BULLETIN DE L'INSTITUT ROYAL DES SCIENCES NATURELLES DE BELGIQUE, BIOLOGIE, **66** suppl. : 89-101, 1996
BULLETIN VAN HET KONINKLIJK BELGISCH INSTITUUT VOOR NATUURWETENSCHAPPEN, BIOLOGIE, **66** suppl. : 89-101, 1996

The genus *Batzella* : a chemosystematic problem

by Rob W.M. van SOEST, Jean-Claude BRAEKMAN, D. John FAULKNER, Eduardo HAJDU, Mary Kay HARPER & Jean VACELET

Abstract

Biogenetically unrelated cyclic guanidine alkaloids and pyrroloquinoline alkaloids have been reported from sponges assigned to the genus *Batzella*. These sponges have been assigned to this genus because of their possession of a simple complement of thin strongyles in irregular plumoreticulate arrangement. Cyclic guanidine alkaloids were first reported from an alleged axinellid species from the Caribbean, *Ptilocaulis* aff. *P.spiculifer*, and subsequently from a second Caribbean specimen identified as *Ptilocaulis spiculifer* and at the same time from a Red Sea poecilosclerid, *Hemimycale* sp. Closely related compounds were described from a Caribbean specimen identified as *Batzella* sp. and also from the poecilosclerids *Crambe crambe* (Mediterranean) and *Monanchora arbuscula* (Brazil). Isobatzellins (pyrroloquinoline alkaloids) were reported from a black deep-water species from the Bahamas identified as *Batzella* sp. Chemically related pyrroloquinoline alkaloids were found in Pacific representatives of the fistular poecilosclerid genus *Zyzzya*, the hadromerid genus *Latrunculia* and the ?haplosclerid genus *Prianos*. Most of the voucher specimens involved in this puzzle were re-examined and several conclusions can be drawn : When inspected closely it appears, that the cyclic guanidine alkaloids are produced by sponges containing anisostrongyles, often in two categories, a thicker and a thinner one. *Monanchora arbuscula*, which has been recently discovered to produce these compounds, has monactinal spicules differentiated into a thinner subtylostyle and a thicker (tylo-) style, but many specimens have anisostrongylote modifications. Microscleres in *Monanchora* can be absent or very rare. By association, all the sponges from which cyclic guanidine alkaloids are known may be united in one family, possibly in a single wider defined genus *Monanchora*. However, further relationships with *Crambe* need to be studied. Both have cyclic guanidine alkaloids, both have megascleres of very variable shape and thickness, differentiated mostly into two overlapping categories, microscleres and other additional spicules are often rare or absent. Relationships with the type of *Hemimycale*, viz. *H. columella* remain obscure, but in view of the much larger spicules of that species and the intricate ectosomal specialization (lacking in the above mentioned specimens) it is possible that similarities between the Red Sea *Hemimycale* and the European species are the product of parallel evolution. The strongyles of sponges producing pyrroloquinoline alkaloids are perfect isostrongyles and in the ectosome these are arranged in a definite ectosomal tangential crust. A good proportion of these strongyles have a faint spination on the apices. Assignment of these sponges to *Batzella* rests on the properties of its type species *Batzella inops*. Examination of a type spicule slide of that species did not solve that question, but until further notice *Batzella* may be used for the deep-water material. A further unsolved problem that remains is the phylogenetic relationships of *Batzella* with *Zyzzya* and *Latrunculia*. The likelihoods of possible causes for this distribution of compounds are discussed.

Keywords : Chemosystematics, Poecilosclerida, *Batzella*, *Monanchora*, *Crambe*, *Zyzzya*, *Latrunculia*, cyclic guanidine alkaloids, pyrroloquinoline alkaloids.

Résumé

La présence d'alcaloïdes guanidiniques cycliques et pyrroloquinoliniques chez des éponges ayant été identifiées comme appartenant toutes au genre *Batzella* a été démontrée bien que ces deux types d'alcaloïdes ne soient pas biogénétiquement apparentés. Ces éponges ont été classées dans le genre *Batzella* car elles possèdent de fines strongyles disposés selon un arrangement plumoréticulé irrégulier. Des alcaloïdes guanidiniques cycliques ont été isolés pour la première fois à partir d'une axinellide des Caraïbes identifiée à *Ptilocaulis* aff. *P. spiculifer*, et ensuite simultanément à partir d'une seconde espèce des Caraïbes (*Ptilocaulis spiculifer*) et d'une poeciloscleride de la mer Rouge (*Hemimycale* sp.). Des alcaloïdes de structure apparentée ont aussi été isolés à partir de *Batzella* sp. (Caraïbes), de *Crambe crambe* (Méditerranée) et de *Monanchora arbuscula* (Brésil). Des isobatzellines (alcaloïdes pyrroloquinoliniques) ont été isolées d'une *Batzella* sp. de couleur noire vivant dans les eaux profondes au large des Bahamas. Des alcaloïdes apparentés ont été isolés d'une poecilosceride du genre *Zyzzya*, d'une hadroméride du genre *Latrunculia* et d'une haploscléride du genre *Prianos*. Ces trois dernières éponges sont toutes originaires du Pacifique. Les spécimens types de toutes ces éponges ont été réexaminés et il est apparu que les alcaloïdes guanidiniques cycliques sont systématiquement présents chez les éponges contenant deux catégories d'anisostrongyles, des fins et des épais. *Monanchora arbuscula*, qui contient également ce type de composés possède des spicules monactinaux différentiés en substylostyles fins et en tylostyles plus épais; de nombreux spécimens possèdent cependant des anisostrongylotes modifiés. Par ailleurs chez *Monanchora*, les microsclères peuvent être absents ou très rares. Il résulte de l'ensemble de ces observations que les éponges possédant des alcaloïdes guanidiniques cycliques pourraient être réunies en une seule et même famille et éventuellement en un seul et même genre élargi, le genre *Monanchora*. Cette éventualité demande cependant à être confirmée par une étude comparative plus détaillée entre les éponges des genres *Crambe* et *Monanchora*. Les éponges de ces deux genres possèdent des alcaloïdes guanidiniques, des mégasclères de taille et de forme variables, séparables grossièrement en deux catégories se recouvrant partiellement. Les microsclères et d'autres types de spicules sont rares ou absents. Les relations de ces espèces avec *Hemimycale columella* ne sont pas évidentes. Cependant, compte tenu des spicules très larges et de la spécialisation ectosomale complexe de cette dernière, il est possible que les similitudes entre l'*Hemimycale* de la mer Rouge et les espèces Européennes soient le résultat d'une évolution parallèle. Les strongyles des éponges produisant des alcaloïdes pyrroloquinoliniques sont des isostrongyles parfaits et dans l'ectosome ceux-ci sont disposées en une couche ectosomale tangentielle bien nette. Une partie importante de ces strongyles ont une spination fine au niveau des sommets. Le rattachement de ces éponges au genre *Batzella* se base sur les propriétés de l'espèce type *Batzella inops*. L'examen d'une coupe d'un spicule typique de cette espèce n'a pas permis de résoudre la question, mais jusqu'à preuve du contraire, la dénomination *Batzella* peut être utilisée pour décrire le spécimen collecté en eaux profondes. Un autre problème non résolu est la relation phylogénétique entre les genres *Batzella*, *Zyzzya* et *Latrunculia*. Les causes possibles de la répartition des alcaloïdes dans ces genres sont discutées.

Mots-clés : Chemosystematique, Poecilosclerida, *Batzella*, *Monanchora*, *Crambe*, *Zyzzya*, *Latrunculia*, guanidine alkaloïdes cycliques, pyrroloquinoline alkaloïdes.

Introduction

Secondary metabolites have been shown to be useful taxonomic markers in several groups of sponges, notably Dictyoceratida and Verongida (BERGQUIST & WELLS, 1983). In groups with easily recognizable and unequivocal morphological characters, it has been established, that chemistry may reflect different levels of morphological similarities. From this it is concluded that phylogeny of sponges may be deduced from chemical as well as morphological evidence. Ongoing studies (e.g. BRAEKMAN *et al.*, 1992; VAN SOEST *et al.*, this volume; ANDERSEN *et al.*, submitted) show that classification of taxa with controversial status at the ordinal or family level may be considerably improved by using chemical evidence.

However, in many investigated cases no apparent congruence between chemical and morphological similarity has been found. There are four possible reasons for such incongruence:

- identical molecular structures may be independently derived through different biogenetic pathways,
- molecules are produced by non-specific microsymbionts,
- molecules are produced by unidentified epibionts,
- morphological characters are not homologous : the identification/classification is wrong.

It is of paramount importance to taxonomists and chemists to establish which of the four explanations apply to cases in hand. Taxonomy of many groups of sponges is hampered seriously by the poor resolution of available morphological classifications and the applicability of chemical characters may help considerably to solve problems (KELLY-BORGES *et al.*, 1994; FROMONT *et al.*, 1994). Chemists need an accurate identification to be able to repeat and develop their chemical studies. Such a case in hand is the genus *Batzella* TOPSENT (1894), which features prominently in recent chemical literature, but is of uncertain taxonomic status. Two unrelated classes of compounds were described from species identified as *Batzella*, viz. cyclic guanidine alkaloids, some of which show *in vitro* inhibitory activity against HIV (MAI *et al.*, 1992; PATIL *et al.*, 1995), and cytotoxic pyrroloquinoline alkaloids (SAKEMI *et al.*, 1989). These molecule types are shared with sponges identified in other genera : *Ptilocaulis*, *Hemimycale*, *Crambe*, *Monanchora* (cyclic guanidine alkaloids) and *Zyzzya*, *Damiria*, *Histodermella*, *Latrunculia*, *Prianos* (pyrroloquinoline alkaloids). It is the purpose of this study to demonstrate that the chemically studied *Batzella*'s are members of different genera, and thus to explain the discrepancy of chemistry and morphology due to the non-homology of the morphological characters used for assigning them to the same genus.

Material and methods

Dissociated spicules and thick sections of the following type and voucher specimens were studied under light- and SEM microscopy :

Halichondria inops TOPSENT (1891), labeled "type", microscopic slide only, from Ile Verte, Roscoff, France, MNHN-L.B.I.M. DT. 2109

Ptilocaulis spiculifer, voucher of KASHMAN *et al.* (1989), Bahamas, HBOI/DBMR # 10-VI-86-2-18, identified by S.A. POMPONI

Batzella sp. voucher of MAI *et al.* (1992), PATIL *et al.* (1995), Chub Cay, Bahamas, 18 m, ZMA POR. 8788, identified by R.W.M. VAN SOEST

Hemimycale sp., voucher of KASHMAN *et al.* (1989), microscopic slides only, Shahag Rock, Suez, #1582, 10.7.86, identified by M. ILAN.

Batzella sp., vouchers of SAKEMI *et al.* (1989), Bahamas, off Grand Bahama Isl., deep water (115-126 m), HBOI/DBMR # 3-VI-84-3-001 & 14-XI-87-85-1-001, identified by S.A. POMPONI.

Damiria sp., voucher of STIERLE & FAULKNER (1991), Palau Isl., ZMA POR. 8441, identified by R.W.M. VAN SOEST.

Monanchora arbuscula (DUCHASSAING & MICHELOTTI, 1864), voucher of TAVARES *et al.* (1994a, 1994b), from Salvador, Brazil, identified by E. HAJDU & R.W.M. VAN SOEST.

Prianos aurantiaca LÉVI (1958), labeled "type", Red Sea, DCl. 1300.

Suberites crambe SCHMIDT (1862), dry "type", ex Mus. Hist. nat. Genève., LMJG 15672.

Suberites fruticosus SCHMIDT (1862), dry "type", ex Mus. Hist. nat. Genève, LMJG 15105.

Pandaros arbuscula DUCHASSAING & MICHELOTTI (1864), lectotype, ZMA POR. 1728.

In addition to these, specimens and slides of the following sponges were studied for comparison :

Crambe crambe (SCHMIDT), Mediterranean, ZMA POR. 10966, 10971.
Ptilocaulis walpersi, Curaçao, ZMA POR. 3625
Ptilocaulis spiculifer, Red Sea, ZMA POR. 197
Zyzzya fuliginosa (CARTER), Seychelles, ZMA POR. 10537
Monanchora arbuscula, Curaçao, ZMA POR. 4610
Monanchora arbuscula, Colombia, ZMA POR. 6150
Monanchora stocki VAN SOEST, Cape Verde Islands, ZMA POR. 6937
Monanchora unguiculata (DENDY) Indonesia, ZMA POR. 7911
Hemimycale columella (BOWERBANK) N. Brittanny, France, ZMA POR. 4819
Hemimycale sp. Red Sea, ZMA POR. 10949

History and affinities of the genus *Batzella*

Batzella TOPSENT, 1894, with type species *Halichondria inops* TOPSENT, 1891, has a very simple skeleton of thin strongyles arranged in ill-defined plumose bundles. According to the description, the type species is thinly encrusting, smooth, soft and has a yellow colour. A slide made from the type was studied. Strongyles measure 170-200 by 2-3 μm (n = 25), they are isodiametric, in general equally shaped at both apices, but a few may be found with one end somewhat mucronate. There is very little variation in length and thickness and all belong obviously to a single category. Most have a wide axial canal. The spicule bundles in the microscopic slide are vague, not visibly bound by spongin, and they follow a wispy course. There are no obvious bouquets at the surface (but these were reported by TOPSENT), and an ectosomal cover of spicules is not apparent. No chemistry is known from *B. inops*.
Sponges similar to that description are recorded from all over the world. Examples are *B. rosea* VAN SOEST, 1984 from the Caribbean and *B. frutex* PULITZER-FINALI, 1982 from Australia. No chemistry is known from these sponges.
TOPSENT (1928) described Mediterranean specimens of *B. inops* with different colour (brilliant carmine red), with larger spicules and with distinctly anisodiametric apices verging towards styles. Because of the apparent possession of both pure strongyles and anisostrongyles, he associated his genus with *Desmacidon columella* BOWERBANK, 1866, later made type of the genus *Hemimycale* BURTON, 1934. No chemistry is known from *Hemimycale columella* (but see below). TOPSENT also discussed the similarity of *Batzella inops* to *Crambe crambe* (SCHMIDT, 1862). Later authors, e.g. SARÀ (1958), ARROYO *et al.* (1976), and PULITZER-FINALI (1978), assigned specimens to *B. inops*, with strongyles and styles in widely varying sizes and shapes. In view of the great variability of *Crambe crambe* (cf. below) it is likely that some of the records of *B. inops* were in fact that species.
A second *Batzella* species described from the Mediterranean, *B. friabilis* PULITZER-FINALI (1978) possesses two categories of spicules, short and fat strongyles and long and thin "strongylotornotes". Through this differentiation it also approaches reduced *Crambe crambe*.
The family assignment of *Batzella* is problematic. TOPSENT (1894, 1928) thought it belonged to the family Desmacididae (as Desmacidonidae). Many later authors (e.g. PULITZER-FINALI, 1978) reporting on Mediterranean *Batzella* considered it to belong to the Halichondrida, family Hymeniacidonidae. Recent authors (VAN SOEST, 1984; WIEDENMAYER, 1989; HOOPER & WIEDENMAYER, 1994) allocate the genus to the Poecilosclerida, family Desmacididae. Unfortunately, that family is a dustbin (cf. HAJDU *et al.*, 1994), and a reassignment is inevitable. The poecilosclerid nature of *Batzella* is derived from its apparent similarity to *Strongylacidon* LENDENFELD (1894) (a genus with chelas and/or sigmas) and "sand sponges" of the genus *Phoriospongia* MARSHALL (1880). Its larvae have been described as big, yellow and having a bare posterior pole (TOPSENT, 1894), which is characteristic of Poecilosclerida.

Description of the *Batzella* sp. producing cyclic guanidine alkaloids

MATERIAL

ZMA POR. 8788, Bahamas, shallow water.

Shape, size, surface and consistency
An erect, bush-like branching-lobate sponge (Fig. 1) with highly penicillate surface projections. Specimen 12 cm high and wide. Lobate branches partly fused, clathrate, 2-4 cm in diameter.
Individual surface projections 5-10 mm long, irregular in outline, spiny. No oscules or other surface apertures are visible in the preserved specimens. Consistency tough, not easily torn or damaged.

Colour
Red in life. In alcohol it is beige.
Ectosomal skeleton : irregular, formed by the brushed endings of choanosomal spicule tracts. Individual tracts project quite variably beyond the ectosome.

Choanosomal skeleton
A strong but very irregular system of spongin fibres cored by variable amounts of spicules. Longitudinal spongin fibres follow a meandering course towards the surface, here and there anastomosing with neighbouring fibres. Towards the surface the fibres contain less spongin and consist

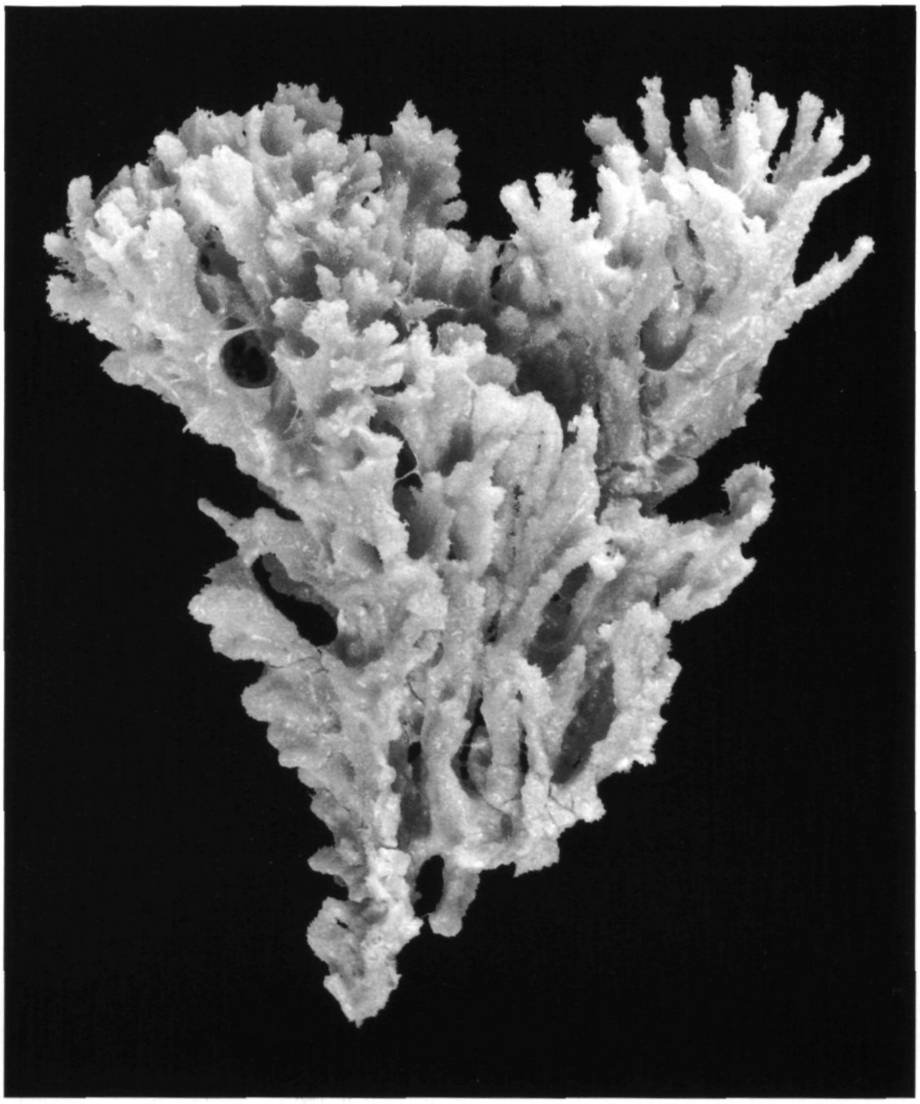


Fig. 1. - Habit of "*Batzella" sp. ZMA* POR. 8788, from shallow-water, near Chub Cay, Bahamas. Height 8 cm. The specimen is now considered a deviating specimen of *Monanchora arbuscula.*

of ill-defined bundles of spicules. Individual spongin fibres 50-70 μm in diameter, but with irregular, knotty outline; coring 5-10 spicules per cross section.

Spicules
Most spicules are rounded at both ends, but many are definitely anisostrongyles with one end somewhat swollen, the other tapering gradually (Fig. 2B). There appear to be two categories, although there is considerable overlap : long and thin spicules of which one end has a constricted "neck", 215-258 x 3-5 μm, and (relatively) short and fat (Fig. 2A), 174-216 x 4-6.5 μm.

Chemistry (Table 1)
Cyclic guanidine alkaloids isolated from this specimen include ptilocaulin, crambescidin and crambescidin-type alkaloids (cf. PATIL *et al.*, 1995).

OTHER SPONGES PRODUCING CYCLIC GUANIDINE ALKALOIDS (Table 1)

HARBOUR *et al.* (1981) had previously recorded similar cyclic guanidine alkaloids [ptilocaulin (**1**) and isoptilocaulin (**2**)] from a Bahamian specimen identified as *Ptilocaulis* aff. *spiculifer* (LAMARCK, 1814) by G.J. BAKUS. We have been unable to obtain the voucher. In 1989, KASHMAN *et al.* reported the isolation from another Bahamian specimen identified as *Ptilocaulis spiculifer* by S.A. POMPONI of ptilomycalin A (**4**), a cyclic guanidine alkaloid which is biogenetically related to the ptilocaulins. We have examined this material and it was found to be identical in shape and skeleton characteristics to the above described *Batzella* sp. Its spicules measured 189-249 x 2-4 μm and 177-210 x 4.5-6 μm respectively, closely similar to the above measurements. Its identification as *Ptilocaulis spiculifer* is

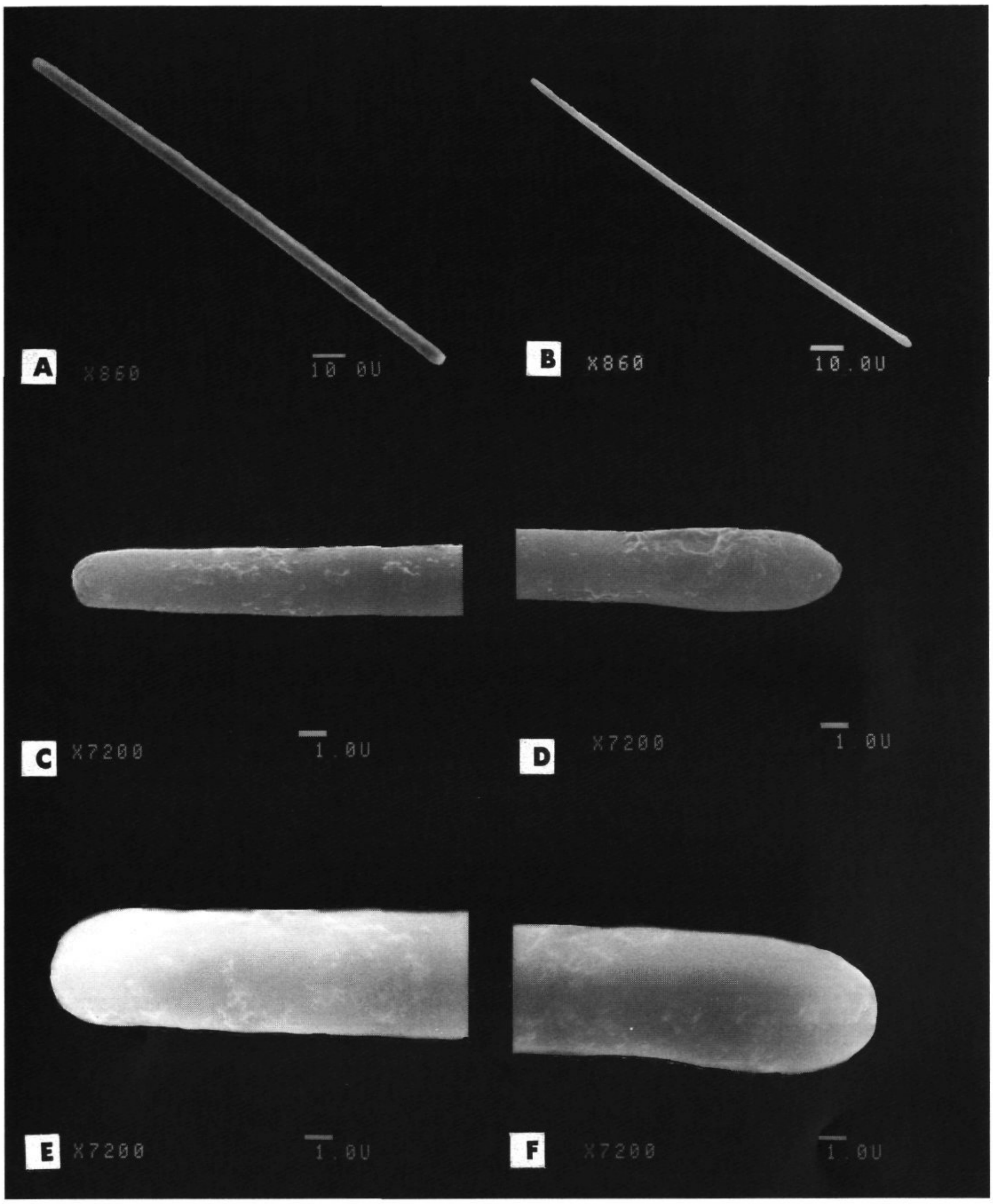


Fig. 2. - SEM pictures of spicules of "*Batzella*" sp., ZMA POR. 8788, from shallow-water (18 m) , near Chub Cay, Bahamas. - A. Thicker, almost isodiametric strongyle. - B. Thinner, strongly anisodiametric "strongyle". - C, D. Apices of B. - E, F. Apices of A.

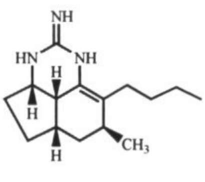
due to WIEDENMAYER's (1977) erroneous description of that species. Skeletal reticulation and spicule sizes and shapes of Central West Atlantic *Ptilocaulis* species (*P. walpersi* & *P. marquezi,* see ALVAREZ *et al.*, in the press) are considerably different. Notably the spiculation with styles up to 650 or 1000 x 18 µm make this clear.

In *P. walpersi* (identified by S.A. POMPONI) an unrelated, typical axinellid compound, oroidin, was found by WRIGHT *et al.* (1991), while in *P. trachys* (identified by R.W.M. VAN SOEST) a likewise unrelated, probably alga-derived peptide (majusculamide C) was found by WILLIAMS *et al.* (1993). It can be stated with certainty that sponges of the genus *Ptilocaulis* do not produce the cyclic guanidine alkaloids.

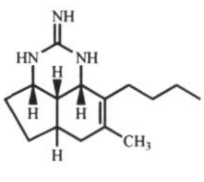
KASHMAN *et al.* (1989) also reported ptilomycalin A (**4**) from a Red Sea *Hemimycale* sp. We have been able to examine thick sections and a spicule slide of the voucher, and one of us (J.V.) has studied fresh material of this species. It is encrusting, with a blue-green or black-green colour. Its surface is a reticulum of polyangular or rounded porefields; oscules are raised into low tubes. The skeleton consists of bundles of strongyles and styles without visible binding

Table1:
Cyclic guanidine alkaloids recorded from sponges.

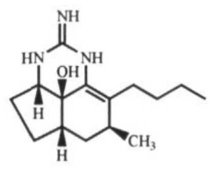
"*Batzella*" sp. shallow water	**1, 4-6, 11, 15-19**
"*Ptilocaulis* aff. *P. spiculifer*"	**1, 2**
"*Ptilocaulis spiculifer*"	**4**
"*Hemimycale*" sp.	**4**
Monanchora arbuscula	**1, 3,5**
Crambe crambe	**5, 14**



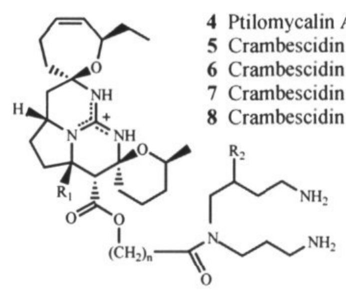
1 Ptilocaulin



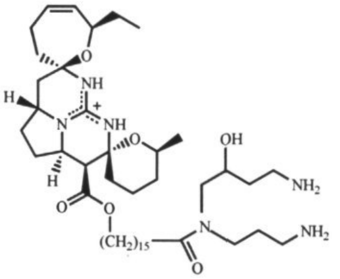
2 Isoptilocaulin



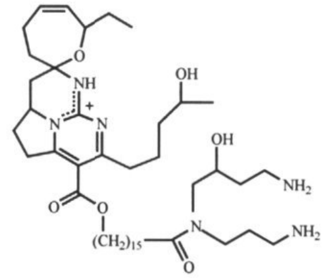
3 8b-hydroxyptilocaulin



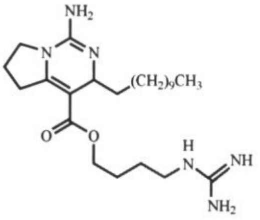
4 Ptilomycalin A; $n = 15$; $R_1 = R_2 = H$
5 Crambescidin 800; $n = 15$; $R_1 = H$; $R_2 = OH$
6 Crambescidin 816; $n = 15$; $R_1 = R_2 = OH$
7 Crambescidin 830; $n = 16$; $R_1 = R_2 = OH$
8 Crambescidin 844; $n = 17$; $R_1 = R_2 = OH$



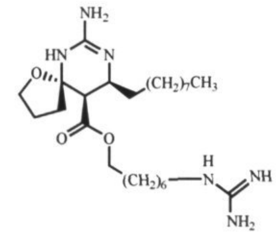
9 Isocrambescidin 800



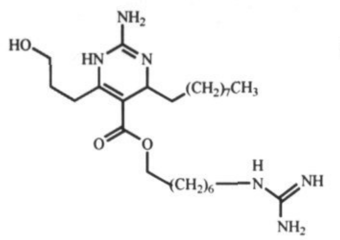
10 Crambidine



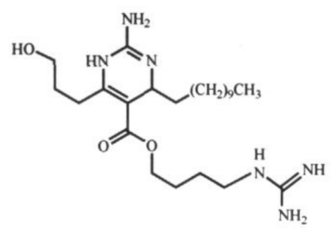
11 Crambescin A



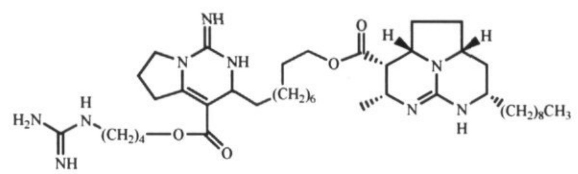
12 Crambescin B



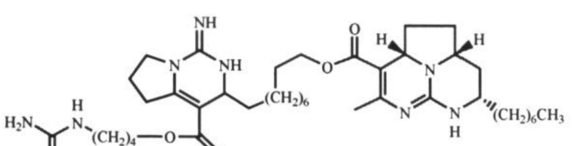
13 Crambescin C1



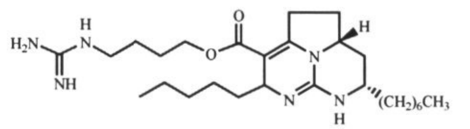
14 Crambescin C2



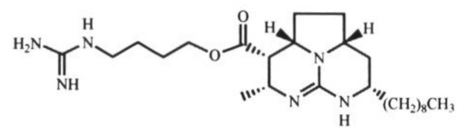
15 Batzelladine A



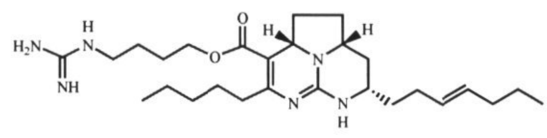
16 Batzelladine B



17 Batzelladine C



18 Batzelladine D



19 Batzelladine E

spongin; there is no special ectosomal skeleton. The spicules are thin with a wide axial canal. Many are anisostrongyles, with a swollen and a mucronate end. Possibly there is a category of shorter, mostly isodiametric strongyles (about 200-230 µm), and longer anisodiametric ones (260-315 µm), but a clear separation in size is not evident. VACELET *et al.* (1987) described calcareous spherules from this species and WILLENZ (1982) described spherulous and sacculiferous cells from it.

BERLINCK *et al.* (1990, 1992) and JARES-ERIJMAN *et al.* (1991) reported cyclic guanidine alkaloids of the crambescin- (**11-14**) and crambescidin-types (**5-10**) from the Mediterranean species *Crambe crambe* (SCHMIDT, 1862), identified by N. BOURY-ESNAULT. This is a vivid-red encrusting species with smooth or lumpy surface and a well developed venal canal system. The skeleton normally shows irregular bundles of megascleres bound by variable amounts of spongin; no special ectosomal skeleton. The spiculation is extremely variable, which has led to confusion in the past (cf. RÜTZLER, 1965). Completely developed specimens contain thin ectosomal subtylostyles of 220-320 x 3-6 µm, thicker choanosomal styles of 350-400 x 6-13 µm, unguiferate isochelas, and aster-like desmas. In many specimens, the desmas and microscleres are extremely rare or absent; also size and form of the megascleres are quite variable and may include anisostrongylote modifications.

TAVARES *et al.* (1994, 1995) reported further cyclic guanidine alkaloids [crambescidin 800 (**5**), ptilocaulin (**1**) and 8b-hydroxyptilocaulin (**3**)] from Brazilian and Belizean specimens of *Monanchora arbuscula* (DUCHASSAING & MICHELOTTI, 1864). This species is red, encrusting, with well-developed venal canal system emphasized by a whitish lining of the canals. Its skeleton consists of irregular bundles of megascleres, bound by spongin and anastomosing irregularly. No special ectosomal skeleton. Its spiculation includes thin ectosomal subtylostyles of 190-380 x 2-5.5 µm, thick ectosomal styles of wide size variation : 140-450 x 5-13 µm, occasionally with a well-developed tyle, anchorate-unguiferate isochelas and reduced sigma-like smaller chelas. Microscleres are often rare.

CONCLUSION

Based on morphological characters (skeletal structure, spicule shape variation, subdivision into two spicule categories, frequently occurring rarity or absence of microscleres) and on the closely related or identical chemistry we conclude that *"Batzella"* sp. *sensu* MAI *et al.* (1992) and PATIL *et al.* (1994), "*Ptilocaulis spiculifer*" *sensu* HARBOUR *et al.* (1981) and KASHMAN *et al.* (1989), *Hemimycale* sp. *sensu* KASHMAN *et al.* (1989), *Monanchora arbuscula* and *Crambe crambe* are all closely related. We suggest they should eventually be united in the same genus, which for priority reasons has to be *Crambe* , but a formal proposal will have to await a revision of all species. It remains to be investigated whether or not *Batzella inops* and *Hemimycale columella* also belong to this group. WILLENZ (1982) reports that the latter species shares sacculiferous cells with the Red Sea "*Hemimycale"* sp., but such cells are not exclusive as they have been also described from unrelated *Cyamon neon* by SMITH & LAURITIS (1969).

Alternative explanations for this distribution of chemical compounds (cf. above) are judged to be unlikely : there are no known microsymbionts or epibionts reported in the studied material and the chemistry is sufficiently unique to exclude convergent biogenetic pathways. In all studied specimens the cyclic guanidine molecules were the major compounds present. In view of the above, we assume that the occurrence of ptilomycalin A, crambescidin 800 and related guanidine alkaloids in New Caledonian asteroids (Echinodermata), as recently reported by PALAGIANO *et al.*, 1995, is not the result of biosynthesis of the compounds by these organisms. It is likely that these echinoderms have sequestered the compounds from sponges of the genus *Crambe/Monanchora*, as several asteroids are known sponge predators.

As to the specific status of "*Batzella "* sp., we propose to follow ALVAREZ *et al.* (in the press) in assigning it to a variable species *Monanchora arbuscula* until further revision of the Caribbean members of the group has been completed. Further corroboration for this is the unpublished chemistry ("almost identical to that produced by the Chub Cay specimen save minor derivatives", source M.K. HARPER) of a bushy specimen of *Monanchora arbuscula* from Jamaica (voucher ZMA POR. 11009), containing all the typical spicules, including two categories of styles, unguiferate isochelae as well as reduced sigma-like chelae and acanthose microrhabds. The habit of this specimen is strikingly similar to the Bahamas "*Batzella* ".

Description of the *Batzella* sp. producing pyrroloquinoline alkaloids

MATERIAL

HBOM 3-VI-84-3-001 & 14-XI-84-3-005, Bahamas, 115-125 m.

Shape, size, surface and consistency
Small, amorphous, sponge. Available voucher fragments are about 1 cm in thickness. Surface smooth, no apparent oscules. Consistency compressible but firm.

Colour
Black in life, tan in alcohol.

Ectosome
Detachable. There is a tangential crust of intercrossing spicules.

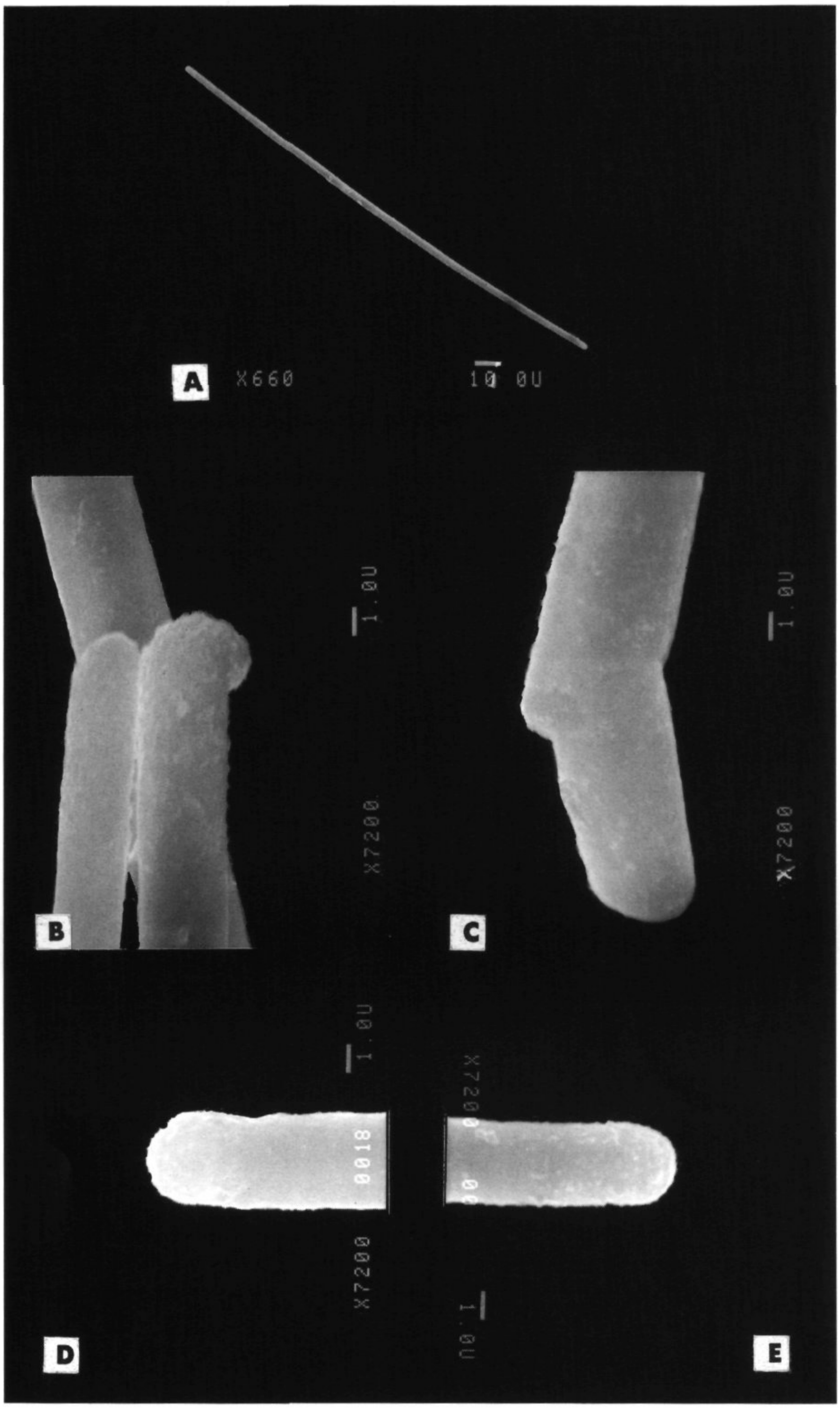


Fig. 3. - SEM pictures of spicules of "*Batzella*" sp., HBOI/DBMR # 3-VI-84-3-001, from deep water (115-126 m), off Grand Bahama Isl. - A. Strongyle. - B-E. Apices of strongyles showing incipient or vestigial spination and "malformations".

Choanosome
Plumoreticulate, with irregularly anastomosing tracts. Spongin not visibly developed, tracts rather loose.

Spicules (Fig. 3)
Strongyles, isodiametric with equally shaped apices, many rather bluntly ending. Under SEM many show a faint apical microspination (Fig. 3A, B, C),

but entirely smooth apices are also found. Quite a few malformed, shepherd-staff or crooked strongyles occur (Fig. 3d). Size uniformly 315-350 x 3-5 µm; definitely no differentiation into categories.

Chemistry (Table 2)
SAKEMI *et al.* (1989) and SUN *et al.* (1990) reported pyrroloquinoline alkaloids [batzellines A-C (**20-22**) and isobatzellines A-D (**23-26**)] from this sponge.

OTHER SPONGES PRODUCING PYRROLOQUINOLINE ALKALOIDS (Table 2)

STIERLE & FAULKNER (1991) reported similar molecules [damirones A-B (**27-28**)] from a Palauan sponge identified as *Damiria* sp. by R.W.M.VAN SOEST (later re-identified as *Zyzzya fuliginosa* (CARTER, 1880), cf. VAN SOEST *et al.*, 1994). This is the same species as was studied by RADISKY *et al.* (1993), and these authors indeed found similar compounds (makaluvamines A-F (**29-34**), makaluvone (**35**), discorhabdin A (**37**) and damirone B (**28**)]. And it is also the same species as was studied by CARNEY *et al.* (1993) (an Indonesian specimen identified as *Histodermella* sp. by M. KELLY-BORGES, but re-identified as *Zyzzya fuliginosa* in a personal communication), and these authors found damirones A-B (**27, 28**), makaluvamine A (**29**), C (**31**), and G (**36**). *Zyzzya fuliginosa* is a member of the poecilosclerid family Iophonidae, and at first glance has a spiculation quite different from that of the deep water *Batzella*, as it has verticillated acanthostrongyles, spined tylotes and palmate isochelas (cf. revision in VAN SOEST *et al.*, 1994). Still, two similarities may be pointed out : both have a tangential spicule cover at the surface, and the faint microspination of the strongyles of *Batzella* sp. may be homologous to the spines on the tylote apices.

PERRY *et al.* (1986, 1988a, 1988b) reported the isolation of the pyrroloquinoline alkaloids discorhabdins A-C (**37-40**)] from a presumably distant genus, viz. *Latrunculia* DU BOCAGE, 1870. This genus is well-characterized by its chessman-microscleres. It is usually assigned to a family of its own in the order Hadromerida (cf. BERGQUIST, 1978). However, it has been associated with poecilosclerid sponges by several authors (e.g. LÉVI, 1973), and it may turn out to be a polyphyletic genus.

Finally, four pyrroloquinoline alkaloids (prianosins A-B) have been isolated [KOBAYASHI *et al.* (1987, 1991) and CHENG *et al.* (1988)] from the Okinawan sponge *Prianos melanos* DE LAUBENFELS (1954), identified by T. HOSHINO. Prianosin A is identical to discorhabdin A (**37**) and prianosin D to discorhabdin D (**40**). Prianosin B (**41**) and prianosin C (**42**) are new pyrroloquinoline alkaloids. The identity of this sponge remains obscure. The genus *Prianos* is of uncertain status; it is characterized by a reticulation of strongyles. Close morphological (and phylogenetic) relationship with *Zyzzya* and/or the deep-water *Batzella* sp. is quite possible.

CONCLUSION

The single category of isodiametric strongyles found in the deep-water *Batzella* is shared with *Batzella inops* and this similarity could be explained as evidence for these sponges being congeneric. A distinct difference is the lack of an ectosomal spicule cover in *Batzella inops*. The faintly spined apices of the strongyles could be explained as evidence for generic affinity with *Zyzzya*. Both alternatives lack a firm basis. Until we have SEM studies of the strongyle apices of *Batzella inops* and know its chemistry it remains uncertain how the Bahama deep-water material relates to the genus *Batzella*. We propose to retain the deep-water *Batzella* specimens in that genus for the time being.

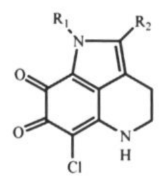
The specimen identified as *Prianos melanos* in the study by KOBAYASHI *et al.* (1987) is likely to be assignable either to the same genus as the deep-water *Batzella* or to *Zyzzya*. The close relationships of the deep-water *Batzella*, *Zyzzya fuliginosa* and *Prianos melanos*, whatever their exact generic status, is obvious.
This cannot be said for the relationship of these sponges with the genus *Latrunculia*. Morphologically, this genus seems quite distinct, and the chemical similarity demands an explanation other than close phylogenetic relationships. PERRY *et al.* (1988b) point out that possibly related compounds are produced by *Amphimedon*, an ascidian and a sea anemone. They suggest that an unknown microsymbiont or food organism may be the producer of the discorhabdins. From underwater observations of *Latrunculia* specimens they conclude that epibionts are absent on the surface of these sponges.

Discussion

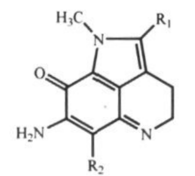
Reduced morphology in many sponges may seriously hamper a correct assignment to families and genera. The chemistry of these sponges may considerably strengthen the generic or specific assignment or in contrast refute it. However, care should be exercised because similar chemistry may be due to symbionts or epibionts. A further problem is that chemistry usually is reported only if "new" compounds have been found. Thus many occurrences of compounds remain unpublished because they were already known from other sponges. Taxonomists identifying sponges for chemists should get a full report on their chemistry and be consulted

Table 2:
Pyrroloquinoline alkaloids recorded from sponges.

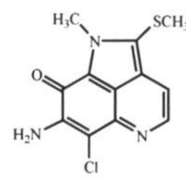
"*Batzella*" sp. deep water	**20-26**
"*Damiria*" sp.	**27,28**
Zyzzya "massalis"	**28-35,41**
"*Histodermella*" sp.	**27-29,31,36**
Latrunculia spp.	**37-40**
"*Prianos melanos*"	**37,40-42**



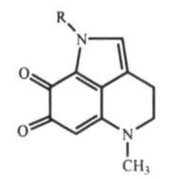
20 Batzelline A; $R_1 = CH_3$; $R_2 = SCH_3$
21 Batzelline B; $R_1 = H$; $R_2 = SCH_3$
22 Batzelline C; $R_1 = CH_3$; $R_2 = H$



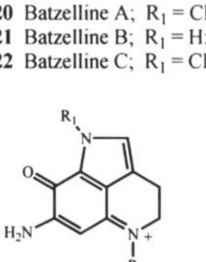
23 Isobatzelline A; $R_1 = SCH_3$; $R_2 = Cl$
24 Isobatzelline B; $R_1 = SCH_3$; $R_2 = H$
25 Isobatzelline C; $R_1 = H$; $R_2 = Cl$



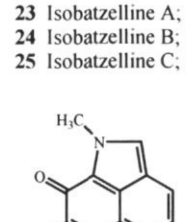
26 Isobatzelline D



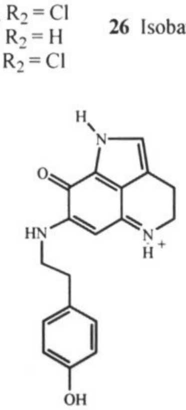
27 Damirone A; R = H
28 Damirone B; R = CH3



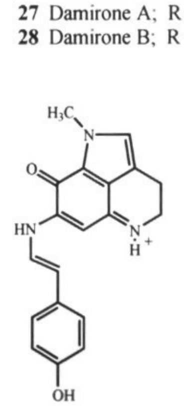
29 Makaluvamine A; R = H; $R_1 = CH_3$
30 Makaluvamine C; $R = CH_3$; $R_1 = H$



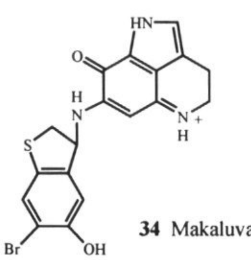
31 Makaluvamine B



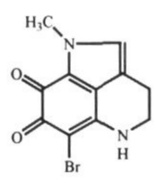
32 Makaluvamine D



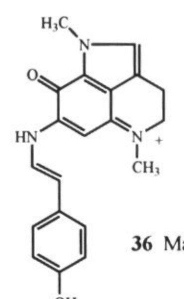
33 Makaluvamine E



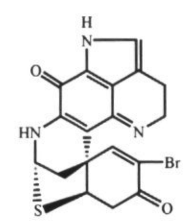
34 Makaluvamine F



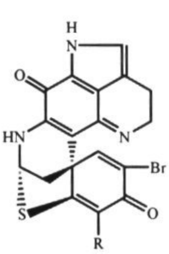
35 Makaluvone



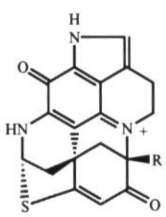
36 Makaluvamine G



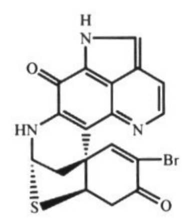
37 Discorhabdin A



38 Discorhabdin B; R = H
39 Discorhabdin C; R = Br



40 Discorhabdin D; R = H
41 Prianosin C; R = OH



42 Prianosin B

whether or not the information has chemosystematic significance. Also, if taxonomists would get feed-back from their chemical colleagues on the biogenetic relationships of the compounds that are found, erroneous identifications or overlooked epi/symbionts would be detected earlier.

Although a full revision still needs to be done, it is increasingly obvious that *Monanchora* and *Crambe* are likely synonyms. The alkaloids reported for *Crambe crambe*, *Monanchora arbuscula*, "*Hemimycale*" sp. and "*Batzella*" sp. are biogenetically related and are not reported from other sponges. Morphologically there are considerable differences between extreme specimens, but through a considerable variability in many intermediate specimens, these are linked. All are united in a basic differentiation into two categories of megascleres arranged in irregularly plumose bundles. Specimens with this reduced spiculation of only megascleres have been assigned to *Crambe crambe*, *Monanchora arbuscula* (and of course "*Batzella*" sp. and "*Hemimycale*" sp.). Additional spicules in this group may include unguiferate chelas (in *Crambe* spp. and *Monanchora* spp.), reduced sigma-like chelas (in *Crambe tailliezi* VACELET & BOURY-ESNAULT (1982), *C. erecta* PULITZER-FINALI (1993), *Monanchora arbuscula*), spined microrhabds [in *Crambe tailliezi*, *C. chelastra* LÉVI (1963), and *Monanchora unguifera* (DE LAUBENFELS, 1953 as *Echinostylinos*)], and desmas [in *Crambe crambe*, *C. tailliezi*, *C. acuata* (LÉVI, 1961), and *C. tuberosa* MALDONADO & BENITO (1991)]. Most species in this group are red, with raised oscules and clear venal canal patterns. The Red Sea "*Hemimycale*" sp. is dark blue-green and has characteristic rounded or angular porefields. Growth forms vary from encrusting (most *Crambe* and *Monanchora* species) to massive-lamellate (*Crambe erecta*) and bushy (*Monanchora arbuscula*).

Based on shared possession of related pyrroloquinoline alkaloids the close relationships of the deep-water *Batzella*, *Zyzzya fuliginosa* and *Prianos melanos* is obvious. The relationship of these sponges with some representatives of the genus *Latrunculia*, which also produce these compounds, remains undecided for the time being. A morphological revision of *Latrunculia* might solve some of the present inconsistencies.

Chemistry has failed to solve the systematic position of the genus *Batzella* because no information on the chemistry of its type species, *Batzella inops*, is currently available. BARROW & CAPON (1992) reported sulfolane from a sponge/tunicate "composite", of which the sponge was identified as *Batzella* sp. However, the spicules of this sponge were described as styles which were erect on the basal spongin layer covering the substrate. This description does not fit with any described *Batzella*, and thus is not relevant for the present study.

Guanidinic and pyrrolic compounds are also known to occur in other sponges (CHEVOLOT, 1985; BRAEKMAN *et al*., 1992), but these are structurally unrelated to the molecules discussed in this paper.

Acknowledgments

B. CARTÉ is thanked for providing the shallow-water Bahamas specimen. A. PATIL (SmithKline) has done the chemistry of the bushy form of *Monanchora arbuscula* (voucher ZMA 11009 from Jamaica). We thank C. LÉVI (Paris), M. ILAN (Tel Aviv), K. SMITH & K. RÜTZLER (Washington), R. DESQUEYROUX-FAÚNDEZ (Geneva) and J. REED (Fort Pierce) for the loan of specimens. J. VERMEULEN (Amsterdam) made the SEM pictures. B. ALVAREZ (Canberra), M.C. DÍAZ (Santa Cruz), B. CARTÉ (San Diego), G.J. BAKUS (Los Angeles), M. KELLY-BORGES (London), S.A. POMPONI & A.E. WRIGHT (Fort Pierce) and S. ZEA (Santa Marta) provided relevant information. The Conselho Nacional of Desenvolvimento Científico e Tecnológico (CNPq), Brazil supported E. HAJDU financially.

References

ALVAREZ, B., VAN SOEST, R.W.M. & RÜTZLER, K., (in press). A revision of the Western Atlantic Axinellidae. *Smithsonian Contributions to Zoology*.

ANDERSEN, R.J., VAN SOEST, R.W.M. & KONG, F., (submitted). 3-Alkylpiperidine alkaloids from marine sponges in the order Haplosclerida.

ARROYO, C., URIZ, M.J., RUBIO, M., 1976. *Inachus thoracicus* (Crustacea Decapoda) substrato pasivo de Demospongia. *Investigacion Pesquera*, 40 (1) : 17-57.

BARROW, R.A. & CAPON, R.J., 1992. Sulfolane as a natural product from the sponge/tunicate composite, *Batzella* sp./ *Lissoclinum* sp. *Journal of Natural Products*, 55 (9) : 1330-1331.

BERGQUIST, P.R., 1978. Sponges. Hutchinson, London, 268 pp.

BERGQUIST, P.R. & WELLS, J.W., 1983. Chemotaxonomy of the Porifera : the development and current status of the field. *In* : SCHEUER, P.J. (Ed.), Marine Natural Products; Chemical and Biological Perspectives. Academic Press, New York, Vol. 5, pp. 1-50.

BERLINCK, R.G.S., BRAEKMAN, J.C., DALOZE, D., HALLENGA, K., OTTINGER, R., BRUNO, I. & RICCIO, R., 1990. Two new guanidine alkaloids from the Mediterranean sponge *Crambe crambe*. *Tetrahedron Letters*, 31 : 6531-6534.

BERLINCK, R.G.S., BRAEKMAN, J.C., DALOZE, D., BRUNO, I., RICCIO, R., ROGEAU, R. & AMADE, P., 1992. Crambines C1 and C2 : two further ichthyotoxic guanidine alkaloids from the sponge *Crambe crambe*. *Journal of Natural Products*, 55 (4) : 528-532.

BRAEKMAN, J.C., DALOZE, D., STOLLER, C. & VAN SOEST, R.W.M., 1992. Chemotaxonomy of *Agelas* (Porifera : Demospongiae). *Biochemical Systematics and Ecology*, 20 : 417-431.

CARNEY, J.R., SCHEUER, P.J. & KELLY-BORGES, M., 1993. Makaluvamine G, a cytotoxic pigment from an Indonesian sponge *Histodermella* spec. *Tetrahedron*, 49 (38) : 8483-8486.

CHENG, J., OHIZUMI, Y., WALCHLI, M.R., NAKAMURA, H., HIRATA, Y., SASAKI, T. & KOBAYASHI, J., 1988. Prianosins B, C and D, novel sulfur-containing alkaloids with potent antineoplastic activity from the Okinawan marine sponge *Prianos melanos*. *Journal of Organic Chemistry*, 53 : 4621-4624.

CHEVOLOT, L., 1981. Guanidine derivatives. *In* : SCHEUER P.J. (Ed.), Marine Natural Products. Academic Press, London, Vol. 4, pp. 54-91.

FROMONT, J., KERR, S., RIDDLE, M. & P. MURPHY, 1994. Chemotaxonomic relationships within, and comparison between, the orders Haplosclerida and Petrosida (Porifera : Demospongiae) using sterol complements. *Biochemical Systematics and Ecology*, 22 (7) : 735-752.

HAJDU, E., VAN SOEST, R.W.M. & HOOPER, J.N.A., 1994. Proposal for a phylogenetic subordinal classification of poecilosclerid sponges. *In* : VAN SOEST, R.W.M., VAN KEMPEN, Th.M.G. & BRAEKMAN, J.C. (Eds), Sponges in Time and Space. Proceedings of the IVth International Porifera Congress. A.A. Balkema, Rotterdam, pp. 123-140.

HARBOUR, G.C., TYMIAK, A.A., RINEHART, K.L., SHAW, P.D., HUGHES, R.G., MIZSAK, S.A., COATS, J.H., ZURENKO, G.E., LI H. LI & KUENTZEL, S.L., 1981. Ptilocaulin and isoptilocaulin, antimicrobial and cytotoxic cyclic guanidines from the Caribbean sponge *Ptilocaulis* aff. *P. spiculifer* (LAMARCK, 1814). *Journal of the American Chemical Society*, 103 : 5604-5606.

HOOPER, J.N.A. & WIEDENMAYER, F., 1994. Porifera. *In*: WELLS, A. (Ed.), Zoological Catalogue of Australia, 12. CSIRO, Melbourne, Australia, xiii + 624 pp.

JARES-ERIJMAN, E.A., SAKAI, R. & RINEHART, K.L., 1991. Crambescidins : new antiviral and cytotoxic compounds from the sponge *Crambe crambe*. *Journal of Organic Chemistry*, 56 : 5712-5715

KASHMAN, Y., HIRSH, S., MCCONNELL, O.J., OHTANI, I., KUSUMI, T. & KAKISAWA, H., 1989. Ptilomycalin A : a novel polycyclic guanidine alkaloid of marine origin. *Journal of the American Chemical Society*, 111 : 8925-8926.

KOBAYASHI, J., CHENG, J.-F., ISHIBASHI, M., NAKAMURA, H., OHIZUMI, Y., HIRATA, Y., SASAKI, T., LU, H. & CLARDY, J., 1987. Prianosin A, a novel antileukemic alkaloid from the Okinawan sponge *Prianos melanos*. *Tetrahedron Letters*, 28 (42) : 4939-4942.

KELLY-BORGES, M., ROBINSON, E.V., GUNASEKERA, S.P., GUNASEKERA, M., GULAVITA, N.K. & POMPONI, S.A., 1994. Species differentiation in the marine sponge genus *Discodermia* (Demospongiae : Lithistida) : the utility of ethanol extract profiles as species-specific chemotaxonomic markers. *Biochemical Systematics and Ecology*, 22 (4) : 353-365.

KOBAYASHI, J., CHENG, J., YAMAMURA, S. & ISHIBASHI, M., 1991. Revised structures of prianosins C and D, antineoplastic alkaloids from the Okinawan marine sponge *Prianos melanos*. *Tetrahedron Letters*, 32 : 1227-1228.

LÉVI, C., 1973. Systématique de la classe des Demospongiaria (Démosponges). *In* : GRASSÉ, P.-P. (Ed.), *Traité de Zoologie : Anatomie, Systématique, Biologie*, 3 (1) : 577-631.

MAI, S.H., NAGULAPALLI, V.K., PATIL, A.D., TRUNEH, A. & WESTLEY, J.W., 1992. Marine sponge *Batzella* polycyclic guanidine alkaloid compounds as HIV virus inhibitors. CA 118 (23) : 225662x. PCT International Application.

PALAGIANO, E., DEMARINO, S., MINALE, L., RICCIO, R., ZOLLO, F., IORIZZI, M., CARRE, J.B., DEBITUS, C., LUCARAIN, L. & PROVOST, J., 1995. Ptilomycalin A, crambescidin 800 and related new highly cytotoxic guanidine alkaloids from the starfishes Fromia monolis and Celerina heffernani. *Tetrahedron*, 51 : 3675-3682.

PATIL, A.D., KUMAR, N.V., KOKKE, W.C., BEAN, M.F., FREYER, A.J., DE BROSSE, C., MAI,S., TRUNEH, A., FAULKNER, D.J., CARTE, B., BREEN, A.L., HERTZBERG, R.P., JOHNSON, R.K., WESTLEY, J.W. & POTTS, B.C.M., 1995. Novel alkaloids from the sponge *Batzella* sp. : Inhibitors of HIV gp120-human CD4 binding. *Journal of Organic Chemistry*, 60 :1182-1188.

PERRY, N.B., BLUNT, J.W., MCCOMBS, J.D. & MUNRO, M.H.G., 1986. Discorhabdin C; a highly toxic pigment from a sponge of the genus *Latrunculia*. *Journal of Organic Chemistry*, 51 : 5476-5478.

PERRY, N.B., BLUNT, J.W. & MUNRO, M.H.G. 1988a. Cytotoxic pigments from New Zealand sponges of the genus *Latrunculia* : discorhabdins A, B and C. *Tetrahedron*, 44 (6) : 1727-1734.

PERRY, N.B., BLUNT, J.W. & MUNRO, M.H.G, HIGA, T., & SAKAI, R., 1988b. Discorhabdin D, an antitumour alkaloid from the sponges *Latrunculia* and *Prianos* sp. *Journal of Organic Chemistry*, 53 : 4127-4128.

PULITZER-FINALI, G., 1978. Report on a collection of sponges from the Bay of Naples. III. Hadromerida, Axinellida, Poecilosclerida, Halichondrida, Haplosclerida. *Bollettino dei musei e degli istituti biologici della (R.) Università di Genova*, 45 : 7-89.

RADISKY, D.C., RADISKY, E.S., BARROWS, L.R., COPP, B.R., KRAMER, R.A. & IRELAND, C.M., 1993. Novel cytotoxic topoisomerase II inhibiting pyrroloiminoquinones from Fijian sponges of the genus *Zyzzya*. *Journal of the American Chemical Society*, 115 : 1632-1638.

RÜTZLER, K., 1965. Systematik und Ökologie der Poriferen aus Littoral-Schattengebieten der Nordadria. *Zeitschrift für Morphologie der Tiere* 55 : 1-82.

SAKEMI, S., SUN, H.H., JEFFORD, C.W. & BERNARDINELLI, G., 1989. Batzellines A, B, and C. Novel pyrroloquinoline alkaloids from the sponge *Batzella* sp. *Tetrahedron Letters*, 30 (19) : 2517-2520.

SARÀ, M., 1958. Studio sui Poriferi di una grotta di marea del Golfo di Napoli. *Archivo di Zoologia Italiana*, 43 : 203-280.

SMITH, V.E. & LAURITIS, J.A., 1969. Cellular origin and fine structure of mesoglea in a marine sponge, *Cyamon neon* DE LAUBENFELS. *Journal of Microscopy*, 8 : 179-188.

SOEST, R.W.M. VAN, 1984. Marine sponges from Curaçao and other Caribbean localities. Part III. Poecilosclerida. *Studies on the Fauna of Curaçao and other Caribbean Islands*, 66 : 1-167.

SOEST, R.W.M. VAN, DESQUEYROUX-FAÚNDEZ, R., WRIGHT, A.D. & KÖNIG, G.M., 1995. *Cymbastela hooperi* n.sp. (Halichondrida; Axinellidae) from the Great Barrier Reef, Australia. *In* : WILLENZ, Ph. (Ed.). Recent Advances in Sponge Biodiversity Inventory and Documentation. *Bulletin de l'Institut royal des Sciences naturelles de Belgique, Biologie*, 66 supplément : 103-108.

SOEST, R.W.M. VAN, ZEA, S. & KIELMAN, M., 1994. New species of *Zyzzya, Cornulella, Damiria*, and *Acheliderma* (Porifera :

Poecilosclerida), with a review of fistular genera of Iophonidae. *Bijdragen tot de dierkunde*, 64 (3) : 163-192.

STIERLE, D.B. & FAULKNER, D.J., 1991. Two new pyrroloquinoline alkaloids from the sponge *Damiria* spec. *Journal of Natural Products*, 54 : 1131-1133.

SUN, H.H., SAKEMI, S., BURRES, N. & MCCARTHY, P., 1990. Isobatzellines A, B, C, and D. Cytotoxic and antifungal pyrroloquinoline alkaloids from the marine sponge *Batzella* sp. *Journal of Organic Chemistry*, 55 : 4964-4966.

TAVARES, R., DALOZE, D., BRAEKMAN, J.C., HAJDU, E., MURICY, G. & VAN SOEST, R.W.M., 1994. Isolation of crambescidin 800 from *Monanchora arbuscula* (Porifera). *Biochemical Systematics and Ecology*, 22 (6) : 645-646.

TAVARES, R., DALOZE, D., BRAEKMAN, J.C., HAJDU, E. & VAN SOEST, R.W.M., 1995. 8b-Hydroxyptilocaulin, a new guanidine alkaloid from the sponge *Monanchora arbuscula*. *Journal of Natural Products*, 58 (7) : 1139-1142.

TOPSENT, E., 1894. Campagne de la Melita, 1892. Éponges du Golfe de Gabès. *Mémoires de la Societé zoologique de France*, 7 : 37-44.

TOPSENT, E., 1928. Les affinités de *Batzella inops* Tops. d'après des spécimens d'Arzew. *Bulletin des Travaux publiés par la Station d'Aquiculture et de Pêche de Castiglione*, 1 : 83-88.

VACELET, J., DONADEY, C. & FROGET, C., 1987. The calcium carbonate spherules of *Hemimycale columella* (Demosponges, Poecilosclerida) and their taxonomic value. *In* : VACELET, J. & BOURY-ESNAULT, N. (Eds), Taxonomy of Porifera from the N.E. Atlantic and Mediterranean Sea. NATO ASI Series G, 13. Ecological Sciences, Springer-Verlag, Berlin, pp. 259-274.

WIEDENMAYER, F., 1977. Shallow-water sponges of the western Bahamas. *Experientia Supplementum*, 28 : 287 pp., + 43 pls.

WIEDENMAYER, F., 1989. Demospongiae (Porifera) from Northern Bass Strait, Southern Australia. *Memoirs of the Museum of Victoria*, 50 (1) : 1-242.

WILLENZ, Ph., 1982. Aspects cinétiques, quantitatifs et ultrastructuraux de l'endocytose, la digestion et l'exocytose chez les éponges. Thèse, Université Libre de Bruxelles, Belgique : 107 pp. + 180 figs.

WILLIAMS, D.E., BURGOYNE, D.L., RETTIG, S.J., ANDERSEN, R.J., FATHI-AFSHAR, Z.R. & ALLEN, T.M., 1993. The isolation of majuscalamide C from the sponge *Ptilocaulis trachys* collected in Enewetak and determination of the absolute configuration of the 2-methyl-3-aminopentanoic acid residue. *Journal of Natural Products*, 56 : 545-551.

WRIGHT, A.E., CHILES, S.A. & CROSS, S.S., 1991. 3-Amino-1-(2-aminoimidazolyl)-prop-1-ene from the marine sponges *Teichaxinella morchella* and *Ptilocaulis walpersi*. *Journal of Natural Products*, 54 : 1684-1686.

Rob W.M. VAN SOEST
Institute for Systematics and Population Biology
(Zoölogisch Museum)
University of Amsterdam
PO Box 94766, 1090 GT Amsterdam
The Netherlands

Jean-Claude BRAEKMAN
Chimie Bio-Organique
Université Libre de Bruxelles
CP 160, 50 Avenue F. Roosevelt, 1050 Brussels
Belgium

D. John FAULKNER
Marine Research Division
Scripps Institute of Oceanography
University of California
La Jolla, CA 92093
USA

Eduardo HAJDU
Departamento de Zoologia
Instituto de Biociências
Universidade de São Paulo
Cx Postal 11461 - 05422-970 São Paulo
Brazil

Mary Kay HARPER
Marine Research Division
Scripps Institute of Oceanography
University of California
La Jolla, CA 92093
USA

Jean VACELET
Centre d'Océanologie
Station Marine d'Endoume
Rue de la Batterie-des-Lions
13007 Marseille
France