MORPHOLOGICAL STUDIES OF *Hapalospongidion macrocarpum* AND *Nemoderma tingitanum* (PHAEOPHYCEAE) FROM THE SALVAGE ISLANDS (MADEIRA ARCHIPELAGO)

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Two species of encrusting brown algae (Phaeophyceae) are newly recorded for the Salvage Islands (Madeira Archipelago) viz. *Nemoderma tingitanum* and *Hapalospongidion macrocarpum*. The species are described, and information is presented concerning their ecology, morphology and geographical distribution in the Atlantic.

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INTRODUCTION

Relatively little attention has been given to the study of the marine macroalgae of the Salvage Islands. Phycological investigations have only been conducted sporadically, mainly by visiting scientists over short periods of time (PICCONE 1884; GAIN & MIRANDE 1912; GAIN 1914; PICKERING & HANSEN 1969; LEVRING 1974; GIL-RODRÍGUEZ et al. 1978; WEISSCHER 1982, 1983; AUDIFFRED & WEISSCHER 1984; AUDIFFRED & PRUD'HOMME VAN REINE 1985; PRUD'HOMME VAN REINE et al. 1994; PARENTE et al. 2000a). Since LEVRING (1974), all the published information has been widely scattered in the literature, making it difficult to complete any biogeographical assessment of the Macaronesian marine algal flora. NETO et al. (2001), however compiled a checklist and bibliography of the marine plants of the Madeira Archipelago which listed a total of 68 species of Phaeophyceae, 42 of them occurring in the Salvage Islands. The latter number was increased to 53 by PARENTE et al. (2000a). When compared to the Canary Islands (125 Phaeophyceae, see HAROUN et al. 2002), this number can be considered low, particularly when the tuft-forming and crustose brown algae are considered. In fact, the checklist of NETO et al. (2001) recorded only six small tufted and two crustose species of brown algae for the Madeira Archipelago.

To provide a better knowledge of the marine algal flora of this archipelago, a scientific expedition was sent to the Salvage Islands in May 1999, under the project "TFCM Macaronesia 2000 - Museo de Ciencias Naturales de Tenerife". Particular attention was given to searching for crustose species of brown algae, and the present paper reports on two new additions.

The archipelago of Madeira, co-ordinates 30° to 33°N and 16° to 17°W, located in the Central Eastern Atlantic Ocean, comprises Madeira, Porto Santo, Deserta Grande, Bugio, and the Salvage group of Selvagem Grande, Selvagem Pequena

and Ilhéu de Fora. Most of the islands are close to each other, except for the Salvage Islands, which are situated about half way between Madeira and the Canaries, coordinates 30º N and 16º W.

The systematic position of the brown crusts is not yet completely understood, and the present understanding of the phylogeny of these species is likely to change as a result of further investigations, in particular molecular studies. The delimitation of families in the crustose brown algae, in particular, remains unresolved. The characteristics used to distinguish the crustose brown algae families such are the number of chloroplasts and the position of reproductive organs are not sufficient to delimitate them (see HOLLENBERG 1969). Accordingly, HOLLENBERG (1969) placed all the crustose brown algae into a single family, the Ralfsiaceae, which he basically characterised as having a pseudoparenchymatous mode of construction and a life history involving either an alternation of isomorphic generations or with only one type of plant body present. Frequently placed in the order Ectocarpales, it was removed from this order by TAN & DRUEHL (1994) based on analysis of the nucleotide sequences of rDNA of *Analipus japonicus* (Harvey) *Wynne* and *Ralfsia fungiformis* (Grunnerus) Setchell et Gardner. These authors did not, however, suggest an alternative ordinal placement. In the present study, in agreement with most authors (HOLLENBERG 1969; WOMERSLEY 1987; SILVA et al. 1996), and until further studies are undertaken, the crustose brown algae are included in the family Ralfsiaceae.

MATERIAL AND METHODS

Sampling was undertaken in the period 22-27 May 1999, in several locations on Selvagem Grande (SG), Selvagem Pequena (SP) and Ilhéu de Fora (IF). Intertidal collections were carried out at Ponta da Atalaia, Enseada da Fonte das Galinhas, Enseada das Pedreiras, Ponta do Inferno (SG), Ponta do Oeste, Ponta do Norte, Ponta de Leste and Ponta dos Garajaus (SP) and on Ilhéu de Fora. Subtidal collections were undertaken on SP in Ponta do Norte (22 m), on IF (20 m) and on SG, in Portinho das Cagarras (20 m), Enseada da Atalaia (25 m), Ilhéu Preto (18 m) and Ponta de Leste (25 m).

Sites were surveyed at random, and collections were made of stones, shells and chiselled portions of bedrock, with obvious brown spots, crusts or small tufts. All collections were numbered and deposited at the University of La Laguna Herbarium (TFC Phyc.) and the University of the Azores. In the laboratory, algae were identified using a combination of stereo and compound microscopes. Where appropriate, measurements were made of cells and other structures using a calibrated micrometer eye piece. Permanent slides were made of all algae collected using glycerol jelly and crystal violet. A reference collection was made by giving each specimen a herbarium code number and storing the algae in 5% buffered formaldehyde seawater solution. In the results, under each species name, the code numbers of the more representative specimens are provided. The systematic arrangement, including taxonomic grouping, follows SILVA et al. (1996). Authors of taxa are given in full, mainly to avoid different abbreviation systems. For all species, data on their morphology, local distribution and ecology are provided.

RESULTS

Ralfsiaceae

Nemoderma tingitanum Schousboe ex Bornet 1898

SEL-99-25a; SEL-99-154c; SEL-99-157b; SEL-99-162a; SEL-99-166c.

Plants epilithic, rarely epizoic, forming, smooth, slightly spongy, quite thick, yellow-brown crusts, often confluent and irregularly spreading to several centimetres, and closely adherent and firmly attached to the substratum, usually without obvious rhizoids. In squash preparations (Fig. 1), plants reveal a basal region of 2-3 layers of cells, with cells quadrate to rectangular in shape, measuring 8-23 µm in length and 6-12 µm in height. These cells give rise to erect, monosiphonous, unbranched filaments up to 22 cells (593 µm) in length, easily

separable under light pressure. Central cells of erect filaments commonly rectangular, measuring 13-33 x 8-16 µm, with cells 1.25-4 x longer than wide; upper cells more quadrate, slightly elongate, with pyriform apical cells, measuring 9-18 x 7-13 µm. Cells light coloured with several small discoid plastids without obvious pyrenoids. Hairs abundant, grouped and emerging from pits. In the older crusts, the lower cells appear slightly colourless. The specimens observed were sterile.

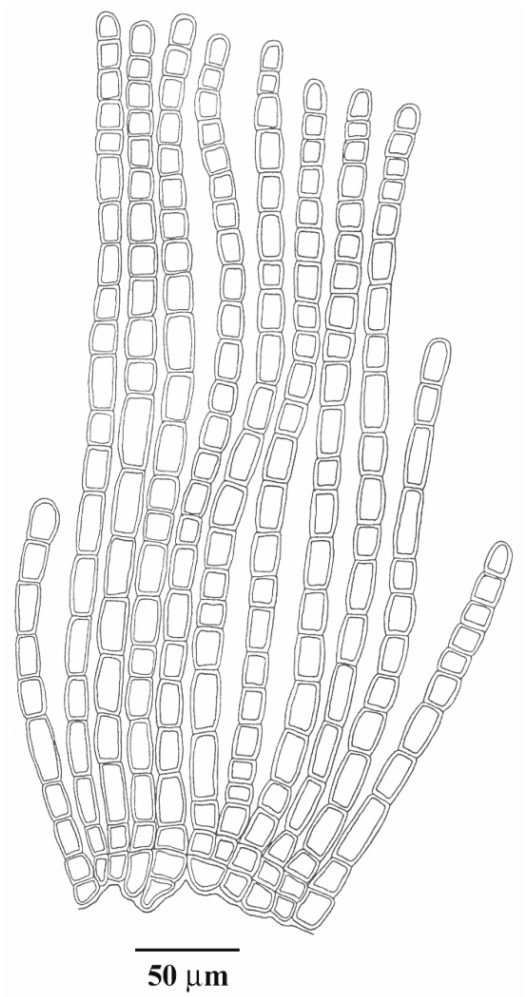

Fig. 1. *Nemoderma tingitanum*: squash preparation showing erect vegetative filaments.

Plants were found covering the bedrock in the mid-to low-tidal region of exposed shores on Selvagem Grande, Selvagem Pequena and Ilhéu de Fora.

In summary, the main diagnostic features of the Salvage Island's material are as follows: crusts yellow-brown in colour, soft and gelatinous, comprising erect, unbranched filaments, easily separable under light pressure; filaments cells up to 16 µm in diameter, with numerous discoid plastids.

Hapalospongidion macrocarpum (Feldmann) León Álvarez & González 1993

SEL-99-25b; SEL-99-154; SEL-99-157a; SEL-99-162b.

Plants epilithic, irregular and confluent, up to 5 cm in extent, forming quite thick, brown crusts, adhering firmly to the substratum. In squash preparations (Fig. 2), the crusts comprise a basal region of three to four layers of adjoined quadrate to rectangular cells, measuring 7-18 µm in length and 4-8 µm high. This basal region gives rise to straight, erect, free, unbranched, monosiphonous filaments up to 69 cells (670 µm) in length, which are loosely attached by mucilage and easily separated under light pressure. These filaments are slightly elongate-clavate in appearance, with the lower and mid-region cells either quadrate on rectangular in shape, measuring 6-17 x 4-8 µm, with cells 1-2.5 longer than wide, and the upper region cells being distinctly subglobose in shape, measuring 8-13 x 9-12 µm, with cells 1-1.25 x longer than wide. Erect filament cells quite thick-walled, especially in the upper subglobose regions, occasionally showing sheath-like remains, with each cell possessing 1, rarely 2, plate-like plastids. Mature unilocular sporangia oblong/elongate-oblong in shape, measuring 120-144 x 36-42 µm, arising laterally from the basal regions of the free filaments and on pedicels of 11-22 cells (max. 284 µm) long (Fig. 3); pedicel cells usually rectangular is shape measuring 9-18 x 5-9 µm. Plurilocular sporangia were not seen.

Plants were common at Selvagem Grande, Selvagem Pequena and Ilhéu de Fora, occurring in the lower part of the intertidal on bedrock moderately exposed to wave action and in pools.
In summary, the main diagnostic features of this crust in the Salvage Islands are as follows: crusts

brown and strongly mucilaginous; erect filaments elongate-clavate in shape, up to 69 cells in length and readily separating under light pressure; unilocular sporangia terminal on long pedicels, arising laterally in the basal regions of the erect filaments, without associated paraphyses.

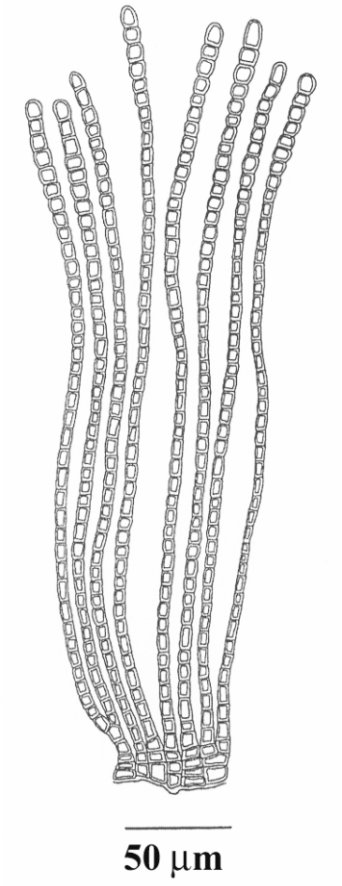

Fig. 2. *Hapalospongidion macrocarpum* squash preparations showing erect vegetative filaments.

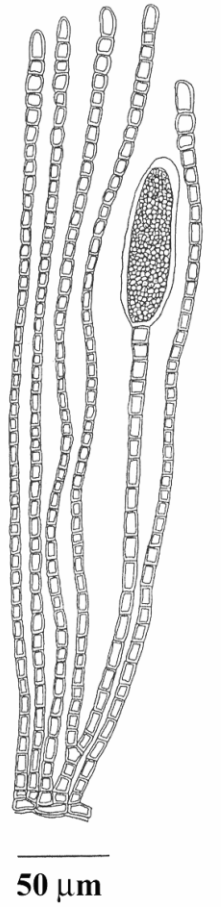

Fig. 3. *Hapalospongidion macrocarpum* squash preparations showing unilocular sporangia on stalks.

DISCUSSION

The two new records described here increase the known number of marine brown algal crusts of the Salvage Islands to 4; only *Ralfsia bornetii* Kuckuck (included within *Stragularia clavata* phase, FLETCHER 1987) and *Ralfsia verrucosa* (Areschoug) Areschoug have previously been recorded. The brown algal flora now totals 120 species.

It is not surprising to record *Nemoderma tingitanum* from the Salvage Islands. This species is known from the Canary Islands (AFONSO-CARRILLO & SANSÓN 1999; HAROUN et al. 2002), Morocco (DANGEARD 1949) and from the Mediterranean (COPPEJANS 1972; SOTO & CONDE 1989), and was recently reported for the Azores by PARENTE et al. (2000b), which extended its northern limit of distribution in the Atlantic. Its presence in the Salvage Islands, therefore, lies within its known distributional range. It seems strange that this species has not been reported from previous expeditions in view of the extent of spread of the yellow-brown crusts observed in some localities.

The material collected from the Salvage Islands agrees well with the descriptions and drawings given by KUCKUCK (1912) for material

collected from Tangier, Morocco, by HAMEL (1935), for Mediterranean (France) specimens, and by PARENTE et al. (2000b) for Azorean specimens. The only difference relates to the thickness of the thallus, which is smaller in the Salvage Islands material when compared to the specimens described from other localities.

The other soft crust collected from the Salvage Islands has been provisionally placed in the genus *Hapalospongidion* SAUNDERS (1899). *Hapalospongidion* is characterised by the following combination of characters; a crustose thallus, comprising erect, gelatinous, loosely associated, sparsely branched filaments, with terminal unilocular sporangia, and intercalary plurilocular sporangia in the upper part of the erect filaments. *Hapalospongidion* is easily confused with the genus *Basispora*, which is only known from the West African coast (JOHN & LAWSON 1974) and the genus *Mesospora*, which is distributed in warm temperate waters (WEBER VAN BOSSE 1911). The only consistent difference between *Hapalospongidion* and these two genera is the presence in the former of terminal unilocular sporangia on the erect filaments (see Tab. 1). Other diagnostic features commonly used to separate these genera are the number of plastids (one, or several per cell) and the morphology of the vegetative filaments (free and straight as opposed to free and upwardly curved). However, these characteristics were not consistently observed by different authors for the same species (WOMERSLEY 1987). Based on these inconsistent observations, this author suggested that *Basispora* and *Mesospora* should be relegated to the synonymy of *Hapalospongidion*. More recently LLUNCH (2002) disagreed and proposed that *Basispora* should be kept separate until the number and morphology of chloroplasts be elucidated. According to the author, the genus *Hapalospongidion* is characterized by the presence of one to a few blade-like chloroplasts rather than several discoid chloroplasts as originally described to *Basispora* by JOHN & LAWSON (1974).

Apart from the apparent smaller size of the erect filaments of *Basispora* and *Mesospora* spp., and the smaller number (1-2) of pedicel cells given for *Mesospora macrocarpa* (Tab. 1), there are no distinguishing features that allow a positive identification of the Salvage Islands material. This major discrepancy in the pedicel number of cells between Selvages material and the FELDMANN (1931) description suggests a different morphology. However, pending further work, notably molecular studies, the Salvage Island´s material was provisionally identified as *Hapalospongidion macrocarpum*, which is the oldest name for this crust in this part of the world.

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Table 1
Principal characteristics used to distinguish species of the genera Basispora, Hapalospongidion and Mesospora.

Species name	Localities	Source	Crust diameter (cm)	No of basal layers	Erect filaments			No of Plastids	Reproductive features			
					Maximum length (µm)	No of cells	Cells diameter (µm)		US (µm)	Position of US	No cells of US Pedicel	PS (µm)
Hapalospongidion sp.	Salvage Islands	Present study	5	3-4	670	> 69	7-12	1-2?	120-144 x 36-42	Lateral	11-22	Not observed
Hapalospongidion saxigenum	New Zealand	LINDAUER (1949)		2-3	725	> 60	6-18		105-170 x 36-45	Terminal		Unknown
Hapalospongidion gelatinosum	California	SAUNDERS (1899)	1	2	750		4-10	Several		Terminal		
‘’	‘’	ABBOTT & HOLLENBERG (1976)	10 or more	2	750		7-10	Mostly one	80-140 x 25-35			60-200 x 20-25
Hapalospongidion capitatum	Australia	WOMERSLEY (1987)	Few to one	2-3	750	40-60	3-12	1-few		US unknown		40-60 (-80) x 15-20
Mesospora macrocarpa (as mediterranea)	Mediterranean France	FELDMANN (1935)	1-2	2-4	550	20-25	10-15			Lateral	1-2	45-60 x 70-100 (140-200)
Mesospora schmidtii	Malaysian Archipelago	WEBER-VAN BOSSE (1911)	1-2	2-4	299*	8-20	4-10			Lateral		140 x 48
Mesospora schmidtii	Japan	TANAKA & CHIHARA (1982)	0.5-1.5	1-4	310**	20-32	5-13	1-3	80-190 (-220) x 30-35 (-38)	Lateral	5-12	30-50 x 10-15
Basispora africana	West African Coast	JOHN & LAWSON (1974)		3-several	247***	> 40?	8-11	Several	70-106 (-125) x 15-33 (-45)	Lateral	(4-) 6-10 (-15)	PS unknown

* Data obtained from Fig.43 (WEBER VAN-BOSSE 1913). ** Data obtained from Fig.7 (TANAKA & CHIHARA 1982). *** Data obtained from Fig.2 (JOHN & LAWSON 1974).

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