

ITS1 sequences of type specimens of *Gigartina* and *Sarcothalia* and their significance for the classification of South African Gigartinaceae (Gigartinales, Rhodophyta)

JEFFERY R. HUGHEY¹, PAUL C. SILVA² AND MAX H. HOMMERSAND¹

¹Department of Biology, University of North Carolina, Coker Hall CB #3280, Chapel Hill, NC 27599-3280, USA

²Herbarium, University of California, Berkeley, CA 94720-2465, USA

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Uncertainties about the identity of type specimens of red algae have frequently led to taxonomic and nomenclatural confusion. A procedure for extracting PCR-amplifiable DNA from formalin-fixed material and herbarium specimens was used to investigate the taxonomic status of several South African Gigartinaceae. We compared nucleotide sequences in the internal transcribed spacer 1 (ITS1) region in type specimens and other historically important collections presently referred to *Gigartina bracteata*, *G. radula* and *G. stiriata*. The following opinions are supported: (1) *Fucus bracteatus* S.G. Gmelin, *Chondrodictyon capense* Kützing and *Iridaea clathrata* Decaisne represent a single species of South African *Gigartina* (*G. bracteata* (S.G. Gmelin) Setchell & Gardner) in which the disintegration of tetrasporangial sori results in a reticulate thallus. (2) *Mastocarpus polycarpus* Kützing, *M. incrassatus* Kützing and *Iridaea lapathifolia* Kützing represent a single species of South African *Gigartina* (*G. polycarpa* (Kützing) Setchell & Gardner) that has often, but erroneously, been called *G. radula* (Esper) J. Agardh. (3) *Mastocarpus verrucosus* Kützing is a later heterotypic synonym of *Iridaea papillosa* Bory (*Sarcothalia papillosa* (Bory) Leister) and was based on material that probably came from southern South America rather than from South Africa, the provenance given by Kützing. (4) *Fucus stiriatus* Turner and *Sphaerococcus burmannii* C. Agardh represent a single species of South African *Sarcothalia* (*S. stiriata* (Turner) Leister).

Key words: DNA, Gigartinaceae, *Gigartina*, *Sarcothalia*, type material, ITS1, PCR, red algae, South Africa

Introduction

During the brief history of molecular systematics, protocols have been developed for the purpose of extracting and amplifying sequenceable DNA. These protocols vary from one taxonomic group to another, but almost all entail extraction of DNA from fresh tissues or from tissues prepared specifically for DNA extraction by being dried in the air or using silica gel. The need to develop protocols for extracting and amplifying useful DNA from herbarium specimens became apparent in studies of population genetics requiring a large number of samples and in phylogenetic studies for which certain taxa were not otherwise readily available. Pioneering work was done as early as 1985 (Rogers & Bendich, 1985), and now herbarium specimens are routinely used as a source of DNA in phylogenetic studies of flowering plants. Success using herbarium specimens of fungi was reported as early as 1990 (Bruns *et al.*, 1990). Phycologists have generally considered it futile to attempt to extract

useful DNA from herbarium specimens of algae, many of which, prior to being dried, were immersed in formalin for varying lengths of time, a method of preservation shown to be highly destructive of DNA (Pyle & Adams, 1989). Goff & Moon (1993) reported success when using genomic material extracted from a cystocarp taken from a specimen of *Gracilaria* that had been collected 11 years earlier and not fixed in formalin. Those authors obtained sequences from the ribosomal ITS region and the plastid Rubisco spacer region. Brodie *et al.* (1996, 1998) were successful in using herbarium specimens of *Porphyra* collected as early as 1926 but of unstated preservational history. One of the herbarium specimens was an isotype.

In the present paper we give a protocol specifically designed for the extraction and amplification of regions of the chloroplast and nuclear genomes of red algae, using vegetative or reproductive fragments of herbarium specimens without regard for their preservational history. Then, in an attempt to resolve problems entailing certain South African algae that have been assigned to the genus *Gigartina* Stackhouse (Gigartinaceae), we pursue the sugges-

tion made by Taylor & Swann (1994) that 'Disputes about nomenclature might be settled by examination of DNA from type specimens'.

In their preliminary survey of *Gigartina*, Setchell & Gardner (1933) recognized nine South African species. Several of these were considered by Hommersand *et al.* (1993) in their morphology-based generic revision of the Gigartinaceae. Molecular data (sequence analysis of the *rbcL* gene) were provided for two of these species (*G. clathrata* and *G. polycarpa*) using recent collections (Hommersand *et al.*, 1994). The resulting taxonomic and nomenclatural changes, however, were noted but not accepted by Stegenga *et al.* (1997). In the present paper we provide molecular data (nucleotide sequences from the ITS1 region) from various collections, including seven type specimens relevant to these species, for the purpose of confirming or revising taxonomic opinions previously published by Hommersand *et al.* (1993, 1994).

Materials and methods

DNA extraction protocol

Ten to twenty milligrams of dried tissue (~5 mm²) was ground in a 1.7 ml microcentrifuge tube with a pellet pestle in 700 µl of extraction buffer (Dellaporta *et al.*, 1983) containing 100 mM Tris (pH 8.0), 50 mM EDTA, 500 mM NaCl, 10 mM 2-mercaptoethanol (added just before use, 7 µl ml⁻¹ throughout extraction buffer), 50 µl of 20% SDS, 10 µl of 0.1 M dithiothreitol and 4 mg of lyophilized Proteinase K (Boehringer Mannheim, Indianapolis, IN), and incubated at 65 °C for 3 h or overnight. Polysaccharides were removed with 250 µl of potassium acetate (5 M) by incubating on ice for 30 min and centrifuging for 30–40 min at 12000 g. The supernatant (750 µl) was extracted with an equal volume of chloroform in a sterile tube and then centrifuged for 15 min. DNA was precipitated from the aqueous phase (600 µl) with 2/3 volume isopropanol for 1 or more hours at -20 °C, then spun down for 20–30 min and washed with 450 µl of 70% ethanol. Samples were air-dried before resuspending DNA in 100 µl of distilled water or TE (10 mM Tris-Cl, 1 mM EDTA, pH 8.0). A working solution of 10:1 (water:DNA) was prepared for polymerase chain reaction (PCR) in a separate tube. A negative control which contains no organic material was performed with each set of extractions. Vortexing was averted during all steps of the extraction to avoid shearing the DNA.

DNA amplification protocol

Six to twelve microlitres of diluted DNA was added to each 50 µl reaction containing 5 µl of 10× reaction buffer (containing 15 mM magnesium chloride), 10 µl of Q-solution (Quiagen, Valencia, CA), 1–3 µl from each 10 µM primer, 200 µM of each dNTP, and 2.5–5.0 units of *Taq* DNA polymerase. To avoid PCR amplification of fungal contaminants common on herbarium specimens, genus-specific primers were synthesized. The ITS1 region was amplified and sequenced with the following forward

primers (K = T+G, R = A+G): ITS1G *Gigartina*-specific, 5'-GGATCATTCTTAGTGAGATAGC-3'; ITS1S *Sarcothalia*-specific, 5'-GGATCATTCKTAGT-GRGAT-3'. The ITS2 reverse primer, 5'-GCTGCG-TTCTTCATCGATGC-3' (White *et al.*, 1990) was used in all reactions. Reactions were cycled using a PTC-100 PCR Thermocycler (MJ Research, Watertown, MA) with the following parameters: 94 °C for 3 min, followed by 40 cycles of 95 °C for 30 s, 50 °C for 90 s, and 72 °C for 90 s, with a final extension of 72 °C for 5 min. The PCR products were electrophoresed on 1.5% agarose gels for 30 min at 120 V. Positive reactions were purified using the QIAquick PCR Purification Kit following the manufacturer's instructions (Qiagen, Valencia, CA). A negative control was run with each set of PCR reactions to check for contamination. The PCR products were sequenced with Big Dye Terminators (ABI Automated 377, PE Applied Biosystems, Foster City, CA).

The boundaries of the ITS1 region were determined from published sequences in Goff *et al.* (1994) and Van Oppen *et al.* (1995). Sequences were pasted directly into MacDraw Pro 1.5v2 and aligned manually. All references to sequences included gaps. The names, locality data and GenBank numbers of the species sequenced are listed in Table 1.

Results

Sequences obtained for the ITS1 region from holotype or lectotype specimens of species described from South Africa by Turner, C. Agardh, Decaisne and Kützing that are currently referred to *Gigartina bracteata*, *G. radula* or *G. stiriata* (Stegenga *et al.*, 1997) were compared. Our observations (Fig. 1) are summarized as follows:

An ITS1 sequence, length 141 bp, of the holotype of *Iridaea clathrata* Decaisne was identical to that of a recently collected tetrasporangial plant of *Gigartina bracteata* (S.G. Gmelin) Setchell & Gardner and a sterile plant of the same species collected by Menzies in 1779.

ITS1 sequences, length 141 bp, of holotype or lectotype specimens of *Mastocarpus polycarpus* Kützing, *Iridaea lapathifolia* Kützing and *Mastocarpus incrassatus* Kützing and the intended type of *Gigartina kuetzingii* Setchell & Gardner (nom. inval.) were identical.

An ITS1 sequence, length 144 bp, of *Mastocarpus verrucosus* Kützing, said to come from South Africa, differed from that of a recent collection of *Sarcothalia papillosa* from the Falkland Islands by 1 bp. It differed from a South African collection of *S. stiriata* by 4 bp, from a Chilean collection of *S. crispata* (Bory) Leister by 5 bp, and from a New Zealand collection of *S. circumcincta* (J. Agardh) Hommersand by 9 bp.

An ITS1 sequence, length 144 bp, of the lectotype of *Fucus stiriatus* Turner (*Sarcothalia stiriata* (Turner) Leister) was identical to that of the holotype of *Sphaerococcus burmannii* C. Agardh (*Sarcothalia burmannii* (C. Agardh) Kützing). It differed from a

Table 1. Species, authorities, collection and herbarium information, and GenBank accession numbers for taxa analysed in this study

Species and authorities	Collection information	ITS1
<i>Gigartina bracteata</i> (S.G. Gmelin) Setchell & Gardner	Cape of Good Hope, South Africa, specimen collected by A. Menzies labelled <i>G. radula</i> (BM)	AF397430
<i>Gigartina bracteata</i> (S.G. Gmelin) Setchell & Gardner	Swakopmund, Namibia, coll. M.H. Hommersand, 6.vii.1993 (NCU)	AF397431
<i>Gigartina bracteata</i> (S.G. Gmelin) Setchell & Gardner	Africa australi, coll. C.R.A. Leclancher, in 1844 (right-hand specimen on holotype sheet of <i>Iridaea clathrata</i> (PC))	AF397429
<i>Gigartina polycarpa</i> (Kützinger) Setchell & Gardner	Cape of Good Hope, South Africa, coll. Pappe, prior to 1849 (lectotype of <i>Mastocarpus polycarpus</i> Kützinger, here designated: L 938, 334360)	AF397425
<i>Gigartina polycarpa</i> (Kützinger) Setchell & Gardner	Cape of Good Hope, South Africa, coll. Pappe, prior to 1849 (lectotype of <i>Iridaea lapathifolia</i> here designated: L 938, 334384)	AF397428
<i>Gigartina polycarpa</i> (Kützinger) Setchell & Gardner	Cape of Good Hope, South Africa, coll. Pappe, prior to 1849 (lectotype of <i>Mastocarpus papillatus</i> sensu Kützinger: L 938, 334404)	AF397427
<i>Gigartina polycarpa</i> (Kützinger) Setchell & Gardner	Cape of Good Hope, South Africa, coll. Pappe, prior to 1867 (lectotype of <i>Mastocarpus incrassatus</i> Kützinger, here designated: L 938, 334380)	AF397426
<i>Sarcothalia circumcineta</i> (J. Agardh) Hommersand	Wellington, New Zealand, Coll. W.A. Nelson, 23.v.1993	AF397438
<i>Sarcothalia crispata</i> (Bory) Leister	Capelmapu, Chile, coll. M.H. Hommersand, 5.i.1995 (NCU)	AF397437
<i>Sarcothalia papillosa</i> (Bory) Leister	Sea Lion Is., Falkland Is., coll. M.H. Hommersand, 8.i.1998 (NCU)	AF397436
<i>Sarcothalia papillosa</i> (Bory) Leister	Cape of Good Hope, South Africa, coll. R. Hohenacker, prior to 1867 (lectotype of <i>Mastocarpus verrucosus</i> Kützinger, here designated: L 939, 673)	AF397435
<i>Sarcothalia stiriata</i> (Turner) Leister	Dutch Cape Colony, South Africa, coll. unknown, 1700s (lectotype of <i>Fucus stiriatus</i> Turner, here designated: BM; pl. 16, fig. c in Turner 1808, Fig. 4 in this paper)	AF397432
<i>Sarcothalia stiriata</i> (Turner) Leister	Cape of Good Hope, South Africa, coll. Pappe, prior to 1849 (L 938, 334406, as <i>Mastocarpus stiriatus</i>)	AF397434
<i>Sarcothalia stiriata</i> (Turner) Leister	Cape of Good Hope, South Africa, coll. P. Hermann, in 1673 (lectotype, here designated: unnumbered specimen labelled <i>Fucus burmanni</i> , Lam ^x ined. N ^o U. C.B.sp. Burmann, PC-cabinet 251, Fig. 5, upper, this paper)	AF397433

collection identified as *Mastocarpus stiriatus* by Kützinger by 1 bp.

An attempt to amplify DNA from the ITS1 region of syntype material of *Iridaea fissa* Suhr, a species thought to be referable to *Sarcothalia*, failed.

Discussion and taxonomic conclusions

Gigartina bracteata/*Gigartina clathrata*

The most distinctive foliose species of *Gigartina* in South Africa is one in which mature tetrasporangial sori disintegrate centrifugally, leaving holes that gradually enlarge to produce a reticulate thallus (Stegenga *et al.*, 1997: 347, pl. 126: 1, as *G. bracteata*). This alga was first described and illustrated by Seba (1758: 192, pl. CIII: figs. 1–3) in a book in which binomial nomenclature was not used. Seba's treatment of the South African alga, together with Rumphius's treatment of an alga from the Indonesian island of Amboina (Rumphius, 1750: 186), was cited by S. G. Gmelin (1768: 212) in the

protologue of *Fucus bracteatus*. Seba's illustrations were considered to constitute the type of *F. bracteatus* by Setchell & Gardner (1933: 295), but Parkinson (1981: 19) gave arguments for overturning this lectotypification in favour of the Rumphius alga. According to Art. 9.17 of the St Louis Code (Greuter *et al.*, 2000), the first lectotypification of a specific name must be followed unless it can be shown that it is in serious conflict with the protologue. Although the Rumphius alga would arguably be the logical lectotype, it may not replace the Seba illustrations as lectotype since they are an integral part of the protologue and not in conflict with it.

Parkinson, after concluding that the name *Fucus bracteatus* should apply to an Indonesian alga, sought the correct name for the South African alga. He pointed out that the latter was described by Kützinger (1843: 396; 1867: 6, pl. 21) as *Chondrodictyon capense*, the only species in his new genus *Chondrodictyon*, but that the epithet cannot be used in *Gigartina* because of the existence of *Gigartina capensis* (J. Agardh) D. H. Kim (1976: 39), based

Gigartina

	1	37	73
Mas. polycarpus Type	TTAGTGATATAGCAATAGGGGTTTCGGCCGTCTATTGCCTGCTTTTCTAACA	-GTTTCGTGAAACTTTGAA	
Mas. incrassatus Type
Mas. papillatus S.A.
Ird. lapathifol. Type
Ird. clathrata TypeG.....C.....T.....T.....C.....		
Gig. bracteata S.A.G.....C.....T.....T.....C.....		
Gig. bracteata NamibiaG.....C.....T.....T.....C.....		
	74	110	145
Mast. polycarpus Type	ACCATATTTTTTTTACAAC-A	AATACAACCCAAACAACAACAATAAATAATTCTTAACAGAAAAAAA	
Mast. incrassatus Type
Mast. papillatus S.A.
Ird. lapathifol. Type
Ird. clathrata TypeT.TT.....T.....C.....		
Gig. bracteata S.A.T.TT.....T.....C.....		
Gig. bracteata NamibiaT.TT.....T.....C.....		

Sarcothalia

	1	37	73
Fcs. stiriatus Type	GTAGTGGGATGGTTAGTTGGAATCGGCGATTCCGCCGTTTTC	-AGCCTACCTTTCTATCATGTTTCGAGAACA	
Sph. burmanni Type
Mas. stiriatus S.A.
Mas. verrucosus Type
Sar. papillosa Falklands
Sar. crispata ChileT.....		
Sar. circumcincta N.Z.T.....A.....T.....		
	74	110	147
Fucus stiriatus Type	AATCTGACCTTTTTT-CTAAACTACTAAAACACTAAACAAAAACCATAACAAACTTTTATAAC-AAACATAAA		
Sph. burmanni Type
Mas. stiriatus S.A.
Mas. verrucosus Type	.C.....T.....C.T.....		
Sar. papillosa Falklands	.C.....T.....C.T.....		
Sar. crispata Chile	.T.....C.T.....C.....		
Sar. circumcincta N.Z.	.T.....C.T.....T.....A.C.....C.....		

Fig. 1. Alignments of ITS1 sequences from Gigartinaceae. Dots are identical to uppermost line, dashes indicate gaps. Refer to the Materials and Methods for specific primer sequences and Table 1 for collection data. Mas., *Mastocarpus*; Ird., *Iridaea*; Gig., *Gigartina*; Sph., *Sphaerococcus*; Sar., *Sarcothalia*; S.A., South Africa; Fcs., *Fucus*.

on *Iridaea capensis* J. Agardh (1848: 47). Parkinson also pointed out that the next available epithet for the South African alga is provided by *Iridaea clathrata* Decaisne (1844: 236), a name applied to a species that was transferred to *Gigartina* by Rabenhorst (1878: 71). There is no indication that anyone subsequent to Decaisne has examined the type of this species. Recently, Bruno de Reviers located the type at the Muséum National d'Histoire Naturelle, Paris (PC) (Fig. 2). It consists of a single sheet on which are mounted two specimens collected by C.R.A. Leclancher, the naturalist on board *La Favorite*, during a voyage around the world (1830–1832) led by C.P.T. Laplace. The original label carries the inscription '*Iridaea clathrata* Dne Africa australis' in Decaisne's handwriting. Bornet has annotated the sheet '*Gigartina Radula* γ *clathrata* J. Ag.' and another label, in an unknown handwriting, reads 'Afrique australe M. Leclancher, 1844'.

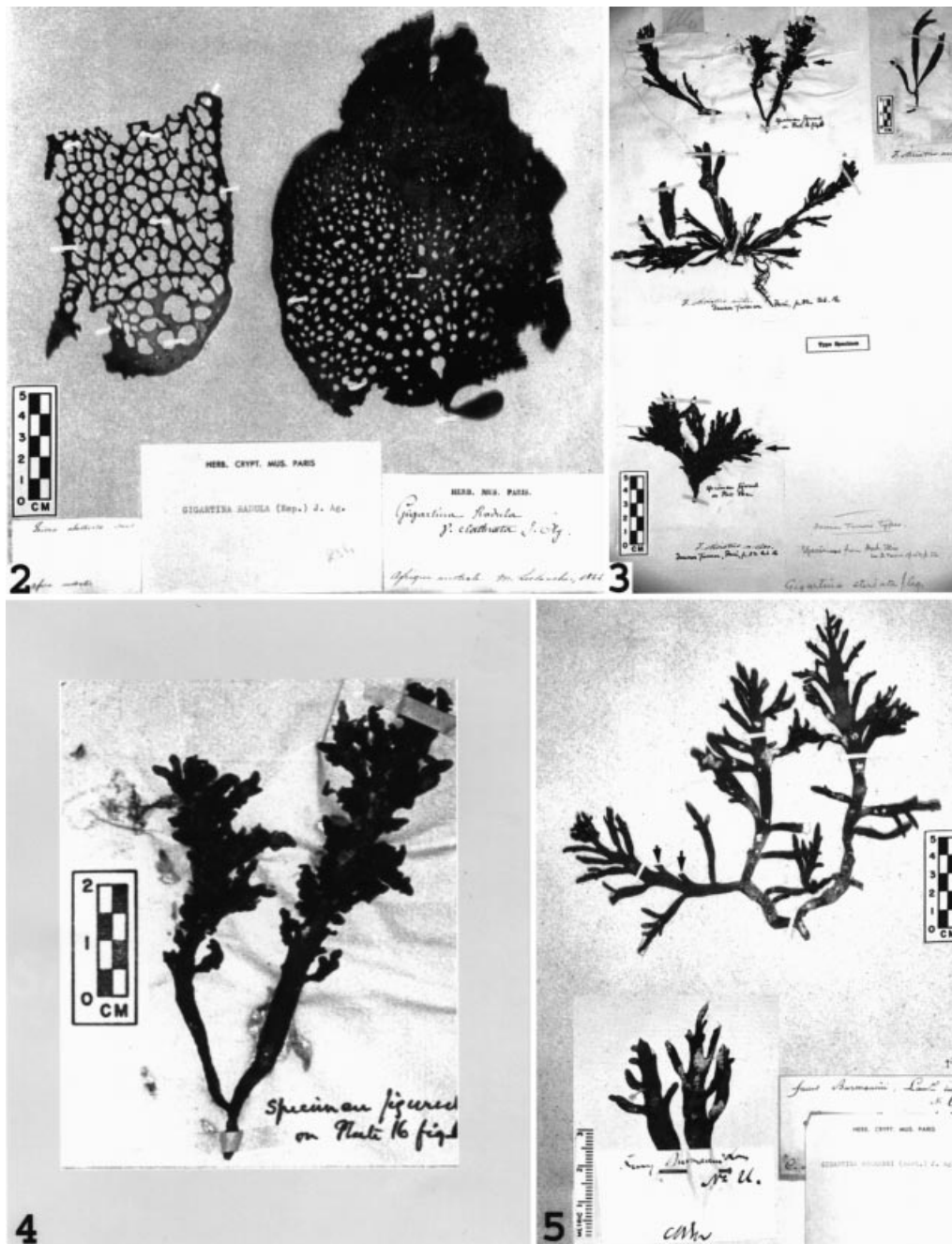
The ITS1 region of the right-hand specimen on the type sheet of *Iridaea clathrata* proved identical in length (141 bp) and nucleotide sequence (Fig. 1) to that of two specimens identified as *G. bracteata* on the basis of morphology: one a recently collected tetrasporangial plant from Namibia, the other a sterile plant at the Natural History Museum (BM) collected at the Cape of Good Hope by Menzies in 1779. The type specimen of *Chondrodictyon capense* Kützing could not be located; however, Kützing's illustration of the type (1867, pl. 21) shows that it is referable to *Gigartina bracteata*. We conclude that

Fucus bracteatus, *Iridaea clathrata* and *Chondrodictyon capense* represent a single species of South African *Gigartina* in which the mature tetrasporophyte is reticulate. *Gigartina clathrata* (Decaisne) Rabenhorst would be the correct name for this species if *Fucus bracteatus* were to be typified with the Indonesian alga, as by Parkinson (1981) and Hommersand *et al.* (1993, 1994, 1999). We now believe, however, that *F. bracteatus* should be typified with the South African alga, so that the correct name for this species is *Gigartina bracteata* (S.G. Gmelin) Setchell & Gardner (1933: 258, 295).

Gigartina radula/*Gigartina polycarpa*

In a second foliose South African *Gigartina*, the tetrasporangial thallus does not become reticulate. The name *G. radula* (Esper) J. Agardh has usually been applied to this species (as by Stegenga *et al.*, 1997: 350, pl. 127.1, col. pl. 49). *Fucus radula* Esper (1802: 3, pl. CXIII) was based on a collection from 'Neuholland' sent by Turner, who later (1808: 54) revealed that the name was found on a specimen in the Banksian Herbarium. Turner treated the two foliose species as one, to which he reluctantly applied 'Gmelin's unmeaning name of *F. bracteatus*' rather than *F. radula*, a name that he considered 'most happy both as to look and touch'.

When J. Agardh (1851: 278) transferred *F. radula* to *Gigartina*, he circumscribed the species to include a variety of forms from South Africa, New Zealand,



Figs 2–5. Habit of South African type specimens that were DNA sequenced in this study. Fig. 2. Type sheet of *Iridaea clathrata* Decaisne (PC, photo by Bruno de Reviere). Fig. 3. Type sheet of *Fucus stiriatus* Turner (BM). The two specimens marked by arrows were illustrated by Turner (1808: pl. 16) (photo by Leister). Fig. 4. Cystocarpic specimen of *F. stiriatus* (upper middle in Fig. 3) here designated as the lectotype (photo by Leister). Fig. 5. Holotype of *Sphaerococcus burmannii* C. Agardh (photo by Bruno de Reviere). Arrows indicate where two branches (shown below), now in the Agardh Herbarium, LD, were removed (photo by Leister).

the Auckland and Campbell islands, and the Pacific coast of North America. Later, J. Agardh (1876: 202) restricted the name to plants from the Cape of Good Hope, despite the fact that the type locality for this species was given as ‘Neuholland’ by Esper. New Zealand plants formerly assigned to *G. radula* were described as a new species, *G. circumcincta* J. Agardh (1876). Parkinson (1981: 22) suggested that the type of *Fucus radula* was probably collected on Cook’s first voyage (1768–1771) and came from New Zealand. Womersley (1994: 293), however,

argued that ‘several other individuals collected in Australia for Banks before 1800, and the type specimen cannot be excluded from Australia on the present evidence, and some Australian specimens agree well with Esper’s illustration’. An intensive search at Erlangen (ER) by Geoffrey Leister (personal communication) failed to locate a specimen that matched Esper’s illustration.

With the exclusion of *Gigartina radula* from the South African flora, Hommersand *et al.* (1994: 198) adopted what they considered to be the earliest

available name for the second of the two South African foliose species of the genus: *G. polycarpa* (Kützinger) Setchell & Gardner (1933: 295). The species to which this name applies was originally described as *Mastocarpus polycarpus* Kützinger (1849: 733) on the basis of a Pappe collection from the Cape of Good Hope. Hommersand *et al.* (1999: 141) showed that *G. polycarpa* is only distantly related to *G. radula* from Australia.

Several other names have been applied to South African algae that closely resemble *G. polycarpa*. To clarify their status, we sequenced the ITS1 region of type specimens of the following names: *Iridaea lapathifolia* Kützinger (1849: 729; 1867: 5, pl. 16) (*Sarcothalia lapathifolia* (Kützinger) Leister in Hommersand *et al.* 1993: 112); *Mastocarpus incrassatus* Kützinger (1867: 12, pl. 41) (*Gigartina incrassata* (Kützinger) Setchell & Gardner (1933: 295)); and *Mastocarpus verrucosus* Kützinger (1867: 12, pl. 42) (*Gigartina verrucosa* (Kützinger) Setchell & Gardner (1933)). We also sequenced a South African specimen that Kützinger had referred to *Mastocarpus papillatus* (C. Agardh) Kützinger (1867: 13, pl. 45, fig. a). Setchell & Gardner (1933) intended to recognize Kützinger's specimen as representative of a new species, *Gigartina kuetzingii*, but the name is invalid because there was no accompanying or cited description. The ITS1 region in all these specimens except the type of *Mastocarpus verrucosus* was found to be 141 bp in length and identical in sequence (Fig. 1). We conclude that *Mastocarpus polycarpus*, *M. incrassatus* and *Iridaea lapathifolia* represent a single species of South African *Gigartina* that has often, but erroneously, been called *G. radula*. Its correct name is *G. polycarpa*.

Identity of Mastocarpus verrucosus

The ITS1 region of the type of *Mastocarpus verrucosus* was 144 bp in length and not alignable with that of *Gigartina polycarpa*. Rather, it was similar in sequence to that in specimens of *Sarcothalia*, differing from the two South African specimens of *S. stiriata* by 4 bp, from a Chilean specimen of *S. crispata* (Bory) Leister by 5 bp, and from a New Zealand collection of *S. circumcincta* (J. Agardh) Hommersand by 9 bp (Fig. 1). Recalling the morphological resemblance of *M. verrucosus* to *S. papillosa* (Bory) Leister (*Iridaea papillosa* Bory) from the Falkland Islands, we sequenced the ITS1 region of a formalin-fixed specimen of the latter and found that it differed from that in *M. verrucosus* by only 1 bp (Fig. 1). We conclude that *M. verrucosus* is a later heterotypic synonym of *S. papillosa* and was based on material that probably came from southern South America rather than South Africa, the provenance given by Kützinger (1867).

Gigartina stiriata/Sarcothalia stiriata

Fucus stiriatus Turner (1808: 32, pl. 16) was based partly on material collected by Thunberg at the Cape of Good Hope and communicated by Mertens under the manuscript name adopted by Turner, and partly on specimens of unknown provenance in the Ellis and Linnaean herbaria. Turner's illustration was based on specimens in the Ellis herbarium. Turner distinguished this species by its ligulate ramuli and gelatinous substance. The type sheet in the Natural History Museum (BM) comprises five specimens, two of which were illustrated by Turner (Fig. 3, arrows). We herein designate the cystocarpic specimen (Fig. 3, upper middle; Fig. 4) as lectotype. *Fucus stiriatus* was transferred to *Mastocarpus* by Kützinger (1843: 398), to *Gigartina* by J. Agardh (1851: 277) and to *Sarcothalia* by Leister (in Hommersand *et al.*, 1993: 112). Stegenga *et al.* (1997: 352) retained the name *Gigartina stiriata*, but an analysis of *rbcL* gene sequences confirms the placement of this species in *Sarcothalia* (Hommersand *et al.*, 1994, 1999).

J. Agardh (1851: 277) suggested that *Gigartina burmannii* (C. Agardh) J. Agardh (1842: 104), originally described as *Sphaerococcus burmannii* C. Agardh (1822: 272), might be the tetrasporangial phase of *Gigartina stiriata*, for which he knew only cystocarpic plants. *Sphaerococcus burmannii* had been segregated by Kützinger (1849: 739) into a unispecific genus, *Sarcothalia*, which was not recognized by J. Agardh.

C. Agardh described *Sphaerococcus burmannii* on the basis of a specimen that he had seen at the Muséum National d'Histoire Naturelle in Paris (PC) during his visit in 1821. The specimen bore the manuscript name *Fucus burmanni* and was said to come from the Cape of Good Hope. Because it was originally part of the J. Burman herbarium, it seems likely that it was collected by the German-born Dutch explorer Paul Hermann (1646–1695), whose collections of South African flowering plants were treated in a publication by Burman (1737). The specimen seen by C. Agardh at PC has been located by Bruno de Reviers and is shown in Fig. 5 (upper). It is labelled 'Fucus burmannii, Lamx ined. No. U C.B.sp. Burmann' in Lamouroux's handwriting and 'Sphaerococcus burmanni AgSyst J. Agardh' in J. Agardh's script. There are two stubs where branches have been removed. Two branches matching the stubs were found by Geoffrey Leister (personal communication) in the Agardh herbarium (LD 23811; Fig. 5, lower). We consider the specimen in PC to be the holotype. Despite Lamouroux's claim to the name *Fucus burmannii*, C. Agardh's protologue implies that the name originated with Mertens, and Kützinger (1849: 739) explicitly accredited the name to Mertens.

J. Agardh (1876: 204) assigned cystocarpic plants to *Gigartina burmannii*, thus erasing his earlier distinction between this species and *G. stiriata*, but nonetheless recognized both species. *Gigartina burmannii* was not mentioned in Setchell and Gardner's monograph of the genus (1933). However, it was considered conspecific with *G. stiriata* by Leister (in Hommersand *et al.*, 1993: 112), which he transferred to *Sarcothalia*.

The ITS1 region from the type specimen of *Fucus stiriatus* was identical to that from the type specimen of *Sphaerococcus burmannii*. It differed from that of a Pappe collection determined as *Mastocarpus stiriatus* by Kützing (1867: pl. 40: fig. a) by only 1 bp (Fig. 1). We conclude that *Fucus stiriatus* and *Sphaerococcus burmannii* represent a single South African species of *Sarcothalia* whose correct name is *S. stiriata* (Turner) Leister.

Setchell & Gardner (1936: 473) thought it probable that both *Iridaea fissa* Suhr (1836: 340, pl. III: fig. 26) and *I. clavellosa* Suhr (1836: 341, pl. III: figs. 27, 28) were tetrasporangial plants of *Gigartina stiriata*, despite the fact that Suhr had indicated their provenance as Cape Horn. For *I. fissa*, Suhr wrote: 'zwischen dem klauigen Wurzelgeflecht der *L. buccinalis* Ag. vom Cap Horn' and for *I. clavellosa* he wrote 'wie die Vorige vom Cap Horn'. Since *Laminaria buccinalis* (L.) Lamouroux (*Ecklonia maxima* (Osbeck) Papenfuss) occurs at the Cape of Good Hope rather than Cape Horn, Suhr may be considered to have erred. Setchell and Gardner did not see the type of the two Suhr species. While we were able to locate syntype material of *I. fissa* (LD 23839), attempts to amplify DNA from the ITS1 region were unsuccessful.

Conclusions

The distinct profiles of the ITS1 sequences reported in this study support the recognition of *Gigartina* and *Sarcothalia* in the South African flora. The correct names for the three species studied are (1) *Gigartina bracteata* (S.G. Gmelin) Setchell & Gardner, (2) *Gigartina polycarpa* (Kützing) Setchell & Gardner, and (3) *Sarcothalia stiriata* (Turner) Leister. A list of specific names accepted in this paper is given in Table 2 together with their homotypic and heterotypic synonyms.

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Table 2. Specific names accepted in this paper with homotypic and heterotypic synonyms

<i>Gigartina bracteata</i> (S.G. Gmelin) Setchell & Gardner (1933)
<i>Fucus bracteatus</i> S.G. Gmelin (1768)
<i>Delesseria bracteata</i> (S.G. Gmelin) Lamouroux (1813)
<i>Sphaerococcus bracteatus</i> (S.G. Gmelin) C. Agardh (1817)
<i>Halymenia bracteata</i> (S.G. Gmelin) Lamouroux (1824)
<i>Laminaria bracteata</i> (S.G. Gmelin) Steudel (1824)
<i>Mastocarpus bracteatus</i> (S.G. Gmelin) Kützing (1849)
Heterotypic synonym:
<i>Chondrodictyon capense</i> Kützing (1843)
Heterotypic synonym:
<i>Iridaea clathrata</i> Decaisne (1844)
<i>Gigartina clathrata</i> (Decaisne) Rabenhorst (1878)
<i>G. radula</i> var. <i>clathrata</i> (Decaisne) J. Agardh (1851)
<i>G. radula</i> f. <i>clathrata</i> (Decaisne) J. Agardh (1876)
<i>Gigartina polycarpa</i> (Kützing) Setchell & Gardner (1933)
<i>Mastocarpus polycarpus</i> Kützing (1849)
Heterotypic synonym:
<i>Iridaea lapathifolia</i> Kützing (1849)
<i>Sarcothalia lapathifolia</i> (Kützing) Leister (in Hommersand <i>et al.</i> , 1993)
Heterotypic synonym:
<i>Mastocarpus incrassatus</i> Kützing (1867)
<i>Gigartina incrassata</i> (Kützing) Setchell & Gardner (1933)
Invalidly published heterotypic synonym:
<i>Gigartina kuetzingii</i> Setchell & Gardner (1933)
<i>Sarcothalia papillosa</i> (Bory) Leister (in Hommersand <i>et al.</i>, 1993)
<i>Iridaea papillosa</i> Bory (1826)
<i>Gigartina papillosa</i> (Bory) Setchell & Gardner (1936)
Heterotypic synonym:
<i>Mastocarpus verrucosus</i> Kützing (1867)
<i>Gigartina verrucosa</i> (Kützing) Setchell & Gardner (1933)
<i>Sarcothalia stiriata</i> (Turner) Leister (in Hommersand <i>et al.</i>, 1993)
<i>Fucus stiriatus</i> Turner (1808)
<i>Delesseria stiriata</i> (Turner) Lamouroux (1813)
<i>Sphaerococcus stiriatus</i> (Turner) C. Agardh (1817)
<i>Halymenia stiriata</i> (Turner) Lamouroux (1824)
<i>Sphaerococcus radula</i> var. <i>stiriatus</i> (Turner) Rudolphi (1831)
<i>Mastocarpus stiriatus</i> (Turner) Kützing (1843)
<i>Iridaea stiriata</i> (Turner) J.D. Hooker & Harvey (1845)
<i>Gigartina stiriata</i> (Turner) J. Agardh (1851)
<i>Mammillaria stiriata</i> (Turner) Kuntze (1891)
Heterotypic synonym:
<i>Sphaerococcus burmannii</i> C. Agardh (1822)
<i>Gelidium burmannii</i> (C. Agardh) Greville (1830)
<i>Gigartina burmannii</i> (C. Agardh) J. Agardh (1842)
<i>Sarcothalia burmannii</i> (C. Agardh) Kützing (1849)
<i>Mammillaria burmannii</i> (C. Agardh) Kuntze (1891)
<i>Gigartina radula</i> (Esper) J. Agardh (1851)
<i>Fucus radula</i> Esper (1802)
<i>Sphaerococcus radula</i> (Esper) C. Agardh (1822)
<i>Iridaea radula</i> (Esper) Bory (1828)
<i>Mastocarpus radula</i> (Esper) Kützing (1843)
<i>Mammillaria radula</i> (Esper) Kuntze (1891)
<i>Sarcothalia radula</i> (Esper) Edyvane & Womersley (in Womersley, 1994)
<i>Sarcothalia circumcincta</i> (J. Agardh) Hommersand (in Hommersand <i>et al.</i>, 1993)
<i>Gigartina circumcincta</i> J. Agardh (1876)
<i>Mammillaria circumcincta</i> (J. Agardh) Kuntze (1891)

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