem ser encontradas tanto em águas do Atlântico como do Pacífico. *Didemnum ahu, D. granulatum, D. ligulum, D. lutarium* e *D. perlucidum* constituem os primeiros registros para o Brasil.

Palavras-chave: sistemática, Ascidiacea, *Didemnum*, Sudeste brasileiro.

ABSTRACT

Nine species collected in very shallow waters at the São Sebastião Channel (South-Eastern Brazil) were studied. Among the species collected, one is an endemic species, two have a geographical distribution that is restricted to the West Atlantic, and the remaining species are found in both Atlantic and Pacific waters. *Didemnum ahu, D. granulatum, D. ligulum, D. lutarium*, and *D. perlucidum* are recorded from Brazil for the first time.

Key words: sistematics, Ascidiacea, *Didemnum*, South-Eastern Brazil

INTRODUCTION

Ecological studies of the encrusting community underside intertidal boulders, at the São Sebastião Channel, South-Eastern Brazil, resulted in a regular sampling of ascidians.

Most of the ascidians collected were previously known from Brazil, except for those belonging to the family Didemnidae, which proved to be the most diversified in the intertidal area. Most species of this family belonged to the genus *Didemnum*.

The nine species referred herein were common components of the communities, five of them

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are new records for Brazil. Only brief descriptions are given for *D. psammathodes, D. speciosum* and *D. rodriguesi* since they were recently described in previous papers (Rodrigues and Rocha, 1993; Rocha and Monniot, 1993).

It is very difficult to distinguish between the species of *Didemnum* so it was necessary to compare our specimens with the type species. Several types are in the Muséum National d'Histoire Naturelle of Paris, others in the American Museum of Natural History and Amsterdam Museum. Vouchers of the material examined have been deposited in the Muséum National d'Histoire Naturelle, Paris (MNHN), and in the Museu de Zoologia da Universidade de São Paulo, São Paulo, Brazil (MZUSP).

STUDY SITE

Boulder fields in the São Sebastião Channel form narrow strips, 3 to 10m wide, between the rocky walls of the coast and the sandy substrate toward the sea, in the intertidal zone. A map of the region was presented elsewhere (Rodrigues and Rocha, 1993).

Ascidians are rarely found on small pebbles of 30-50 cm^2 in underside surface, but they are very common underside of boulders over 150 cm^2 large. These boulders lie on the sandy substrate on the other layers of boulders.

The boulders are not very exposed to the waves, but they are subject to displacement three to five times a year during strong storms, mainly in the winter. Surface water temperature in the region ranges from 20°C (August and September) to 28°C (February and March).

METHODS

Most of the specimens were collected during low tide, by scraping off the colonies from the boulders with a razor blade. They were then fixed in formalin 4%. Only emerged boulders were sampled. Some specimens were collected from submerged artificial substrate hanging between 0.5 and 1.0m deep.

The techniques of staining and mounting the material on slides were described by Monniot and Monniot (1972).

The figures were prepared under a drawing tube. Small pieces of the tunic were boiled in hypocloride in order to obtain spicules, which were photographed under scanning electronic microscope.

Didemnum ahu Monniot and Monniot, 1987
(Fig. 1A, B, C – Pl. IA)

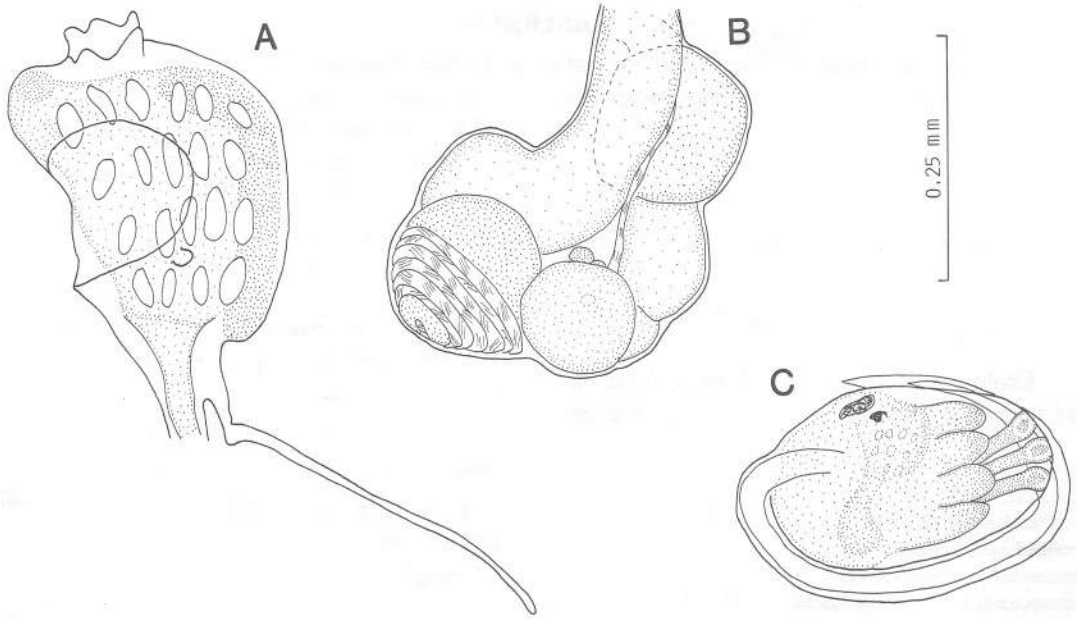

Fig. 1 — *Didemnum ahu* Monniot and Monniot, 1987: A, thorax; B, abdomen; C, larva.

Didemnum ahu: Monniot and Monniot, 1987: 25, Polynesia.

Localities: Ponta do Baleeiro, Praia Grande – intertidal, Praia do Cabelo Gordo de Dentro – shallow water. Material in MNHN: A2 DID C 183.

The thin colonies (1 mm thick) form small crusts of 2 cm in diameter at most. They encrust both natural rocky substrata and submerged metal or ceramic surfaces. They are yellowish or beige. The spicules are not abundant and are restricted to the outermost layer of the tunic which has a soft consistency. There are two types of spicules (Pl. 1A): those with a variable number of rays, which have conical tapered ends and those that have a large number of rays which are cylindrical with blunt ends. The size of the spicules is also variable: from 8 to 42 μm and 64 μm in the same colony.

The zooids are easily removed from the tunic. They are lesser than 1 mm long. The oral siphon is short and the cloacal siphon is usually small in the contracted thorax but it can be rather wide exposing half of the branchial sac. There are six stigmata per half row in the first one, grading down to four in the last row (Fig. 1A). The thoracic lateral organs are small, ear-like, placed over the third transversal vessel or over the fourth row of stigmata. The retractor muscle process is long when the thorax is not contracted and is located between the endostyle and the oesophageal pedicle (Fig. 1A).

The digestive tube is large, with no distinctive feature (Fig. 1B). The single testis is surrounded by six to eight turns of the sperm duct. The ovary has many oocytes, one of them larger than the others.

The larvae are small, between 0.3 and 0.4mm long. They have three adhesive papillae with long peduncles placed in close proximity which gives the larvae an elongated appearance. In two of the colonies we found one larva with only two adhesive papillae. The larvae have four pairs of ampullae. They are not gemmiparous (Fig. 1C).

Didemnum ahu was first found in Polynesia and is recorded in New Caledonia (unpublished). This species is closely related to *D. conchyliatum* (Sluiter, 1898) redescribed by F. Monniot, 1983 based on the type specimen (ZMA TU 578) and caribbean samples. *D. conchyliatum* zooids have a larger oral aperture with pointed lobes, a long thorax, the retractor muscle process becoming free from the oesophageal pedicle very posteriorly; intestinal loop is wider and the larvae contain pigment cells in their body wall.

The specimens described here have more characters in common with *D. ahu* from the Pacific Ocean than with *D. conchyliatum*. Because there are other *Didemnum* species common to São Sebastião Channel and the West Pacific area, these Brazilian specimens are attributed to *Didemnum ahu*. It is worth mentioning that most of those specimens came from the intertidal zone where animals are subjected to drying stress. Thus the slightly different morphology can be attributed to this environmental feature. Deeper collections would be necessary to ascertain the identification.

Didemnum granulatum Tokioka, 1954
(Fig. 2A, B, C – Pl. IB)

Didemnum moseleyi f. *granulatum*: Tokioka, 1954: 244, Japan

Didemnum granulatum: Kott, 1981: 167 (see for synonymy), Fiji; Kott and Goodbody, 1980: 517, Hong Kong; Monniot and Monniot, 1987: 31, Polynesia; Nishikawa, 1990: 103, Japan.

Localities: Praia Grande, Ponta do Jaroba – intertidal. Material in MNHN A2 DID C 180.

The colonies are 1-2 mm thick and 10-15 cm long. They encrust the lateral surfaces of large boulders, growing over algae and other encrusting animals. Their brilliant colors of orange, salmon or brown make them conspicuous. Under magnification we can see that the surface of the colony is not smooth but it contains numerous short spicule-filled papillae that make the colonies distinct. The spicules (Pl. IB) are stellate and of variable sizes (11-34 μm). Some of them have thinner rays than the others. The spicules are densely distributed in the test and they form a thick basal layer.

The apertures of the oral siphons are stellate and sometimes contracted into the surface of the colony. The common cloaca are simple holes. The tunic presents quite extensive thoracic lacunae, these cavities also extend into the abdominal layer in some colonies.

The small zooids (less than 1 mm) are orange. The oral siphon is short and wide. The cloacal aperture is wide, exposing most of the branchial sac (Fig. 2A). It has no languet. The thoracic lat-

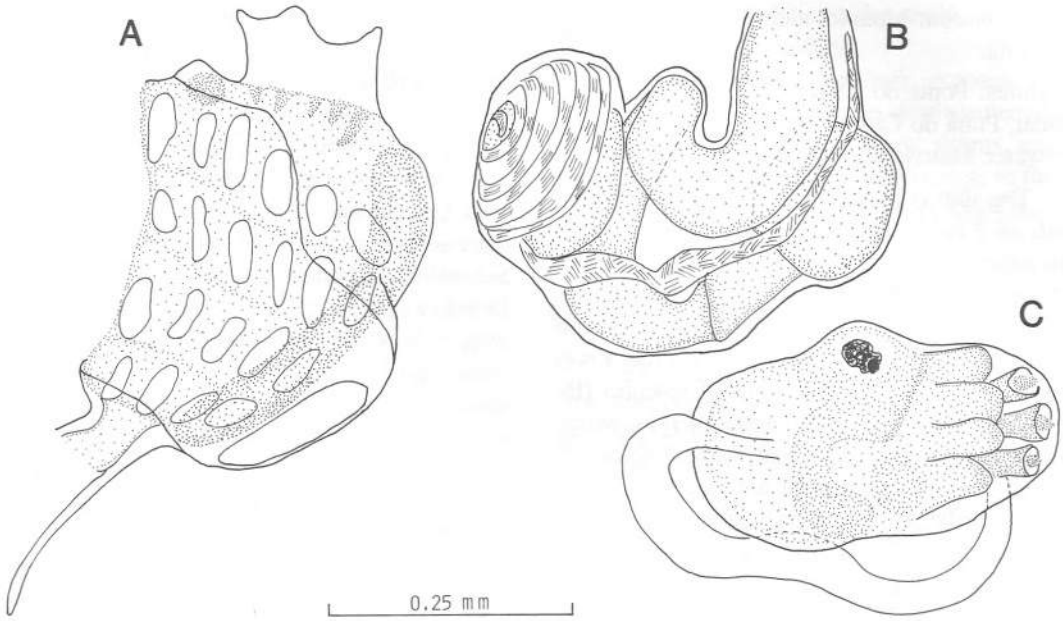

Fig. 2 — *Didemnum granulatum* Tokioka, 1954: A, thorax; B, abdomen; C, larva.

eral organs are either oval or long and vertical and are located in the posterior region of the body wall at the level of the third and fourth rows of stigmata (Fig. 2A). The triangular shape of the branchial sac is due to the fact that there are six stigmata in the first row and only four in the fourth row. Thus the endostyle and oesophageal pedicle are close without space between them. The retractor muscle process is medium to long and it is attached to the middle of the oesophageal pedicle.

The abdomen which is smaller than the thorax is usually located horizontally (Fig. 2B). The testis is a single body surrounded by six to seven coils of the sperm duct. The ovary is placed between the testis and the stomach, but closer to the former, and consists of a large oocyte. The larvae are 0.4 mm long and have the usual three adhesive papillae and four pairs of ampullae. They are generally oval in shape.

The specimens described in this paper are similar to those from Polynesia (Monniot and Monniot, 1987) and others collected in New Caledonia. The difference is the shape and size of the thoracic lateral organs which were larger in the zooids that we observed. The larvae described by Tokioka (1954) and Monniot and Monniot (1987) are smaller but the ones here observed were not as large as those described by Kott (1981).

This is the first time that this species is recorded in the Atlantic Ocean. Its known distribution was limited to the Western Pacific Ocean: Fiji, Suva Barrier Reef, Great Astrolabe Reef, Palau Is., Japan Sea, Hawaii, Circum Australia, and Polynesia.

Didemnum ligulum Monniot F., 1983
(Fig. 3A, B, C, D – Pl. IC, D)

Didemnum ligulum Monniot F., 1983: 27, Guadeloupe; Monniot and Monniot, 1987: 34, Polynesia.

Localities: Ponta do Baleeiro, Ponta do Jaroba – intertidal, Praia do Cabelo Gordo de Dentro – shallow water. Material in MZUSP 11.192.

The colonies are thin, usually 1-2 mm thick. The largest colony is 4 cm long. They are orange or yellowish in color. The spicules are dense but common cloacal openings can be seen as well as the star-shaped apertures of the oral siphons. In the preserved specimens, the oral siphons are often higher than the surface of the colony. The spicules are almost rounded with short rays around a central mass or composed of densely packed blunt rays; their sizes are variable, usually between 10 and 50 µm (Pl. IC, D). The extensive cloacal canals divide the tunic into a superficial and a basal layers.

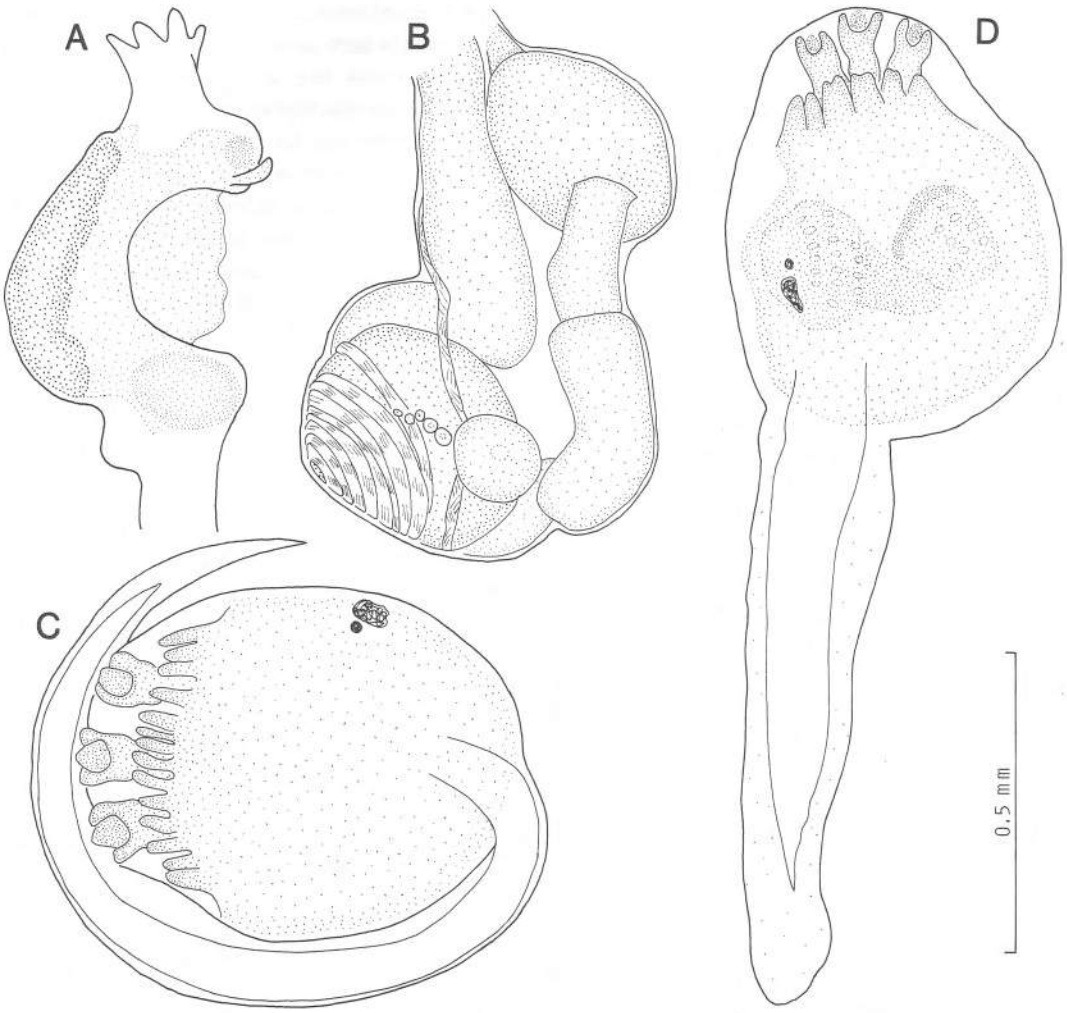

Fig. 3 — *Didemnum ligulum* Monniot F., 1983: A, thorax; B, abdomen; C and D, larvae.

The zooids are orange, and up to 1.5 mm in length. The oral siphon forms a tube with six long lobes. The cloacal opening is wide, and it is provided with a small thin dorsal languet (Fig. 3A). The long retractor muscle process is attached to the anterior end of the oesophageal pedicle.

The testis is a single large body, partially surrounded by the seven to nine coils of the sperm duct. The ovary lies beside the testis and it consists of one large and many small oocytes (Fig. 3B).

The larvae (Fig. 3C, D) are 0.7-0.75 mm long (trunk including the adhesive papillae) with a wide short tail. They are well pigmented. They have three short adhesive papillae and usually ten or more pairs of ampullae. If less than ten pairs were observed, the ampullae were in the process of division. They are gemmiparous.

When compared to the specimens of Guadeloupe (Monniot F., 1983) our colonies have smaller spicules, the languets are not so developed and bifurcated and there is a greater number of sperm duct coils. The size of the larvae is between that of the larvae from Guadeloupe and Polynesia (Monniot and Monniot, 1987). Besides that, the specimen described in this paper is similar to the former descriptions, but the species seems to be very polymorphic.

The presence of this species along the Brazilian coast is not surprising, since it was described from the Caribben Islands. Its presence in

the Western Pacific indicates that it may also be distributed world wide in warm shallow waters.

Didemnum lutarium Van Name, 1910
(Fig. 4A, B, C – Pl. IE)

Didemnum lutarium Van Name, 1910: 371.

Didemnum candidum lutarium: Van Name, 1921: 323; 1945: 86, West Atlantic (and synonymy).

Localities: Ponta do Baleeiro, Ponta do Jaroba – intertidal. Material in MNHN A2 DID C 181.

This species forms small crusts 2 mm thick underside the boulders. It is white, slightly beige or pink when the zooids are exposed through the oral siphons.

Except for the thin outmost layer of tunic that has no spicules, the colony is completely filled with small rounded spicules, with short and blunt rays (Pl. 1E). The spicules are slightly more than 25 µm in diameter.

The zooids are easily removed from the tunic. They may be up to 1.3 mm long. The oral siphon is curved toward the dorsal side. The cloacal siphon forms a small rounded aperture at the level of the second row of stigmata (Fig. 4A); sometimes, when the thorax is very contracted, it forms a short tube. The thoracic lateral organs are posterior to the cloacal siphon (Fig. 4A). There is a strong dorsal muscle band along the dorsal side of the thorax which continues into the long retractor muscle process, that anchors in the tunic when it emerges from the middle of the oesophageal pedicle. The branchial sac has four rows of stigmata with up to nine stigmata per half row.

The stomach is round or oval. The posterior portion of the intestine contains some pyloric vesicles (Fig. 4B). Both sexes can be found in the same individual. The testis is divided into two lobes which are completely encircled by the six to nine large coils of the sperm duct. The ovary has a large oocyte but smaller ones can be seen developing at the same time.

The larva (Fig. 4C) is approximately 0.6 mm long and has three adhesive papillae and four pairs of ampullae. The oozooid has three rows of stigmata with four to six stigmata per half row. The tail makes three fourths the coil around the larva.

Van Name (1910), described *Didemnum lutarium*, however, in later works (1921, 1945), he decided to consider it a sub-species of *D. candidum*. This last species is frequently confused with many other "white didemnids". The characteristics as described by Lafargue (1974) for the neotype of *Didemnum candidum* Savigny, 1816 are somewhat different from those of the West Atlantic species. We dissected many colonies and we found many characters that fully agree with those

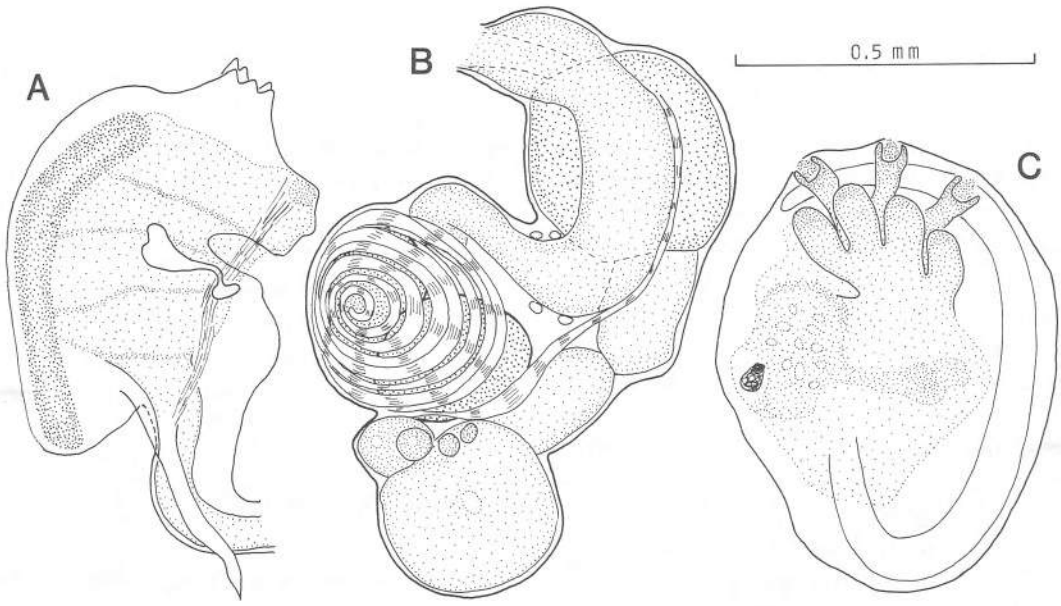

Fig. 4 — *Didemnum lutarium* Van Name, 1910: A, thorax; B, abdomen; C, larva.

in the original description of *Didemnum lutarium*, as the colony thickness and color, the strong muscular bands in the dorsal side of the thorax, numerous stigmata in a row, and two testis vesicles. The only character that did not agree with the description made by Van Name (1945), was the size and shape of the spicules. Thus we concluded that *D. lutarium* should be considered a valid species, in which we include the Brazilian specimens.

In the Caribbean area, *D. duplicatum* Monniot, 1983 has also two testis follicles, and rather small cloacal apertures. But this species differs in several characters such as the insertion of the retractor muscle process, the number of stigmata in a row and, above all, the larval structure.

The distribution of *D. lutarium* extends from New England to Brazil along the West Atlantic Coast.

Didemnum perlucidum Monniot F., 1983

Didemnum perlucidum: Monniot F., 1983: 29, Guadeloupe; Monniot *et al.*, 1985: 486, Polynesia; Monniot and Monniot, 1987: 40, Polynesia.

Localities: Ponta do Baleeiro – intertidal, Praia do Cabelo Gordo de Dentro – shallow water. Material in MNHN A2 DID C 184.

The colonies form very thin sheets (1 mm) that grow on multiple substrates such as rock, wood, ropes, PVC tubes, and other ascidians, in shallow water. The crusts extend several centimeters in diameter.

Externally, the colonies resemble marble with white and gray tones. The gray tones are due to the presence of an extensive net of cloacal canals where the tunic has only sparse spicules. Spicules are always present, especially in the superficial layers of the tunic, and they can reach 40 µm long, however, most spicules reach only 20-30 µm.

The internal characters fully agree with those given in the description of the specimens from Guadeloupe by F. Monniot (1983). The zooids are approximately 1 mm in length; the oral siphon is always short and large; the cloacal siphon is wide exposing almost the entire branchial sac, and has no languet. There are seven to eight stigmata per half row in the anterior rows. The thoracic lateral organs are located near the posterior edge of the body wall at the level of the third or fourth row of stigmata. The retractor muscle process is anterior to the oesophageal pedicle, between it and the end portion of the endostyle. The testis is a single body, surrounded by five to seven coils of the sperm duct and the ovary is located between the testis and the stomach. The ovary consists of one large and a number of smaller oocytes. The larvae are rounded, with three long adhesive papillae and four pairs of ampullae, and they measure 0.4-0.5 mm. The oozooid contains three rows of stigmata with four to six stigmata per half row.

Didemnum perlucidum is common in New Caledonia (unpublished data). Present in all oceans, as a white fouling ascidian, it has probably often been confused with *D. candidum*.

Didemnum psammathodes (Sluiter, 1895)

Leptoclinum psammathodes: Sluiter, 1895: 171, North Australia.

Didemnum psammathodes: Monniot F., 1983: 31, Guadeloupe; Goodbody, 1984a: 68, West Indies; Rodrigues and Rocha, 1993: 729, South-Eastern Brazil.

Localities: Ponta do Baleeiro, Praia Grande – intertidal. Material in MNHN A2 DID C 186, MZUSP 11193, 11194, 11195. Type specimen ZMA TU 588.

This species is very common in the region, encrusting the lateral surfaces and undersides of boulders, and vertical walls, usually shaded sites. The external appearance is exactly the same as that described for the specimens in other places in the world. Spicules are sparse and small (5-30 µm). The zooids are 0.4 to 0.75 mm in length with a single testis surrounded by six to eight coils of the sperm duct. The larvae have three adhesive papillae and four pairs of ampullae, and they are 0.4 to 0.5 mm in trunk length. *D. psammathodes* has been found in all warm seas.

Didemnum rodriguesi Rocha and Monniot, 1993

Didemnum rodriguesi: Rocha and Monniot, 1993: 261, Brazil and New Caledonia.

Localities: Praia Grande – intertidal. Material in MNHN A2 DID C 187, MZUSP 11121 (Type) 11122, 11123.

This species encrusts the lateral surfaces of large boulders, growing directly on the rock, algae, Bryozoa and other organisms. It forms sheets

several centimeters across and it can have an irregular surface due to obstacles to its growth. Two distinct external characteristics are the bright orange or red color and the microscopic reticulate arrangement of spicules on the colony surface. The larvae also have distinctive features, such as the three short and wide adhesive papillae and eight pairs of ampullae.

Its distribution is presently restricted to New Caledonia in the Western Pacific and Brazil.

Didemnum speciosum (Herdman, 1886)

Leptoclinum speciosum: Herdman, 1886: 274.

Didemnum speciosum: Rodrigues and Rocha, 1993: 930 and synonymy, Brazil.

Localities: Ponta do Baleeiro – intertidal. Material in MNHN A2 DID C 185.

The colonies are small, thin and white. The spicules are very abundant and small (15-32 µm). The zooids are less than 1 mm in length with wide cloacal siphons. The retractor muscle process is inserted in an anterior position, located between the oesophageal pedicle and the endostyle. The testis is a single body surrounded by six coils of the sperm duct. The small larvae (0.35 mm) have no distinctive features; they have three adhesive papillae and four pairs of ampullae.

The distribution of this species is restricted to the Brazilian coast.

Didemnum vanderhosti Van Name, 1924
(Fig. 5A, B, C – Pl. IF)

Didemnum vanderhorsti: Van Name, 1924: 25; 1930: 438; 1945: 89; Millar, 1958: 498; 1962: 62; Goodbody, 1984a: 38, 1984b: 65.

Localities: Ilha de São Sebastião, Praia do Araçá, Ponta do Baleeiro, Praia Grande – intertidal. Material in MNHN A2 DID C 182, MZUSP 11196, 11197. Type AMNH 714.

This species is very abundant intertidally. It encrusts the lateral surfaces and undersides of boulders or vertical rocky walls that are protected from direct sunlight. It forms thin sheets 1-2 mm thick and several centimeters in diameter. The edges of the colony are sometimes unattached to the substrate, forming pendent thick masses. The color varies from dark-purple, dark brown or chocolate brown to a marble mixture of these and beige.

The spicules (10-35 µm) are not abundant but always present. They are rounded with numerous short rays (Pl. IF).

The zooids are not arranged in systems but the presence of fecal pellets are evidence that the

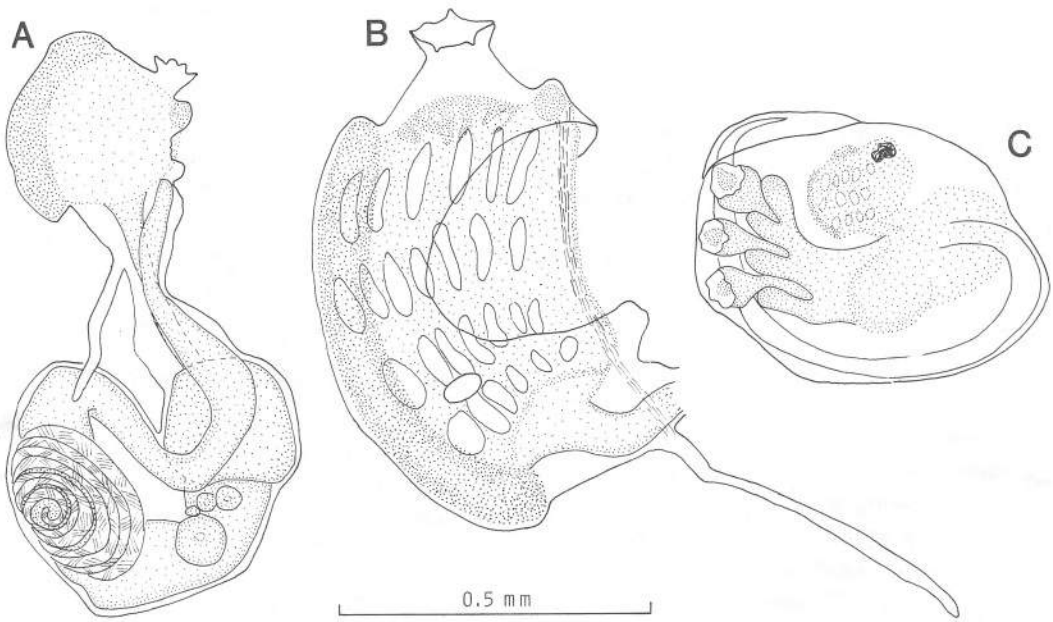

Fig. 5 — *Didemnum vanderhorsti* Van Name, 1924: A, zooid; B, thorax; C, larva.

canals converge to the common cloaca from all directions. Spicules are absent in the thin layer of the transparent test around the common cloacal apertures.

The zooids are rather small and rarely exceed 1 mm in length. The oral siphon is tube-like with short lobes and the cloacal one is generally narrow with no languet (Fig. 5A). The large and elliptical thoracic lateral organs are located at the level of the third row of stigmata or the third transversal vessel between the edge of the body wall and the endostyle (Fig. 5A). The retractor muscle process is long and unattached at the middle of the oesophageal pedicle.

The digestive tract has no distinct characteristics, the stomach is cylindrical. Both sexes are present (Fig. 5B). The testis is a single body, partially surrounded by nine coils of the sperm duct. The ovary consists of one large and several small oocytes located between the testis and the stomach.

Larvae (Fig. 5C) are 0.5 mm long (including adhesive papillae) and each has three adhesive papillae and four pairs of ampullae. Individuals are not gemmiparous and the oozooid has three rows of stigmata with five to six stigmata per side. This is the first description of *D. vanderhorsti* larvae.

Except from the color of the colony, this species differs in many characters from *D. cineraceum* (Sluiter, 1908) redescribed by Monniot F. (1983).

Millar (1958) noted the scarcity of records of this species, however, it is very abundant in south-eastern Brazil. The lack of records of species found between the São Sebastião Channel and the Caribbean Sea (Goodbody, 1984a, b) may be due to the fact that very little collecting has been done in this area.

DISCUSSION

Until recently only two species of the genus *Didermnum* were known in Brazilian waters, *D. speciosum* and *D. vanderhorsti, D. candidum* reported by Van Name (1945) for São Sebastião Island should be reexamined, since this species was redescribed by Lafargue (1974) and probably does not occur in Brazilian waters. This genus is well diversified along all tropical and subtropical coasts in the world and there was no reason to expect that it would not be diversified here. Nevertheless, this work should not be considered as a comprehensive inventory of the species in the São Sebastião region as it was restricted to intertidal or very shallow habitats.

Among the nine species collected, *D. speciosum* is the only one found to be endemic in Brazil. This species has no distinct features that would allow an undoubted identification. It belongs to the "white didemnid" group that has always allowed confusion among its species and it may be found elsewhere with further studies. Two other species are restricted to the western Atlantic: *D. lutarium* and *D. vanderhorsti*.

Surprisingly, we found many species whose distributions are much wider and species known only from the Pacific region. This scattered distribution could reflect a bias of sampling due to the small number of taxonomic papers on ascidian fauna around the world, so that many species could have a wide distribution that is still unknown.

Recent introductions of ascidians have already been reported in many localities (see Monniot *et al.*, 1985 for a bibliographic review). These species are generally transported on the hulls of ships. The newly introduced species usually are restricted to the port area, and they are rarely found among the common species of the region. The São Sebastião Channel is also subjected to the introduction of new species since it has a large harbor for oil-tankers that stay in the port for long periods of time while loading and unloading. Among the species reported in this paper, *D. perlucidum, D. ligulum* and *D. ahu* were found to be very common on artificial substrata and are considered components of the "fouling community", but they were also present on the natural boulders, which might indicate that they were recently introduced.

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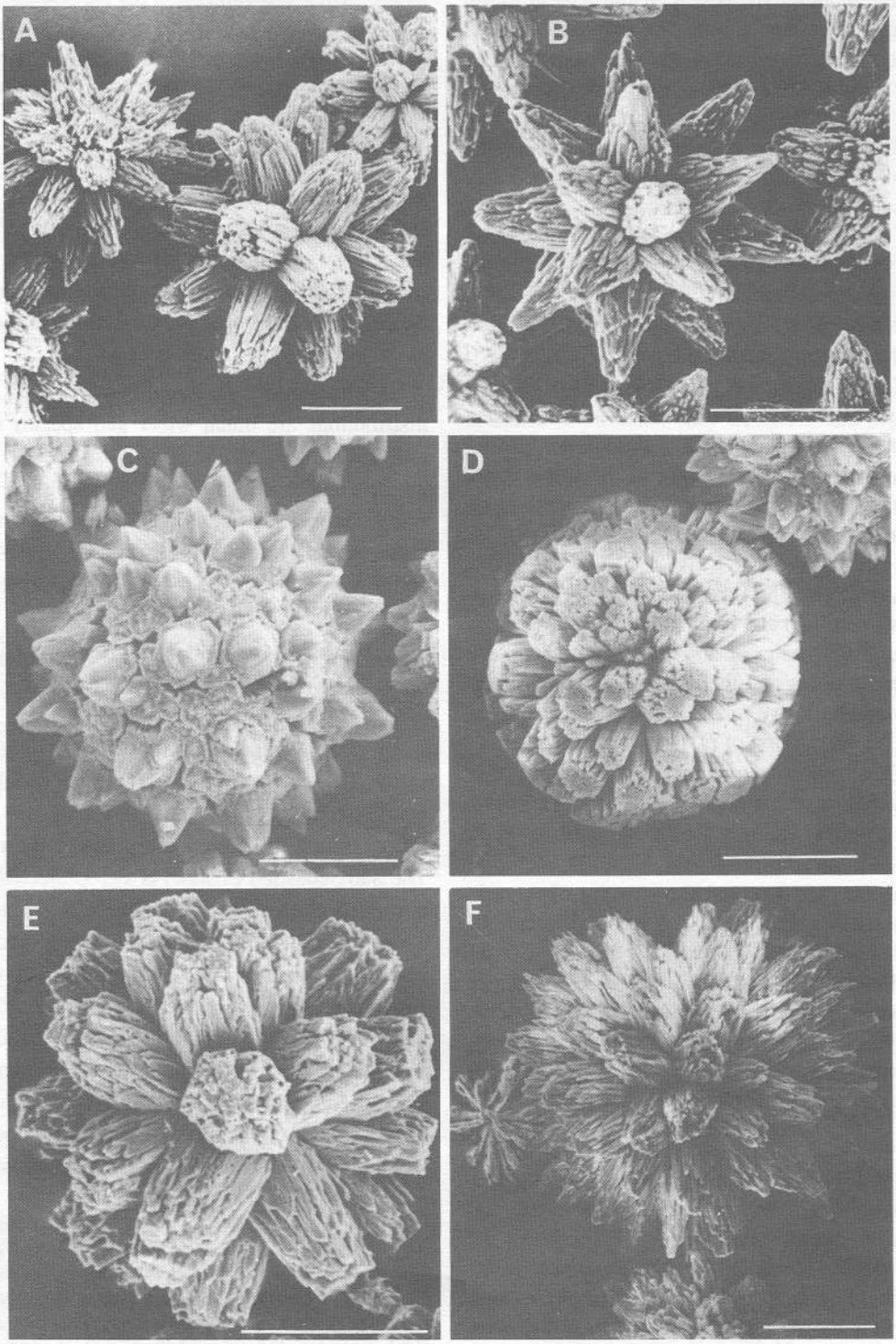
PLATE I



Spicules in A, *Didemnum alui*; B, *Didemnum granulatum*; C and D, *Didemnum ligulum*; E, *Didemnum lutarium*; F, *Didemnum vanderhorsti*. Scale = 10 µm.

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