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Resurrection of *Plocamium pusillum* Sonder (Plocamiaceae, Rhodophyta) from Australia

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ABSTRACT
Molecular analyses have indicated that several Australian taxa previously identified as the red alga *Plocamium cartilagineum* do not group with that species as known from its northern hemisphere type locality. One of these species-level genetic groups corresponds precisely with *Plocamium pusillum*, a species described by Sonder in 1845 based on Preiss specimens from south-western Australia, but regarded as a heterotypic synonym of *P. cartilagineum* in recent morphology-based treatments. A specimen of *P. pusillum* in the Melbourne Herbarium (MEL) has been examined and designated as lectotype; its morphology agrees with recently collected specimens included in molecular analyses and is clearly shown to differ from authentic *P. cartilagineum*. *Plocamium pusillum* is therefore resurrected and is characterized genetically and morphologically.

KEY WORDS
Plocamiaceae,
Plocamium cartilagineum,
resurrected species,
Australia.

RÉSUMÉ
Résurrection de Plocamium pusillum *Sonder (Plocamiaceae, Rhodophyta) en Australie.*
Les analyses moléculaires ont indiqué que plusieurs taxons australiens précédemment identifiés comme l'algue rouge *Plocamium cartilagineum* ne se regroupent pas avec cette espèce telle que connue dans sa localité type de l'hémisphère nord. L'un de ces groupes génétiques au niveau de l'espèce correspond précisément à *Plocamium pusillum*, une espèce décrite par Sonder en 1845 sur la base de spécimens de Preiss du sud-ouest de l'Australie, mais considérée comme un synonyme hétérotypique de *P. cartilagineum* dans les traitements récents basés sur la morphologie. Un spécimen de *P. pusillum* dans l'herbier de Melbourne (MEL) a été examiné et désigné comme lectotype ; sa morphologie est en accord avec les spécimens récemment collectés inclus dans les analyses moléculaires et il est clairement démontré qu'il diffère de l'authentique *P. cartilagineum*. *Plocamium pusillum* est donc ressuscité et est caractérisé génétiquement et morphologiquement.

MOTS CLÉS
Plocamiaceae,
Plocamium cartilagineum,
espèces ressuscitées,
Australie.

INTRODUCTION

Plocamium J.V.Lamouroux is a distinctive red algal genus common on Australian coasts, at its most diverse in the colder southern waters but also with two species, *P. hamatum* J.Agardh and a novel entity (which will be dealt with in a later study), found on the east coast. The southern Australian species of *Plocamium* were monographed by Womersley (1971), his account then included without change in his broader southern Australian flora (Womersley, 1994). One species recorded by Womersley (1971) as being widely distributed on the southern Australian coast (and "in most temperate and cold waters of both northern and southern hemispheres") was *Plocamium cartilagineum* (Linnaeus) P.S.Dixon, a species originally described from northern Europe.

Later studies incorporating DNA sequence analyses (Saunders & Lehmkuhl 2005; Cremades *et al.* 2011), have demonstrated a considerably more restricted distribution for *P. cartilagineum*, excluding it from the Australian flora and prompting a reappraisal of the taxon as interpreted by Womersley (1971). Saunders & Lehmkuhl (2005), who recognized significant cryptic diversity amongst specimens attributed to *P. cartilagineum*, noted in respect to Australia "…there are at least two cryptic entities, neither associated with *P. cartilagineum sensu stricto*, suggesting that a similar situation with respect to cryptic species in '*P. cartilagineum*' exists in Australia as in Europe and further investigation is necessary". Cremades *et al.* (2011) increased the potential number of genetic species in Australian '*P. cartilagineum*' to four.

Included in the synonymy of *P. cartilagineum sensu* Womersley is *Plocamium pusillum* Sonder, a species based on a Johan Preiss collection from south-western Australia. Preiss was a German collector who visited the south-west coast in 1838-1842 (Marchant 1990), collecting numerous algal specimens that were forwarded to Otto Sonder, who, from a collection of 200 species, described 84 that were new to science. Unfortunately, Sonder did not include detailed locality information, ascribing the entire Preiss collection to 'ad oras occidentales Novae Hollandiae' [= coasts of Western Australia]. Thus, we are unable to pinpoint the exact type locality of *P. pusillum*, but we do know that Preiss only collected from the south-west region (including what is now Perth). The Sonder Herbarium, including the Preiss algal collection and the presumed type of *P. pusillum*, was purchased by the National Herbarium of Victoria (MEL) after Sonder's death. This type specimen (MEL 1005815, Fig. 1) is immature and not reproductive, but it nevertheless corresponds morphologically to specimens in the Western Australian Herbarium (PERTH) and to one of the two cryptic Western Australian '*P. cartilagineum*' recognized in molecular analyses (Saunders, unpub. obs.). Womersley (1971) noted that the type of *P. pusillum* had 'ramuli in series of 2 or 3' but was 'similar to better developed specimens in PERTH which agree well with *P. cartilagineum*'. Based on molecular analyses, there are two entities involved, one corresponding to the smaller species with ramuli in series of 2 or 3, and the second a larger plant with ramuli in series of 3-4 (or more, Womersley reports up to 10).

The first of these is compatible with *P. pusillum* Sonder, which we therefore resurrect from synonymy and apply the name to specimens previously represented in the Barcode of Life Data Systems (BOLD; see Ratnasingham & Hebert 2007 for further information) as 'sp. LH2', but now updated to reflect our interpretation. We also provide further morphological observations of the species based on recent collections. The second entity will be described separately as part of a larger study that will examine several other currently un-named species-level genetic groups in Australian *Plocamium* (see: http://v3.boldsystems.org/index.php/TaxBrowser_Taxonpage?taxid=53916).

MATERIAL AND METHODS

Specimens used in our molecular analyses are listed in Table 1. Collections were processed for a voucher, typically either as a press or in silica, with a subsample placed in silica gel for DNA extraction (Saunders & McDevit 2012). Specimens were initially screened with the 5' end of the cytochrome c oxidase subunit I gene (COI-5P), or when this marker failed the 3' end of the ribulose-1, 5-biphosphate carboxylase large subunit gene (*rbc*L-3P) as detailed in Saunders & Moore (2013). These markers were used to assign specimens to genetic groups. To place the previous genetic groups in a phylogenetic context, a longer *rbc*L fragment, as well as the nuclear large subunit ribosomal DNA gene (LSU) were amplified for one representative of each group. Three single-gene alignments were generated: COI-5P, 18 species at 661 base pairs; *rbc*L, 17 species at 1358 base pairs; and LSU, 18 species at 2820 base pairs. All sequences have been submitted to GenBank (Table 1). Each alignment was subjected to maximum likelihood analyses in Geneious 2021.0.3 (https://www.geneious.com; Kearse *et al.* 2012) using RAxML (Stamatakis 2014; model GTR+I+G) with partitioning by codon for the two protein coding genes, and 500 bootstrap replicates. No conflicts were detected and a concatenated alignment was constructed for the three genes (18 species, 4056 base pairs) and analysed as for the single gene alignments, but with partitioning by gene and codon and with 1000 bootstrap replicates. The phylogeny was rooted based on the earlier study of Cremades *et al.* (2011), as well as initial maximum likelihood analyses (multigene as outlined previously) including 28 species of *Plocamium* with six representative Pseudoanemoniaceae and Sarcodiaceae included as outgroups (not shown).

Voucher specimens are lodged in the Western Australian Herbarium (PERTH) or the Connell Memorial Herbarium, University of New Brunswick (UNB). Materials for morphological examination were sectioned by hand and examined under a Nikon SMZ800 stereo microscope and a Nikon Eclipse 80i microscope. Images were taken with a Nikon DS-L4 camera and arranged using Adobe Photoshop 2021. Figure 3 depicts freshly collected, living material.

RESULTS

Molecular analyses

Ongoing barcode surveys in Australia have uncovered numerous species assignable to the genus *Plocamium* (BOLD, link

TABLE 1. — Specimens used in molecular analyses.

Voucher	Species	Habitat	Collector(s)	Date	Country	Province	Locality	Latitude	Longitude	COI-5P GenB	*rbc*L	LSU
GWS008999	*Plocamiocolax pulvinata* Setchell	Subtidal (13 m) on *Plocamium pacificum* (GWS009000)	G.W. Saunders & B. Clarkston	21.IX.2007	Canada	British Columbia	Bamfield, Scotts Bay	48.835	-125.146	HM918870	–	MW770765
KVL72	*Plocamiocolax pulvinata*	Dredged (20 m)	K.V. Lehmkuhl	–	Canada	British Columbia	Bamfield, Wizard I.	48.858	-125.159	–	–	AY881721
GWS000272	*Plocamium cartilagineum* (Linnaeus) Dixon	Low intertidal on rock	G.W. Saunders	21.VIII.1997	France		Cap Gris-Nez	50.867	1.583	–	–	AY881709
GWS001826	*Plocamium cartilagineum*	Drift as an epiphyte	G.W. Saunders	29.VII.2003	Ireland	Co. Galway	Spiddal	53.247	-9.303	–	MW770783	–
GWS001827	*Plocamium cartilagineum*	Drift as an epiphyte	G.W. Saunders	29.VII.2003	Ireland	Co. Galway	Spiddal	53.247	-9.303	JF271573	–	–
GWS036311	*Plocamium corallorhiza* (Turner) J.D.Hooker & Harvey	Mid intertidal on mussel	K. Dixon & J. Ferreira Costa	22.XI.2014	South Africa	Western Cape	Muizenberg, False Bay	-34.110	18.468	MW770751	MW770778	MW770767
GWS036361	*Plocamium cornutum* (Turner) Harvey	Mid intertidal pool on rock	K. Dixon & J. Ferreira Costa	24.XI.2014	South Africa	Western Cape	Rooi-Els	-34.296	18.814	MW770761	MW770790	MW770774
DHO 0162	*Plocamium fimbriatum* M.J.Wynne			–	Oman	–	Sadah baaai 2, Mirbat, Dhofar	16.989	54.692	JF271587	MW770786	MW770771
GWS000254	*Plocamium lyngbyanum* Kützing	Low intertidal as an epiphyte on algal turf	G.W. Saunders	21.VIII.1997	France	–	Cap Gris-Nez	50.867	1.583	JF271598		AY881708
GWS001840	*Plocamium lyngbyanum*	Low intertidal pool on *Ahnfeltia plicata*	G.W. Saunders	30.VII.2003	Ireland	Co. Louth	Ballangar, near Blackrock	53.949	-6.367	JF271592	MW770775	–
G0167	*Plocamium maggsiae* G.W.Saunders & Lehmkuhl	–	C.A. Maggs	14.XI.1993	United Kingdom	Co. Down, Northern Ireland	St. Johns Point	54.246	-5.667	–	–	AF419141
GWS001754	*Plocamium maggsiae*	Intertidal on rock	C.A. Maggs	9.III.2003	Ireland	Co. Donegal	Doaghbeg, Fanad Head	55.276	-7.634	JF271602	JX969788	–
GWS000255	*Plocamium nanum* G.W.Saunders & Lehmkuhl	Low intertidal on rock	G.W. Saunders	21.VIII.1997	France	–	Cap Gris-Nez	50.867	1.583			AY881710
GWS001206	*Plocamium nanum*	Lowest intertidal on rock	G.W. Saunders	16.VIII.2001	France	–	Pointe du Grouin, east of St. Malo	48.717	-1.85	JF271605	MW770782	–
GWS030467	*Plocamium oregonum* Doty	Low intertidal on rock in sand	G.W. Saunders & K. Dixon	6.VI.2012	Canada	British Columbia	Raspberry Cove, Gwaii Haanas	52.167	-131.084	MW770753	MW770779	–
KVL101	*Plocamium oregonum*	Low intertidal on rock in sand	G.W. Saunders	14.V.2001	United States of America	Oregon	Ecola Pt., Canon Beach	45.919	-123.979	JF271608	–	AY881724
GWS030663	*Plocamium pacificum* Kylin	Subtidal (3 m) on rock	G.W. Saunders & K. Dixon	9.VI.2012	Canada	British Columbia	Bay south of Bowles Point, Gwaii Haanas	52.065	-131.119	MW770755	MW770780	–

TABLE 1. — Continuation.

Voucher	Species	Habitat	Collector(s)	Date	Country	Province	Locality	Latitude	Longitude	COI-5P GenB	*rbc*L	LSU
KVL111	*Plocamium pacificum* Kylin	Dredged	K.V. Lehmkuhl	23.5.2001	Canada	British Columbia	Bamfield, Bradys Beach	48.824	-125.159	MW770754		AY881722
PERTH 08715351	*Plocamium pusillum* Sonder	Intertidal on rock	J. Huisman	15.2.2011	Australia	Western Australia	First Rock, S of Penguin Island, Shoalwater Marine Park	-32.312	115.69	MW899161	–	–
GWS002025	*Plocamium pusillum*	Subtidal (15 m) on reef flats & under overhangs	G.W. Saunders	30.1.2004	Australia	New South Wales	Yellow Rock, Lord Howe I.	-31.515	159.034	MW770752	–	MW770768
GWS016698	*Plocamium pusillum*	Subtidal (4 m) on rock	G.W. Saunders, L. Kraft & K. Dixon	2.2.2010	Australia	Victoria	The Springs, Point Lonsdale	-38.276	144.620	HM918320	–	–
GWS025286	*Plocamium pusillum*	Subtidal (3 m) on algae	G.W. Saunders & K. Dixon	13.XI.2010	Australia	Western Australia	Pt. Peron	-32.271	115.682	MW770758		–
GWS029289	*Plocamium pusillum*	Subtidal (1 m) rock	G.W. Saunders & K. Dixon	2.XII.2012	Australia	Norfolk Island	Slaughter Bay, Norfolk I.	-29.059	167.958	MW770757	MW770785	–
PL 19568	*Plocamium raphelisianum* Dangeard	Subtidal (1 m) rock	J. Cremades	30.VIII.2007	Spain	La Coruna	Cambre, Malpica	43.299	-8.757	JF271614	MW770787	–
GWS018247	*Plocamium* sp. 1 Asia	Subtidal (10 m) on *Sargassum*	G.W. Saunders & H-G. Choi	20.V.2010	South Korea	Jeju	Rocky Reef at Lighthouse 'Point' Piyangdo Island	33.420	126.224	MW770759	MW770788	MW770772
DHO 0302	*Plocamium* sp. 1 Oman	–	–	–	Oman	–	Eagle Bay, Mirbat, Dhofar	16.989	54.692	MW770760	MW770789	MW770773
GWS018215	*Plocamium* sp. 2 *cartilagineum* Asia	Subtidal (10 m) tangled in rhodoliths	G.W. Saunders & H-G. Choi	20.V.2010	South Korea	Jeju	Rocky Reef at Lighthouse 'Point' Piyangdo Island	33.420	126.224	MZ870501	MZ870503	MZ870502
KZN 1752	*Plocamium suhrii* Kützing		O. DeClerck & Cocquyt	–	South Africa	Natal	Mission Rocks	-28.267	32.5	JF271667	MW770777	MW770766
GWS035271	*Plocamium tenue* Kylin	Mid intertidal on rock	G.W. Saunders & K. Dixon	23.VIII.2013	Canada	British Columbia	Point between Tana and Gudal Bays, Haida Gwaii	53.194	-132.589	MW770756	MW770784	MW770770
GWS021790	*Plocamium violaceum* Farlow	Low intertridal, on rock wall in upper surge area	B. Clarkston, K. Hind	18.V.2010	United States	California	Jade Cove	35.913	-121.470	HQ544164	MW770781	MW770769

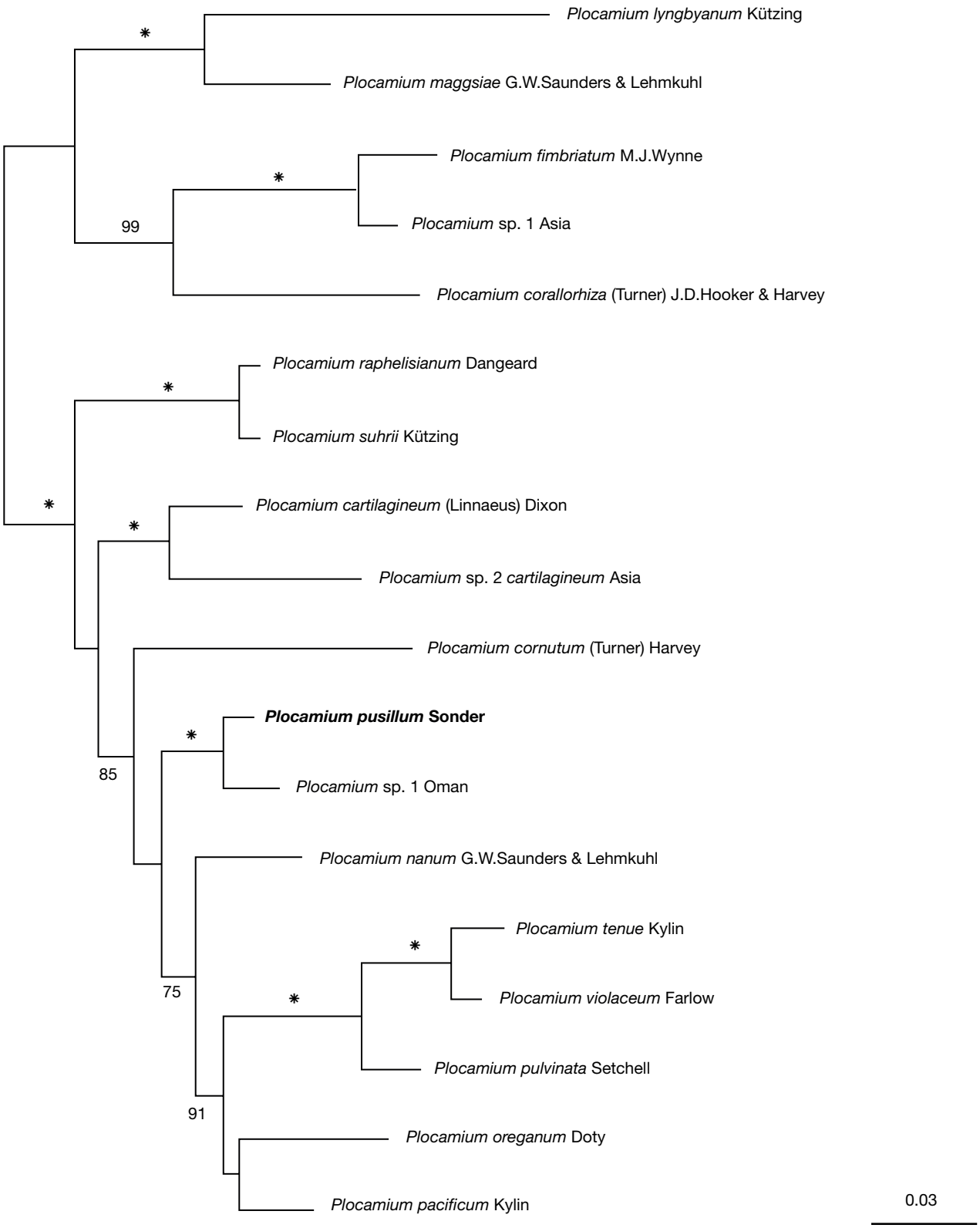

Fig. 1. — Maximum likelihood phylogeny generated for the concatenated alignment. Bootstrap values >70% are provided; *, indicates 100% support. *Plocamium pusillum* is highlighted in bold text. Scale bar: substitutions per site.

provided above). One of these genetic groups, recognized here as *Plocamium pusillum*, contained five specimens from disparate Australian locations including Victoria, Lord Howe Island, Norfolk Island, and Western Australia (Table 1). The five COI-5P genes determined for these specimens were identical, consistent with a single genetic species, and 2.3%

Fig. 2. — Lectotype specimen of *Plocamium pusillum* Sonder (MEL 1005815). Scale bar: 1 cm.

divergent (15 substitutions over 664 base pairs compared) from sequence data for an unassigned species from Oman (Table 1, DHO 0302). This level of divergence indicates separate genetic species.

In phylogenetic analyses *P. pusillum* and *P.* sp. 1Oman (DHO 0302) were strongly supported as sister species in a lineage distinct from that containing the morphologically similar *P. cartilagineum* under which *P. pusillum* was previously synonymized (Fig. 1).

Family Plocamiaceae Kützing
Genus *Plocamium* J.V.Lamouroux

Plocamium pusillum Sonder

Botanische Zeitung 3: 54 (Sonder 1845).

Type locality. — ad oras occidentales Novae Hollandiae. The type specimen was epiphytic on a stem of *Amphibolis*.

Lectotype (here designated). — MEL 1005815 (Fig. 2). Note: Sonder (1845) did not cite a specific specimen, but later (1846) gave 'Herb. Preiss No. 2613'. This number does not appear on MEL 1005815, which may suggest the existence of additional specimens in other herbaria. However, since we are unaware of any further specimens, we herein lectotypify the species with the single MEL specimen.

Specimens examined (DNA sequence vouchers). — **Western Australia**, First Rock, S of Penguin Island, Shoalwater Marine Park, 15.II.2011, *J.Huisman 15.2.11.1.16* (PERTH 08715351); Cape (Point) Peron, W.A., 3 m depth on algae, *G.W.Saunders & K.Dixon* (UNB, GWS025286); New South Wales: Yellow Rock, Lord Howe Island, from 15 m depth on rock, *G.W.Saunders* (UNB, GWS002025); Victoria: The Springs, Point Lonsdale, 4 m depth on rock, *G.W.Saunders & L.Kraft* (UNB, GWS016698); Norfolk Island, 1 m depth on rock, *G.W.Saunders & K.Dixon* (UNB, GWS029289).

Additional specimens examined (unsequenced, based on morphology). — **Western Australia**, Cape Peron, epilithic, 25.II.2021, *J.M.Huisman* (PERTH 09316647); Eglinton Rocks, 17.V.1996, *J.Huisman JH 665* (PERTH 06546757); Burns Beach, drift, 2.IX.1985, *R.D.Royce 2562* (PERTH 05163587); Cottesloe, drift, 12.VI.1951, *R.D.Royce 1062* (PERTH 04017765); Capel Beach, on rocks between tides, 17.IX.1949, *R.D.Royce 368* (PERTH 04017862); Point [Cape] Peron, epiphytic, 28.V.1949, *R.D.Royce s.n.* (PERTH 04017870); Mushroom Rock, Thomson Bay, Rottnest Island, 16.X.1934, *A.Nash s.n.* (PERTH 04017838).

Distribution. — Based on molecular analyses, this species is widely distributed and known from southern Western Australia, Victoria, and New South Wales (Lord Howe Island) and Norfolk Island.

Habit. — Thallus (Fig. 3A) dark red, to 6 cm tall, much branched, attached by a hapteroid holdfast to seagrasses, algae or epilithic (occasionally sand-binding). Primary axes typically sinuous, with lateral branches borne mostly in series of 3 (Fig. 3B), but occasionally in pairs. Lateral branches only slightly more slender than primary axes, therefore often indistinguishable. Lowermost branch in each series simple, straight or curved upwardly, linear with a pointed apex. Axes initially flattened, 250-300 µm broad, 50-100 µm thick (Fig. 3C) but becoming compressed to subterete towards the base, *c.* 600 µm broad, 400 µm thick (Fig. 3D). Structure pseudoparenchymatous, with large hyaline medullary

FIG. 3. — Morphology of recently collected *Plocamium pusillum* (all PERTH 09316647): **A**, pressed specimen showing habit; **B**, detail of apex showing origin of lateral branches in groups of three; **C**, section of young branch; **D**, section of older branch; **E**, surface view of living material showing prominent refringent vacuoles. These are not visible in preserved or dried material; **F**, tetrasporangia borne in groups near apices of lateral branches (**arrows**), not or only slightly distending the bearing branch; **G**, closer view of tetrasporangia showing biseriate arrangement. Scale bars: A, 1 cm; B, F, 300 µm; C, E, 30 µm; D, G, 60 µm.

cells grading abruptly to a cortex of 1-2 cell layers (Fig. 3C, D). Cortical cells with several ovoid to ellipsoid parietal plastids and a spherical central refringent vacuole, the latter only visible in living material (Fig. 3E).

Reproduction. — Tetrasporangia are borne in fertile distal segments of upper branches (Fig. 3F), the branches initially simple but eventually dividing and becoming glomerulate. Fertile segments only slightly distended. Tetrasporangia biseriate (Fig. 3G), zonately divided, ovoid/ellipsoidal, 50-60 µm long, 35-45 µm diam. Other reproductive structures not observed.

DISCUSSION

The type specimen of *P. pusillum* is unfortunately not reproductive, but the slender and slightly sinuous primary axes with lateral branches in groups of three are distinctive and seen in the more recent collections, some of which (GWS025286) are from the vicinity of the type locality and have been included in molecular analyses. We are therefore confident that the specimens included here in molecular analyses are authentic *P. pusillum* and the COI-5P barcode sequence should be considered characteristic for the species.

Cremades *et al.* (2011) provided a detailed description of authentic European *P. cartilagineum*, noting (p. 134) that the species has "considerable morphological variability". *Plocamium pusillum* differs in its more slender axes (to 600 µm broad in *P. pusillum*, up to 1.5 mm in *P. cartilagineum*), the occasional occurrence of lateral branches in groups of 4 in *P. cartilagineum*, and also in the fertile regions in tetrasporangial thalli being only slightly distended in *P. pusillum*, whereas those of *P. cartilagineum* have a "short stalk that is much narrower than the fertile portion" (Cremades *et al.* 2011: 133). The slender axes and overall smaller stature of *P. pusillum* would also appear to distinguish it from other Australian species, but further work is clearly needed to establish the morphological limits of the numerous species-level genetic groups recognised in BOLD.

The refringent vacuoles found in cortical cells of living specimens (Fig. 3E) are similar to the 'corpse en cerise' in species of *Laurencia* and to the 'mevalonosomes' described for *Plocamium brasiliense* (Greville) M.Howe & W.R.Taylor by Paradas *et al.* (2015). These structures have been implicated in the production of antifouling compounds (Paradas *et al.* 2015), however the regular appearance of epiphytic diatoms on *P. pusillum* might suggest this is not the case.

The resurrection of *Plocamium pusillum* has provided a name for one of the Australian genetic groups that would have previously been assigned to *P. cartilagineum* based on morphology, however there is clearly a need for further study and characterization of several additional species-level genetic groups (as seen in BOLD) that may represent novel taxa.

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REFERENCES

Cremades J., Barreiro R., Maneiro I. & Saunders G. W. 2011. — A new taxonomic interpretation of the type of *Plocamium cartilagineum* (Plocamiales, Florideophyceae) and its consequences. *European Journal of Phycology* 46: 125-142. https://doi.org/10.1080/09670262.2011.565129

Kearse M., Moir R., Wilson A., Stones-Havas S., Cheung M., Sturrock S., Buxton S., Cooper A., Markowitz S., Duran C., Thierer T., Ashton B., Mentjies P. & Drummond A. 2012. — Geneious Basic: an integrated and extendable desktop software platform for the organization and analysis of sequence data. *Bioinformatics* 28: 1647-1649. https://doi.org/10.1093/bioinformatics/bts199

Marchant N. G. 1990. — The Western Australian Collecting Localities of J.A.L. Preiss, *in* Short P. S. (ed.), *History of Systematic Botany in Australasia: Proceedings of a Symposium Held at the University of Melbourne*, 25-27 May 1988. Australian Systematic Botany Society, Melbourne: 131-136.

Paradas W. C., Crespo T. M., Salgado L. T., de Andrade L. R., Soares A. R., Hellio C., Paranhos R. R., Hill L. J., de Souza G. M., Kelecom A. G. A. C., Da Gama B. A. P., Pereira R. C. & Amado-Filho G. M. 2015. — Mevalonosomes: specific vacuoles containing the mevalonate pathway in *Plocamium brasiliense* cortical cells (Rhodophyta). *Journal of Phycology* 51: 225-235. https://doi.org/10.1111/jpy.12270

Ratnasingham S. & Hebert P. D. N. 2007. — BOLD: The Barcode of Life Data System (www.barcodinglife.org). *Molecular Ecology Notes* 7: 355-364. https://doi.org/10.1111/j.1471-8286.2007.01678.x

Saunders G. W. & Lehmkuhl K. V. 2005. — Molecular divergence and morphological diversity among four cryptic species of *Plocamium* (Plocamiales, Florideophyceae) in northern Europe. *European Journal of Phycology* 40: 3, 293-312. https://doi.org/10.1080/09670260500192935

Saunders G. W. & McDevit D. C. 2012. — Methods for DNA barcoding photosynthetic protists emphasizing the macroalgae and diatoms. *Methods in Molecular Biology* 858: 207-222.

Saunders G. W. & Moore T. E. 2013. — Refinements for the amplification and sequencing of red algal DNA barcode and RedToL phylogenetic markers: a summary of current primers, profiles and strategies. *Algae* 28: 31-43. https://doi.org/10.4490/algae.2013.28.1.031

Sonder G. 1845. — Nova algarum genera et species, quas in itinere ad oras occidentales Novae Hollandiae, collegit L. Priess, Ph. Dr. *Botanische Zeitung* 3: 49-57.

Sonder O. G. 1846. — Algae L. Agardh. In: Plantae Preissianae sive enumeratio plantarum quas in Australasia occidentali et meridionali-occidentali annis 1838-1841 collegit Ludovicus Preiss. Vol. 2: 148-160. Sumptibus Meissneri, Hamburgi.

Stamatakis A. 2014. — RAxML version 8: a tool for phylogenetic

analysis and post-analysis of large phylogenies. *Bioinformatics* 30: 1312-1313. https://doi.org/10.1093/bioinformatics/btu033

WOMERSLEY H. B. S. 1971. — The genus *Plocamium* (Rhodophyta) in southern Australia. *Transactions of the Royal Society of South Australia* 95: 9-27.

WOMERSLEY H. B. S. 1994. — *The marine benthic flora of southern Australia - Part IIIA - Bangiophyceae and Florideophyceae (Acrochaetiales, Nemaliales, Gelidiales, Hildenbrandiales and Gigartinales sensu lato)*. Australian Biological Resources Study, Canberra, 508 p.

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