

Supplementary Material

Revealing hidden diversity among upside-down jellyfishes (Cnidaria: Scyphozoa: Rhizostomeae: *Cassiopea*): distinct evidence allows the change of status of a neglected variety and the description of a new species

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Table S1. Mitochondrial ribosomal gene *16S* rRNA and mitochondrial protein-encoding gene cytochrome *c* oxidase I (*COI*) sequences used for combined-marker analysis

Terminal ID corresponds to terminal names on maximum likelihood tree based on *16S+COI* (Fig. 2).

–, marker not available for the taxa. GenBank accession numbers of sequences obtained in this study are in bold

Terminal ID	<i>16S</i>	<i>COI</i>
<i>Cassiopea andromeda</i> – French Polynesia	JN700934	JN700934
<i>C. andromeda</i> – Mexico	KY610609	KY610551
<i>C. culionensis</i> _1 – Cebu, Philippines	MW164879	MW160923
<i>C. culionensis</i> _2 – Cebu, Philippines	MW164869	MW160913
<i>C. culionensis</i> _3 – Cebu, Philippines	MW164886	MW160930
<i>C. frondosa</i> – Florida, USA	KY610617	AY319467
<i>C. frondosa</i> – Panama	–	KY610560
<i>C. mayeri</i> – Japan	MW164859	MW160931
<i>C. mayeri</i> _1 – Cebu, Philippines	MW164863	MW160934
<i>C. mayeri</i> _2 – Cebu, Philippines	MW164864	MW160935
<i>C. mayeri</i> _2 – Luzon Island, Philippines	MW164866	MW160937
<i>C. mayeri</i> _1 – Luzon Island, Philippines	MW164865	MW160936
<i>C. ornata</i> – ?Kamo Aquarium, Indonesia	AB720918	AY319472
<i>C. ornata</i> – Indonesia	–	AY319473
<i>C. xamachana</i> _1 – Panama	JN700936	JN700936
<i>C. xamachana</i> _2 – Panama	KY610614	KY610559
<i>Cassiopea</i> sp. 1 – Queensland, Australia	–	AY319471
<i>Cassiopea</i> sp. 2_1 – Papua New Guinea	–	MF742198
<i>Cassiopea</i> sp. 2_2 – Papua New Guinea	–	MF742199
<i>Cassiopea</i> sp. 3_1 – Hawaii, USA	–	AY331594
<i>Cassiopea</i> sp. 3_2 – Hawaii, USA	–	AY331595
<i>Cassiopea</i> sp. 4_1 – Palau	MZ366351	LC198739
<i>Cassiopea</i> sp. 4_2 – Palau	MZ366351	LC198740
<i>Cassiopea</i> sp. 6_1 – Palau	–	LC198754
<i>Cassiopea</i> sp. 6_2 – Palau	–	LC198763
<i>Mastigias papua</i>	KY610621	KU901434
<i>Versuriga anadyomene</i>	KX904852	KX904853

Table S2. Mitochondrial ribosomal gene *16S* rRNA and mitochondrial protein-encoding gene cytochrome *c* oxidase I (*COI*) sequences used for combined-marker analysis without missing data

Terminal ID corresponds to terminal names on maximum likelihood tree based on *16S+COI* (Fig. S3)

Terminal ID	<i>16S</i>	<i>COI</i>
<i>Cassiopea andromeda</i> – French Polynesia	JN700934	JN700934
<i>C. andromeda</i> – Mexico	KY610609	KY610551
<i>C. culionensis_1</i> – Cebu, Philippines	MW164879	MW160923
<i>C. culionensis_2</i> – Cebu, Philippines	MW164869	MW160913
<i>C. culionensis_3</i> – Cebu, Philippines	MW164886	MW160930
<i>C. frondosa</i> – Florida, USA	KY610617	AY319467
<i>C. mayeri</i> – Japan	MW164859	MW160931
<i>C. mayeri_1</i> – Cebu, Philippines	MW164863	MW160934
<i>C. mayeri_2</i> – Cebu, Philippines	MW164864	MW160935
<i>C. mayeri_2</i> – Luzon Island, Philippines	MW164866	MW160937
<i>C. mayeri_1</i> – Luzon Island, Philippines	MW164865	MW160936
<i>C. ornata</i> - ?Kamo Aquarium, Indonesia	AB720918	AY319472
<i>C. xamachana_1</i> – Panama	JN700936	JN700936
<i>C. xamachana_2</i> – Panama	KY610614	KY610559
<i>Mastigias papua</i>	KY610621	KU901434
<i>Versuriga anadyomene</i>	KX904852	KX904853

Table S3. Mitochondrial ribosomal gene 16S rRNA and mitochondrial protein-encoding gene cytochrome *c* oxidase I (*COI*) sequences used for network construction

GenBank accession numbers of sequences obtained in this study are underlined. N/A, Not applicable

Species	Voucher code	<i>16S</i>	<i>COI</i>
<i>Cassiopea culionensis</i> , stat. nov.	gACM00279	<u>MW164876</u>	<u>MW160920</u>
	gACM00281	<u>MW164877</u>	<u>MW160921</u>
	gACM00282	<u>MW164867</u>	<u>MW160911</u>
	gACM00284	<u>MW164868</u>	<u>MW160912</u>
	gACM00285	<u>MW164869</u>	<u>MW160913</u>
	gACM00286	<u>MW164870</u>	<u>MW160914</u>
	gACM00287	<u>MW164875</u>	<u>MW160919</u>
	gACM00288	<u>MW164871</u>	<u>MW160915</u>
	gACM00290	<u>MW164872</u>	<u>MW160916</u>
	gACM00292	<u>MW164873</u>	<u>MW160917</u>
	gACM00293	<u>MW164874</u>	<u>MW160918</u>
	MZUSP 8632, gACM00340	<u>MW164878</u>	<u>MW160922</u>
	MZUSP 8633, gACM00341	<u>MW164879</u>	<u>MW160923</u>
	MZUSP 8634, gACM00342	<u>MW164880</u>	<u>MW160924</u>
	MZUSP 8635, gACM00343	<u>MW164881</u>	<u>MW160925</u>
	MZUSP 8636, gACM00344	<u>MW164882</u>	<u>MW160926</u>
	MZUSP 8637, gACM00345	<u>MW164883</u>	<u>MW160927</u>
	MZUSP 8638, gACM00346	<u>MW164884</u>	<u>MW160928</u>
	MZUSP 8639, gACM00347	<u>MW164885</u>	<u>MW160929</u>
	MZUSP 8640, gACM00348	<u>MW164886</u>	<u>MW160930</u>
N/A	N/A	KF683387	
<i>C. mayeri</i> , sp. nov.	N/A	N/A	AB563739
	N/A	N/A	AB563740
	FLMNH 007505	<u>MW164859</u>	<u>MW160931</u>
	FLMNH 009664/CASIZ 201000	<u>MW164865</u>	<u>MW160936</u>
	FLMNH 009665/CASIZ 200996	<u>MW164866</u>	<u>MW160937</u>
	gACM00283	<u>MW164860</u>	<u>MW160932</u>
	gACM00289	<u>MW164861</u>	<u>MW160933</u>
	gACM00291	<u>MW164862</u>	N/A
	gACM00280	<u>MW164863</u>	<u>MW160934</u>
	MZUSP 8631, gACM00339	<u>MW164864</u>	<u>MW160935</u>

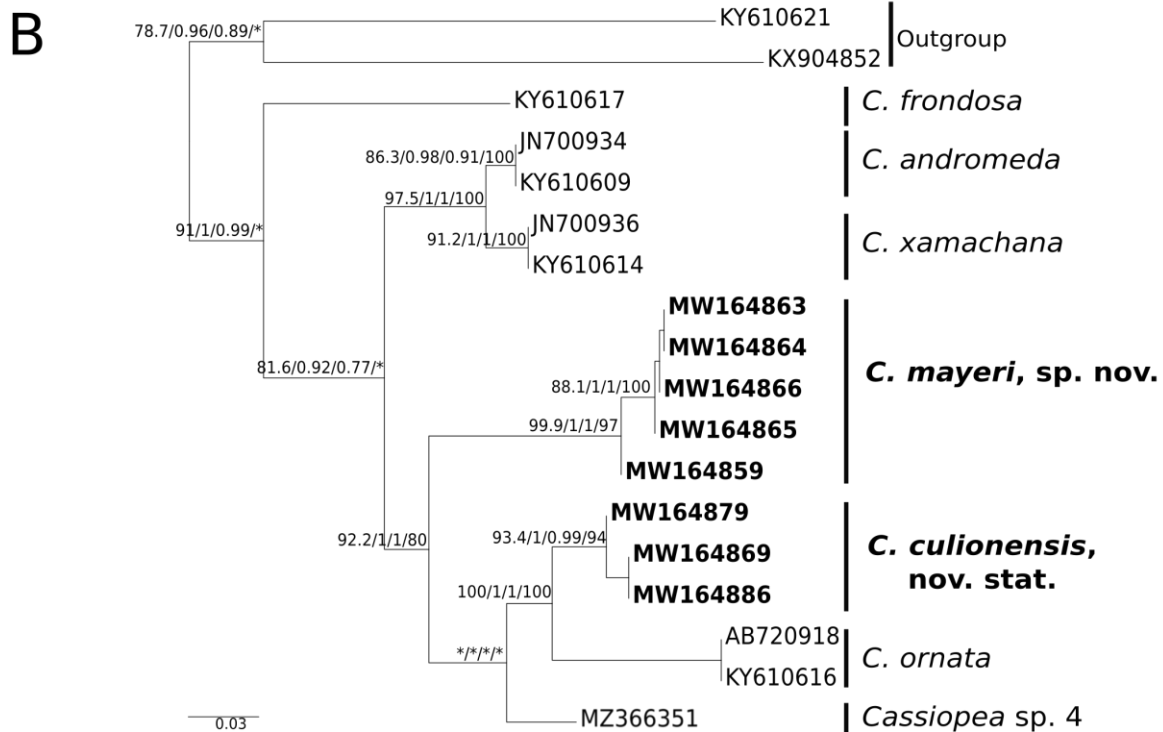
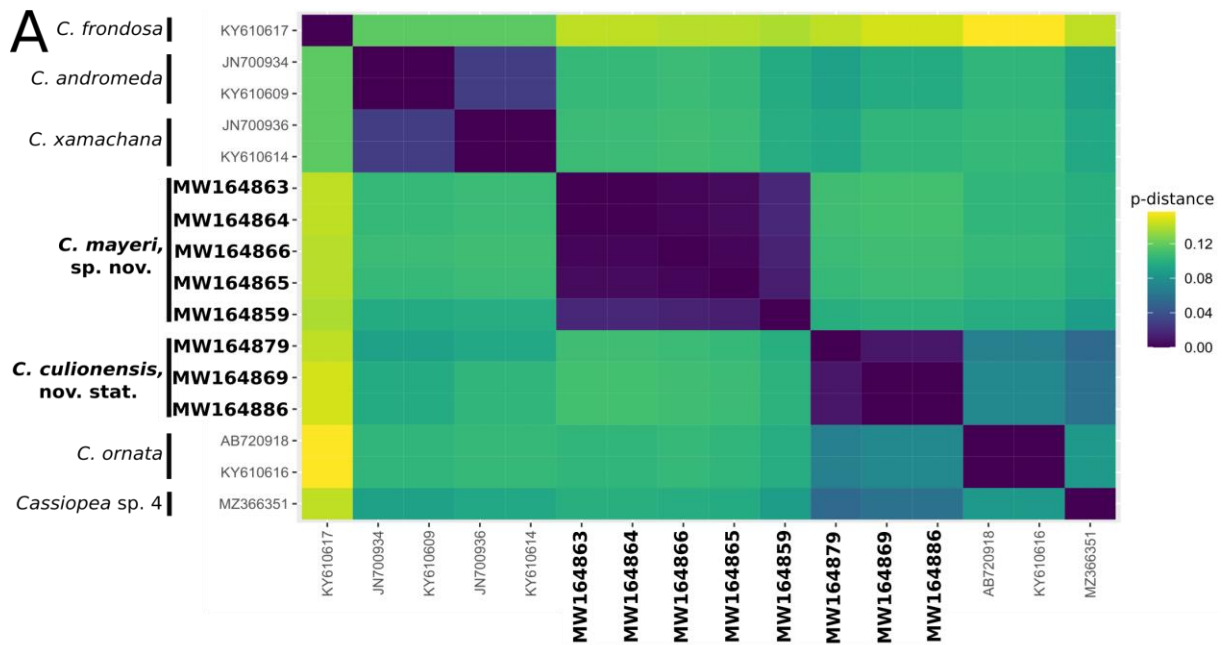


Fig. S1. Heatmap of pairwise uncorrected p -distances and maximum likelihood tree based on mitochondrial ribosomal gene *16S* rDNA data. (A) Heatmap of pairwise uncorrected p -distances. The colours from blue to yellow indicate low to high p -distance. For taxa names and other information, see the column ‘GenBank’ in Table 1. (B) Maximum likelihood tree based on *16S* sequences ($-\ln L$ 2089.353). Clade stability values are shown on branches (as in figure order: Shimodaira–Hasegawa (SH)- approximate likelihood ratio test (aLRT) (%), parametric aLRT/aBayes, bootstrap values (%); Asterisks (*) indicate less than 0.7, 70). For taxa names and other information, see the column ‘GenBank’ in Table 1.

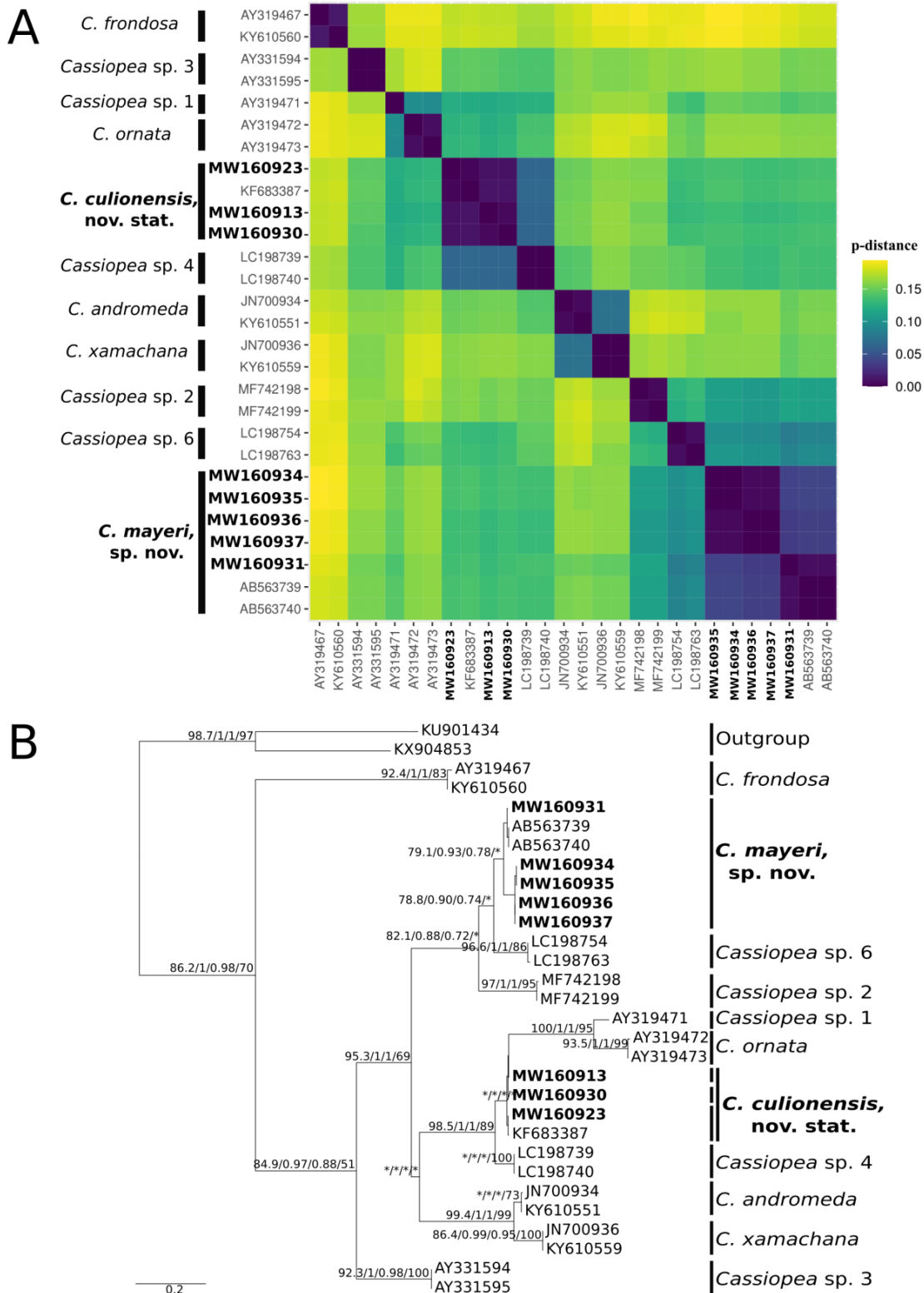


Fig. S2. Heatmap of pairwise uncorrected p -distances and maximum likelihood tree based on mitochondrial protein-encoding gene cytochrome c oxidase I (COI) data. (A) Heatmap of pairwise uncorrected p -distances. The colours from blue to yellow indicate low to high p -distance. For taxa names and other information, see the column ‘GenBank’ in Table 1. (B) Maximum likelihood tree based on COI sequences ($-\ln L$ 2757.711). Clade stability values are shown on branches (as in figure order: Shimodaira–Hasegawa (SH)- approximate likelihood ratio test (aLRT) (%), parametric aLRT, aBayes, bootstrap values (%); Asterisks (*) indicate less than 0.7, 70). For taxa names and other information, see the column ‘GenBank’ in Table 1.

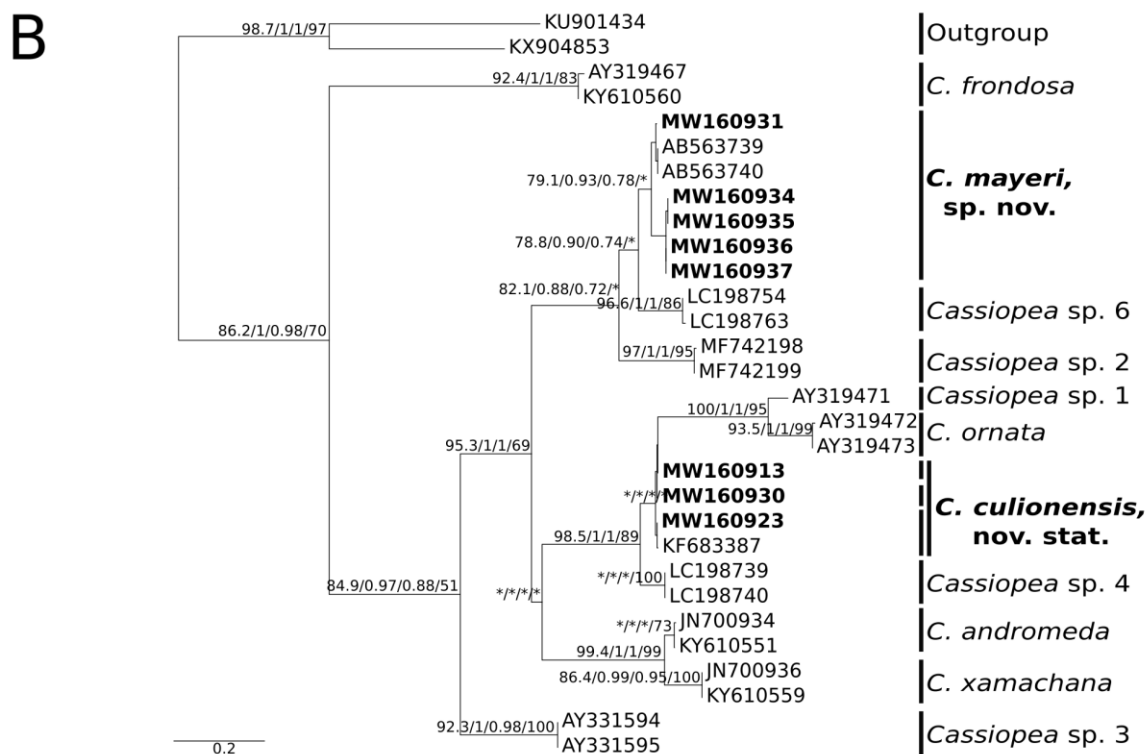
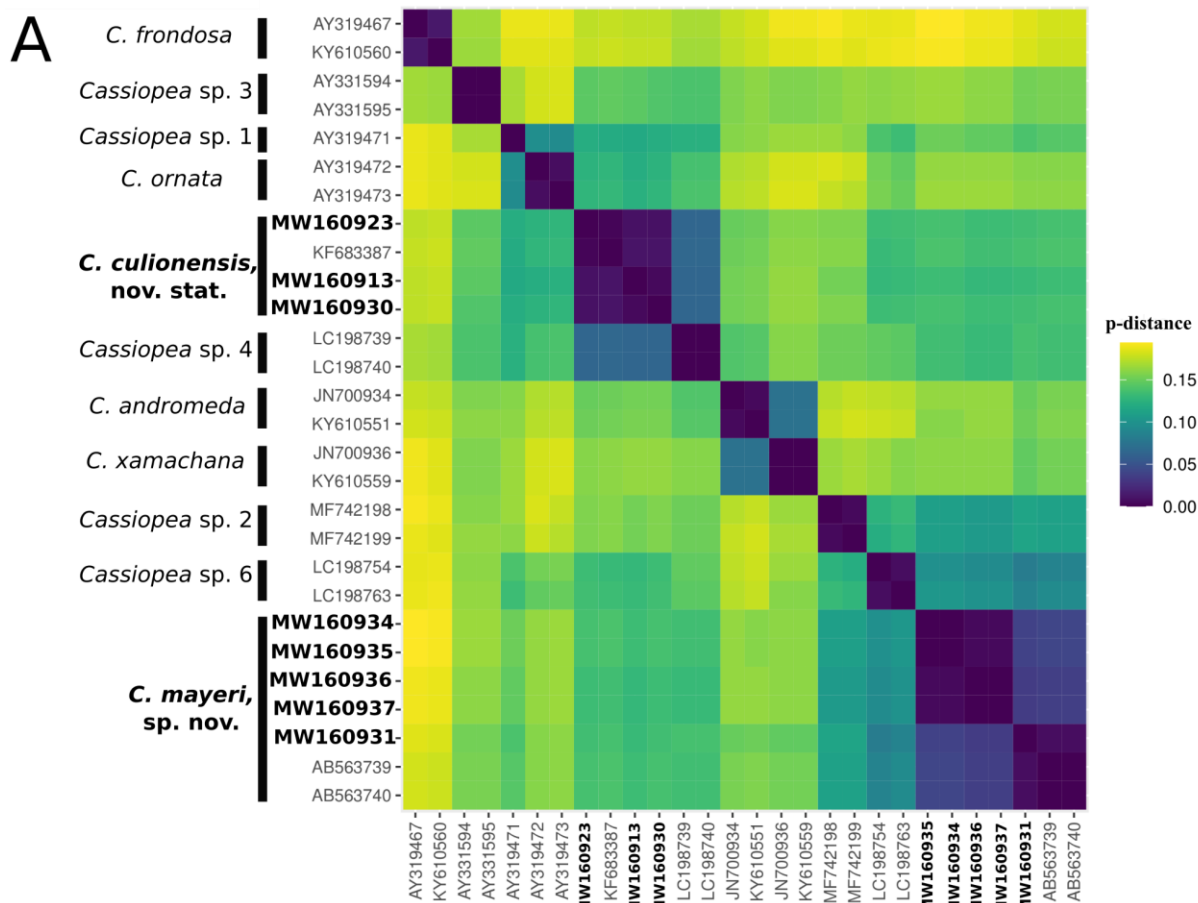


Fig. S3. Maximum likelihood tree based on mitochondrial ribosomal gene *16S* rRNA and mitochondrial protein-encoding gene cytochrome *c* oxidase I (*COI*) data ($-\ln L$ 4322.097). Grey rectangles indicate sequences obtained during this study. Clade stability values are shown on branches (as in figure order: Shimodaira–Hasegawa (SH)- approximate likelihood ratio test (aLRT) (%), parametric aLRT, aBayes, bootstrap values (%). For taxa names and other information, see Table S2.

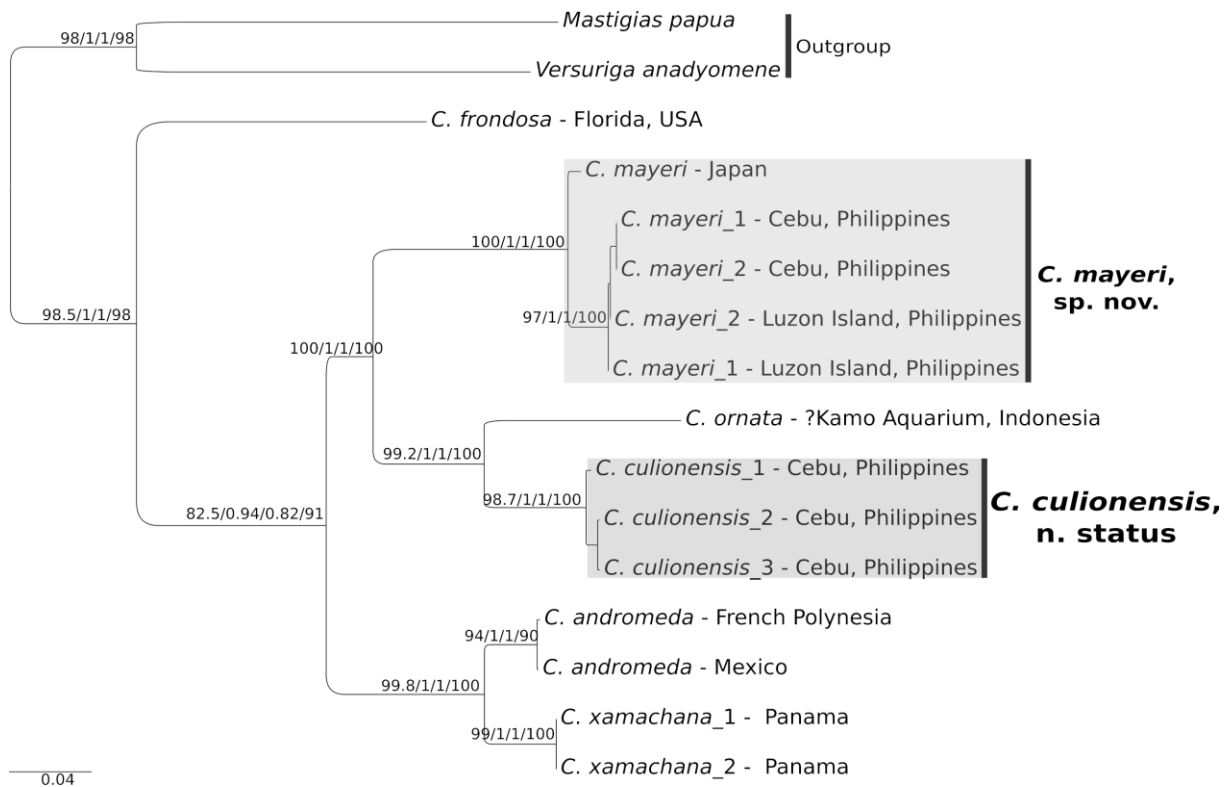


Fig. S4. *Cassiopea* aff. *mayeri*, photographs of the bigger specimen of the lot USNM 27941. (A) Oral view of the oral disc and arms, which were detached. (B) Detail of the appendages at the oral disc. Black arrows indicate fungiform (peltate) appendages, white arrows indicate tuber-shaped appendages. (C) Appendages of the oral arm. White arrows indicate the remainders of small appendages. (D) Exumbrellar view of a rhopalium and a rhopaliar pit.