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FIRST REPORT OF *PROCHLORON* IN ASSOCIATION WITH THE GENUS *POLYSYNCRATON* DIDEMNID ASCIDIAN (TUNICATA)

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PROCHLOROPHYTES
POLYSYNCRATON
ASCIDIE DIDEMNIDAE
ASSOCIATION

RÉSUMÉ. — Pour la première fois des Prochlorophytes ont été trouvées associées au genre *Polysyncraton*, Ascidie Didemnidae (Tunicier). L'espèce *Polysyncraton bilobatum* Lafargue, 1968 est commune aux îles Galapagos ainsi que sur les côtes d'Europe et sur les côtes occidentales d'Afrique. Il s'agit d'une espèce clé dans la lignée évolutive des Didemnidae sur les côtes de France et probablement dans le monde.

PROCHLOROPHYTA
POLYSYNCRATON
DIDEMNID ASCIDIAN
ASSOCIATION

ABSTRACT. — This is the first report of Prochlorophyta in association with the genus *Polysyncraton*, a Didemnid Ascidian (Tunicata). The species *P. bilobatum* Lafargue, 1968 is common near Galapagos islands as well on the European and the West-African coasts. It is a key species in the evolutive line of the Didemnidae in the French coast and probably in the world.

INTRODUCTION

From January 27 to February 10, 1978 an oceanographic cruise aboard the vessel « Alpha Helix » explored Galapagos waters. Several of the SCUBA dive collections yielded a species of didemnid ascidians possessing associated unicellular algae of the genus *Prochloron* Lewin 1977, 1984, belonging to the Prochlorophyta Lewin 1976. We then investigated the distribution of this association within the upper 20 m of the infralittoral zone. The ascidian *Polysyncraton bilobatum* taxonomically intermediate between the genera *Polysyncraton* and *Didemnum* also thrives on European and West-African coasts.

On the Galapagos coasts the ascidians possessing associated procarotic algae either belong to the didemnid genus *Polysyncraton* or to the polycitorid genus *Cystodytes*. The *Prochloron* cells are attached in a very loose manner to these ascidians colony surface. This association is non-obligate.

This is the first report of Prochlorophyta in association with the genus *Polysyncraton*. The species also is common on the European and the West-African coasts.

Most of the colonial ascidians containing associated unicellular Prochlorophyte algae belong to the family Didemnidae (Lewin and Cheng 1975; Newcombe and Pugh 1975; Kott 1977, 1980; Lewin 1984). Examples from other ascidian families have not been found in obligate association (Kott *et al.*, 1984).

MATERIAL AND METHODS

The colonies of didemnids were collected by SCUBA diving. A gentle menthol anaesthesia preceded fixation by 7% neutral formaldehyde in seawater. Zooids and larvae were stained and embedded in resin for microscopic study. For detailed methods see Lafargue (1968) and Lafargue & Wahl (1987).

RESULTS

The Galapagos form of this didemnid species is, the colour except, identical to the European *Polysyncraton bilobatum* Lafargue, 1968, as is apparent from

the following description :

Polysyncraton bilobatum Lafargue, 1968 (Fig. 1 and 2)

Synonymy : *Polysyncraton bilobatum* Lafargue, 1968 p. 401; Lafargue 1975, p. 134; Pars Medioni 1970 p. 38.

Simplified identification file :

Average thickness of colonies is 1 mm. They are white, green or pink colored. The white form is lacking associated algae.

Spicules (Fig. 1D, fig. 2) are of stellate shape with 10-20 rays per optical hemisphere and a mean diameter of approximately 20 μm . Their density in a colony varies within a population : extremely dense in some specimens, coarse in others.

Zooid length is about 1.5 mm. The opening of the oral siphon shows 12 tentacles (Fig. 1,E). There are 4 rows of stigmata with 5-7 stigmata per half-row. The atrial aperture is large in the well relaxed zooid. The wing-shaped vertical spiculogenic organs are to be found close to the rim of the atrial aperture

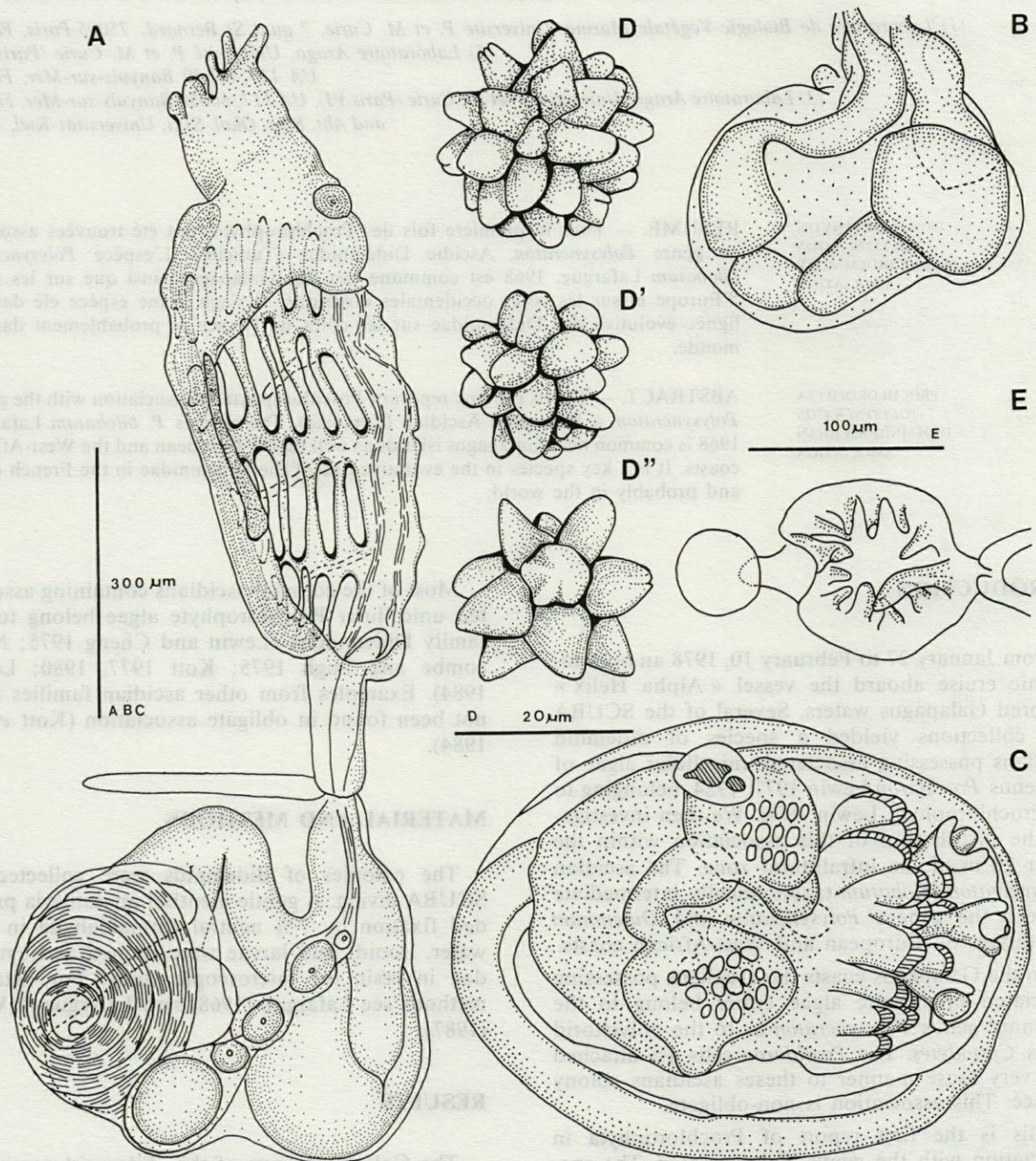


Fig. 1. — *Polysyncraton bilobatum* Lafargue, specimens from Galapagos islands. A, zooid; B, digestive gut; C, larva; D,D', D'', spicules; E, buccal tentacle.

between the second and the third row of stigmata (Fig. 1 A). The muscular appendix is anchored near the oesophageal neck. The intestine is doubly looped (Fig. 1B).

Gonads (Fig. 1A) lie on the left side. The testis is characteristic for the species : it consists of two tightly joined hemispheres. The spermduct forms a spiral of 7-8 tight coils. The ovary does not differ from the family's type. The larva (Fig. 1C) large (600 x 450 μm) and gemmiparous, is characteristic for the genus : 3 adhesive papillae and 7-8 pairs of lateral

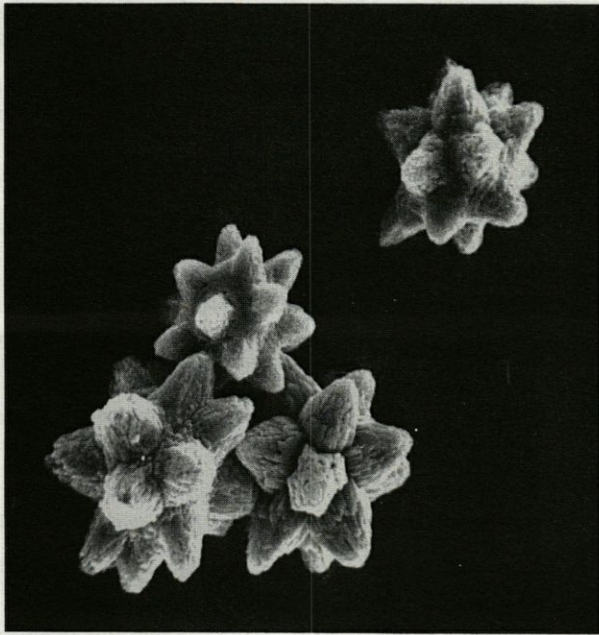


Fig. 2. — Spicules of *Polysyncraton bilobatum* Lafargue seen in the SEM, specimens from Galapagos islands.

ampullae (according to age) in two planes parallel to that defined by the papillae. The oozoid in Fig. 1C has produced a blastozoid. The first can be identified by its sensory vesicle.

Ecology : infralittoral zone. *Prochloron* is found on the colonies between 8-12 m (although the range of the host is from 2 to 20 m).

Distribution : french coasts (Channel, Atlantic, Mediterranean), west-African coast (F. Monniot, 1969) and Galapagos Islands.

TAXONOMIC DISCUSSION

Polysyncraton bilobatum Lafargue, 1968, is an intermediate form between the genera *Polysyncraton* and *Didemnum*. The divided testis, the loosely coiled spermduct and the gemmiparous larva are distinctive characters of the genus *Polysyncraton*. The genus

Didemnum, on the other hand, possesses a whole testis, a tightly coiled spermduct and larvae that are not gemmiparous. Its imperceptibly divided testis and the touching coils of its spermduct make *P. bilobatum* an evolutive link with mixed characters of the two genera.

The specimens collected near the Galapagos Islands have been attributed to *Polysyncraton bilobatum* Lafargue, 1968, because we consider the observed differences with the species type too small to justify the constitution of a new species. These differences are : the color (the species type rarely being white, but normally orange-yellow); the length of the muscular appendix (which too, is smaller in the Galapagos specimens : 25 instead of 35 μm). Neither can the absence or presence of symbiotic algae of the genus *Prochloron* be regarded as an argument in favor of a separation of species : the association *Prochloron* - *Polysyncraton bilobatum* is not obligate and even if, in general, European *P. bilobatum* do not have symbiotic algae, there are exceptions (Lafargue *et al.*, 1986). One of us (M.W.) has recently found filamentous *Cyanobacteria* associated in a quite similarly loose, superficial manner to *P. bilobatum* of Banyuls-sur-Mer, french Mediterranean (unpubl.).

In the evolutionary history of the Didemnidae, *P. bilobatum* has been playing a key-role : 17 didemnid species of the French coast seem to have been descended from it : 12 species of the genus *Didemnum*, 4 belonging to the genus *Trididemnum* and 1 *Didemnopsis* species (Lafargue, 1983).

THE PROCHLORON - P. BILOBATUM ASSOCIATION

Within the Galapagos archipelago the distribution of *Prochloron* is quite heterogenous. It varies with depth and locality. They are most abundant in the western part of the area. Tagus Cove, on the west coast of Isabella Island is by far the richest locality, both with regard to hard and soft bottom. The optimum depth range seems to lie between 8 and 12 m. The volcanic stone may be covered to more than 60% by ascidians, which in their turn, have up to half of their surface occupied by *Prochloron*, thus appearing greenish or pinkish. The algal cover of the colonies is not uniform, either : fissures and those parts sheltered in microcavities are greenest (most densely covered). Those colonies most exposed to sunlight (on horizontal, shallow substratum) lack algae. In this context it is interesting to note that light may play an important role in larvae release and larval behaviour (Olson, 1983) : Didemnid larvae that contain symbiotic algae are released at noon (when light intensity is highest) thus enabling them during fixation to avoid too intensely irradiated localities.

Close-up investigation of *P. bilobatum* colonies show that the localisation of the *Prochloron* cells is positively superficial like in *Cystodytes*. The algae are restricted to the outer colony surface where they are most densely packed around the oral siphon openings. They do not seem to enter the cloacal network nor the larval tunic. This is an example of an extracellular association so loose, that the algae sometimes detach from the colonies during collection. Other species of didemnid ascidians show a much closer association with *Prochloron* and, in some instances, metabolic exchanges have been shown to take place (Hardy *et al.*, 1981; Muller *et al.*, 1984), as has been for phagocytosis (Cox, 1983).

In tropical seas the cases of unicellular procaroytes (*Cyanophyta*, *Prochlorophyta*) living on or within didemnid ascidians are numerous. Species of the didemnid genera *Lissoclinum*, *Diplosoma*, *Polysyncraton*, *Didemnum*, *Trididemnum* may exhibit such associations.

Cox (1986) give comparison of *Prochloron* from different hosts by structural and ultrastructural characteristics. Using different criteria *Prochloron* cells can be divided into three quite clear-cut groups : group I where all *Prochloron* cells living on the external surface of the hosts, group II with *Prochloron* cells apparently engulfed by hosts cells, group III the cloacal-cavity symbiosis which represents the most advanced stage of the symbiosis.

The most primitive stage is seen in *Polysyncraton bilobatum*.

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