

## Supplementary Materials

# **Rambellisea gigliensis and Rambellisea halocynthiae, gen. et spp. nov. (Lulworthiaceae) from the Marine Tunicate Halocynthia papillosa**

**Martina Braconcini<sup>1</sup>, Susanna Gorras<sup>1</sup>, Massimiliano Fenice<sup>1,2</sup>, Paolo Barghini<sup>1</sup> and Marcella Pasqualetti<sup>1,3,\*</sup>**

<sup>1</sup> Department of Ecological and Biological Sciences (DEB), University of Tuscia, 01100 Viterbo, Italy; martina.braconcini@unitus.it (M.B.); gorras<sup>i</sup>@unitus.it (S.G.); fenice@unitus.it (M.F.); barghini@unitus.it (P.B.)

<sup>2</sup> Laboratory of Applied Marine Microbiology, CoNISMa, Department of Ecological and Biological Sciences, University of Tuscia, 01100 Viterbo, Italy

<sup>3</sup> Laboratory of Ecology of Marine Fungi, CoNISMa, Department of Ecological and Biological Sciences, University of Tuscia, 01100 Viterbo, Italy

\* Correspondence: mpasqual@unitus.it; Tel.: +39-0761-357750

## List of Contents

Table S1. Substitution rates, gamma distribution shape parameter and proportion of invariable sites for each partition

Table S2. The variable sites detected in the nrITS region among *Rambellisea halocynthiae*, *R. gigliensis* and its neighbor species belonging to the genera *Paralulworthia*.

Table S3. The variable sites detected in the nrLSU region among *Rambellisea halocynthiae*, *R. gigliensis* and its neighbor species belonging to the genera *Paralulworthia*.

Table S4. The variable sites detected in the nrSSU region among *Rambellisea halocynthiae*, *R. gigliensis* and its neighbor species belonging to the genera *Paralulworthia*.

Figure S1. *Rambellisea gigliensis* sp. nov. HPa3 (MUT 6843). (a) 28-day-old colony: colony texture on ME Aws at 25° C; (b) 28-day-old colony: colony texture on CM Aws at 25° C.

Figure S2. *Rambellisea halocynthiae* sp. nov. HPa52 (MUT 6851). (a) 28-day-old colony on ME Aws (Ø 9 cm) at 25° C (b) and reverse; (c) 28-day-old colony on CM Aws (Ø 9 cm) at 25° C (d) and reverse

**Citation:** Braconcini, M.; Gorras<sup>i</sup>, S.; Fenice, M.; Barghini, P.; Pasqualetti, M. *Rambellisea gigliensis* and *Rambellisea halocynthiae*, gen. et spp. nov. (Lulworthiaceae) from the Marine Tunicate *Halocynthia papillosa*. *J. Fungi* **2024**, *10*, 127. <https://doi.org/10.3390/jof10020127>

Academic Editor: Philippe Silar

Received: 20 November 2023

Revised: 24 January 2024

Accepted: 1 February 2024

Published: 3 February 2024



**Copyright:** © 2024 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (<https://creativecommons.org/licenses/by/4.0/>).

**Table S1.** Substitution rates, gamma distribution shape parameter and proportion of invariable sites for each partition

	Substitution rates	Gamma distribution shape	Proportion of invariable site
<b>Partition 1</b>	gamma	0.6400	0.3970
<b>Partition 2</b>	gamma	0.3760	0
<b>Partition 3</b>	gamma	1.7790	0
<b>Partition 4</b>	gamma	0.2890	0
<b>Partition 5</b>	gamma	1.1200	0
<b>Partition 6</b>	gamma	0.3890	0

**Table S2.** The variable sites detected in the nrITS region among *Rambellisea halocynthiae*, *R. gigliensis* and its neighbor species belonging to the genera *Paralulworthia*.

Species	Position																
	20	23	24	44	46	54	56	57	94	101	102	103	104	108	109	113	114
<i>R. halocynthiae</i>	C	-	-	T	G	T	G	G	C	C	T	C	C	A	-	T	C
<i>R. gigliensis</i>	C	-	-	C	G	A	A	A	C	A	A	A	A	A	C	T	C
<i>P. candida</i>	-	A	A	T	A	-	T	G	A	C	A	C	G	C	A	C	A
<i>P. elbensis</i>	-	A	A	T	A	T	T	G	A	C	A	C	G	C	A	C	A
<i>P. gigaspora</i>	A	A	A	T	A	T	T	G	A	C	A	C	C	C	A	C	A
<i>P. halima</i>	A	A	A	T	A	T	T	G	A	C	A	C	C	C	A	C	A
<i>P. mediterranea</i>	A	A	A	T	A	T	T	G	A	C	A	C	C	C	A	C	A
<i>P. posidoniae</i>	A	A	A	T	A	T	T	G	A	C	A	C	C	C	A	C	A

Species	Position																
	119	120	121	122	123	129	131	132	159	170	171	245	317	329	340	365	366
<i>R. halocynthiae</i>	C	T	A	C	G	T	A	A	A	A	A	A	A	T	T	C	T
<i>R. gigliensis</i>	C	T	G	C	A	C	A	A	A	A	G	A	A	T	C	T	T
<i>P. candida</i>	A	C	C	A	G	T	C	C	G	C	A	C	G	C	T	-	-
<i>P. elbensis</i>	A	C	C	A	G	T	C	C	G	C	A	C	G	C	T	-	-
<i>P. gigaspora</i>	A	C	C	A	G	T	C	C	G	C	A	C	G	C	T	C	C
<i>P. halima</i>	A	C	C	A	G	T	C	C	G	C	A	C	G	C	T	C	C
<i>P. mediterranea</i>	A	C	C	A	G	T	C	C	G	C	A	C	G	C	T	C	C
<i>P. posidoniae</i>	A	C	C	A	G	T	C	C	G	C	A	C	G	C	T	C	C

Species	Position																
	368	369	370	371	373	374	376	377	384	385	386	400	429	430	439	447	464
<i>R. halocynthiae</i>	T	C	C	T	C	G	G	A	T	G	G	C	A	A	T	C	C
<i>R. gigliensis</i>	C	T	T	C	C	A	G	A	C	G	T	T	C	A	G	C	C
<i>P. candida</i>	A	C	C	T	T	T	C	G	T	A	G	C	T	C	G	T	T
<i>P. elbensis</i>	A	C	C	T	T	T	C	G	T	A	G	C	T	C	G	T	T
<i>P. gigaspora</i>	A	C	C	T	T	T	C	G	T	A	G	C	T	C	G	T	T
<i>P. halima</i>	A	C	C	T	T	T	C	G	T	A	G	C	T	C	G	T	T
<i>P. mediterranea</i>	A	C	C	T	T	T	C	G	T	A	G	C	T	C	G	T	T
<i>P. posidoniae</i>	A	C	C	T	T	T	C	G	T	A	G	C	T	C	G	T	T

Species	Position																
	465	470	474	486	487	488	509	512	513	514	515	516	531	532	576	578	582
<i>R. halocynthiae</i>	C	C	T	-	-	-	G	T	C	G	G	T	G	A	T	C	C
<i>R. gigliensis</i>	T	G	T	-	-	-	G	T	C	G	G	T	G	A	C	C	G
<i>P. candida</i>	C	T	C	C	A	A	C	A	-	-	-	A	G	C	A	C	C
<i>P. elbensis</i>	C	T	C	C	C	A	A	C	A	-	-	A	G	C	A	C	C

<i>P. gigaspora</i>	C	C	C	C	C	A	A	C	A	-	-	-	A	G	C	A	C
<i>P. halima</i>	C	C	C	C	C	A	A	C	A	-	-	-	A	G	C	A	C
<i>P. mediterranea</i>	C	C	C	C	C	A	A	C	A	-	-	-	A	G	C	A	C
<i>P. posidoniae</i>	C	C	C	C	C	A	A	C	A	-	-	-	A	G	C	A	C

588 589 600

<i>R. halocynthiae</i>	A	C	<b>A</b>
<i>R. gigliensis</i>	<b>C</b>	<b>T</b>	<b>A</b>
<i>P. candida</i>	A	C	C
<i>P. elbensis</i>	A	C	C
<i>P. gigaspora</i>	A	C	C
<i>P. halima</i>	A	C	C
<i>P. mediterranea</i>	A	C	C
<i>P. posidoniae</i>	A	C	C

**Table S3.** The variable sites detected in the nrLSU region among *Rambellisea halocynthiae*, *R. gigliensis* and its neighbor species belonging to the genera *Paralulworthia*.

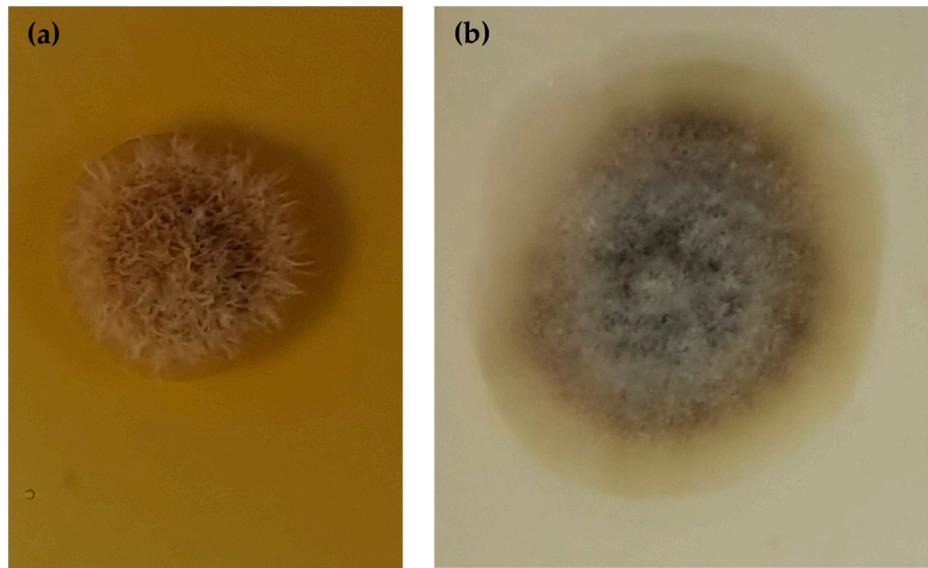
Species	Position																
	102	144	145	173	383	407	408	466	467	468	469	487	488	508	509	540	569
<i>R. halocynthiae</i>	<b>T</b>	<b>G</b>	G	C	C	<b>T</b>	<b>C</b>	<b>C</b>	<b>G</b>	<b>A</b>	<b>T</b>	<b>T</b>	<b>T</b>	<b>C</b>	<b>T</b>	T	<b>T</b>
<i>R. gigliensis</i>	<b>T</b>	<b>A</b>	<b>C</b>	<b>T</b>	<b>A</b>	<b>T</b>	<b>C</b>	T	<b>G</b>	<b>A</b>	C	C	C	<b>C</b>	<b>T</b>	<b>C</b>	C
<i>P. candida</i>	C	T	G	C	C	-	-	T	A	G	C	C	C	T	C	T	C
<i>P. elbensis</i>	C	T	G	C	C	-	-	T	A	G	C	C	C	T	C	T	C
<i>P. gigaspora</i>	C	T	G	C	C	-	-	T	A	G	C	C	C	T	C	T	C
<i>P. halima</i>	C	T	G	C	C	-	-	T	A	G	C	C	C	T	C	T	C
<i>P. mediterranea</i>	C	T	G	C	C	-	-	T	A	G	C	C	C	T	C	T	C
<i>P. posidoniae</i>	C	T	G	C	C	-	-	T	A	G	C	C	C	T	C	T	C

Species	Position															
	601	643	668	764	765	772	808	809	878	1016	1050	1051	1052	1062	1070	1071
<i>R. halocynthiae</i>	<b>T</b>	<b>T</b>	<b>A</b>	<b>A</b>	<b>A</b>	<b>A</b>	<b>T</b>	<b>A</b>	<b>T</b>	<b>T</b>	C	<b>C</b>	<b>T</b>	<b>G</b>	T	<b>G</b>
<i>R. gigliensis</i>	<b>T</b>	<b>T</b>	<b>A</b>	<b>A</b>	<b>A</b>	<b>A</b>	<b>T</b>	<b>A</b>	<b>T</b>	<b>T</b>	<b>T</b>	<b>C</b>	<b>T</b>	<b>G</b>	<b>C</b>	<b>G</b>
<i>P. candida</i>	C	C	C	G	G	G	C	G	C	C	C	A	C	A	T	C
<i>P. elbensis</i>	C	C	C	G	G	G	C	G	C	C	C	A	C	A	T	C
<i>P. gigaspora</i>	C	C	C	G	G	G	C	G	C	C	C	A	C	A	T	C
<i>P. halima</i>	C	C	C	G	G	G	C	G	C	C	C	A	C	A	T	C
<i>P. mediterranea</i>	C	C	C	G	G	G	C	G	C	C	C	A	C	A	T	C
<i>P. posidoniae</i>	C	C	C	G	G	G	C	G	C	C	C	A	C	A	T	C

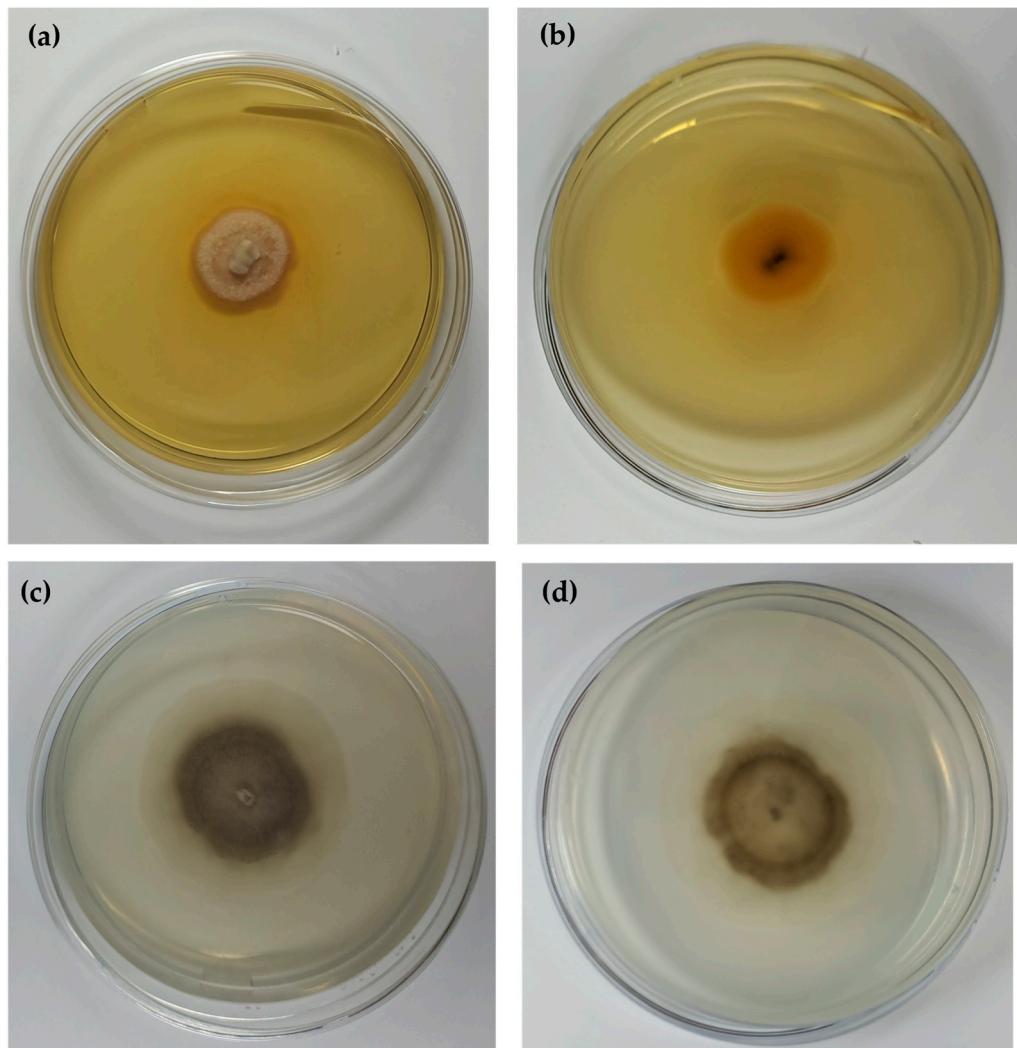
**Table S4.** The variable sites detected in the nrSSU region among *Rambellisea halocynthiae*, *R. gigliensis* and its neighbor species belonging to the genera *Paralulworthia*.

Species	Position																
	43	99	100	101	107	110	121	129	130	140	161	219	323	349	350	376	1126
<i>R. halocynthiae</i>	<b>T</b>	T	C	G	<b>A</b>	<b>G</b>	<b>A</b>	C	<b>T</b>	<b>T</b>	<b>T</b>	<b>A</b>	<b>A</b>	<b>A</b>	<b>G</b>	<b>C</b>	<b>T</b>
<i>R. gigliensis</i>	<b>T</b>	<b>A</b>	<b>A</b>	<b>C</b>	<b>C</b>	T	G	<b>T</b>	<b>T</b>	<b>T</b>	<b>T</b>	<b>A</b>	<b>A</b>	G	A	T	<b>T</b>

<i>P. candida</i>	G	T	C	G	G	T	G	C	A	C	C	G	T	G	A	T	A
<i>P. elbensis</i>	G	T	C	G	G	T	G	C	A	C	C	G	T	G	A	T	A
<i>P. gigaspora</i>	G	T	C	G	G	T	G	C	A	C	C	G	T	G	A	T	A
<i>P. halima</i>	G	T	C	G	G	T	G	C	A	C	C	G	T	G	A	T	A
<i>P. mediterranea</i>	G	T	C	G	G	T	G	C	A	C	C	G	T	G	A	T	T
<i>P. posidoniae</i>	G	T	C	G	G	T	G	C	A	C	C	G	T	G	A	T	A



**Figure S1.** *Rambellisea gigliensis* sp. nov. HPa3 (MUT 6843). (a) 28-day-old colony: colony texture on MEAwS at 25° C; (b) 28-day-old colony: colony texture on CMAws at 25° C.



**Figure S2.** *Rambellisea halocynthiae* sp. nov. HPa52 (MUT 6851). (a) 28-day-old colony on MEA<sub>ws</sub> ( $\varnothing$  9 cm) at 25° C (b) and reverse; (c) 28-day-old colony on CMA<sub>ws</sub> ( $\varnothing$  9 cm) at 25° C (d) and reverse.