

DNA barcoding reveals the coral “laboratory-rat”, *Stylophora pistillata* encompasses multiple identities

Shashank Keshavmurthy¹✉ Sung-Yin Yang^{1,2}✉ Ada Alamaru³✉ Yao-Yang Chuang^{1,23} Michel Pichon^{4,5}
David Obura⁶ Silvia Fontana^{1,7,8} Stephane De Palmas¹ Fabrizio Stefani⁹ Francesca Benzoni⁷ Angus
MacDonald¹⁰ Annika M. E. Noreen¹¹ Chienshun Chen¹² Carden C Wallace⁴ Ruby Moothien Pillay¹³
Vianney Denis¹ Affendi Yang Amri^{14,15} James D. Reimer¹⁶ Takuma Mezaki¹⁷ Charles Sheppard¹⁸
Yossi Loya³ Avidor Abelson³ Mohammed Suleiman Mohammed¹⁹ Andrew C. Baker²⁰ Pargol
Ghavam Mostafavi²¹ Budiyanto A. Suharsono²² Chaolun Allen Chen^{1,8,23*}

¹Biodiversity Research Centre, Academia Sinica, Nangang, Taipei 115, Taiwan

²Graduate school of Engineering and Science, University of the Ryukyus, Okinawa 903-0213, Japan

³Department of Zoology, George S. Wise Faculty of Life Sciences, Tel Aviv University, Tel Aviv 69978, Israel

⁴Museum of Queensland, Townsville 4811, Australia

⁵James Cook University, Australia

⁶Coastal Ocean Research and Development of Indian Ocean (CORDIO), Mombasa, Kenya

⁷Department of Biotechnology and Biosciences, University of Milano-Bicocca, P.zza della Scienza, 2, 20126
Milan, Italy

⁸Taiwan International Graduate Program (TIGP)-Biodiversity, Academia Sinica, Nangang, Taipei 115, Taiwan

⁹Water Research Institute, National Research Council (IRSA-CNR), Via del Mulino 19, 20861 Brugherio (MB),
Italy

¹⁰School of Life Sciences, University of KwaZulu-Natal, Private Bag X54001, Durban, 4001, South Africa.

¹¹Coral Reef Research Centre, School of Environmental Science and Management, Southern Cross University, PO
Box 157, Lismore, New South Wales 2480, Australia

¹²Taiwan Ocean Research Institute, National Applied Research Laboratories, Kaohsiung 852, Taiwan

¹³Mauritius Oceanography Institute, France Centre, Victoria Avenue Quatre Bornes, Mauritius

¹⁴Institute of Biological Sciences, Faculty of Science, Universiti Malaya, 50603 Kuala Lumpur, Malaysia and
School of Plant Biology

¹⁵The University of Western Australia, Crawley, 6009 Western Australia

¹⁶Rising Star Program, Transdisciplinary Research Organization for Subtropical Island Studies (TRO-SIS),
University of the Ryukyus, Okinawa 903-0213, Japan

¹⁷Biological Kuroshio Institute, Otuski, Kochi, Japan

¹⁸School of Life Sciences, University of Warwick, CV4 7AL, United Kingdom

¹⁹Institute of Marine Science, University of Dar es Salaam, Zanzibar, Tanzania

²⁰Division of Marine Biology and Fisheries, Rosenstiel School of Marine and Atmospheric Science, University of
Miami 4600 Rickenbacker Causeway, Miami, FL 33149, USA

²¹Department of Marine Biology, Graduate school of Marine Science and Technology, Science and Research
Branch, 3 Islamic Azad University, Iran

²²Research Center for Oceanography, Indonesian Institute of Sciences (LIPI), Jl. Pasir Putih I, Ancol Timur Jakarta,
Indonesia

²³Institute of Oceanography, National Taiwan University, Taipei 106, Taiwan

✉SK, SY and AA contributed equally to this work

*Correspondence to - cac@gate.sinica.edu.tw

Table S1. Intra-clade (in grey) and inter-clade variation in COI [sequence diversity (p-distance)] for *Stylophora* samples.

p-distance	Clade 1	Clade 2	Clade 3	Clade 4
Clade1	0.00017503± 0.00009578			
Clade 2	0.00378 ± 0.00248	0 ± 0		
Clade 3	0.01592 ± 0.00492	0.01584± 0.00495	0.00462174 ± 0.001932581	
Clade 4	0.01326 ± 0.00470	0.01317 ± 0.00465	0.01274 ± 0.00436	0.000175716 ±0.000117208

Table S2. Information on COI base pair changes, length of CAD, tRNAW duplication and *Symbiodinium* clades in the four clades of *Stylophora*. Asterix represent *Symbiodinium* data that was obtained from published databases.

Location	COI bp changes / Amino acid changes	Length of CAD 3	tRNAW duplication	<i>Symbiodinium</i> clades
Clade 1 Pacific Ocean				
Australia (GBR)	7/3	500bp	—	*C
Taiwan	7/3	500bp	Yes	C
Japan	7/3	500bp	Yes	C
Malaysia	7/3	500bp	Yes	C
Indonesia	7/3	500bp		C
New Caledonia	7/3	500bp		C
Clade 2 Indian Ocean				
Kenya	—	900bp	—	C
Tanzania	7 (3) / 4 (2)	500bp	—	C and D
South Africa	9 (4) / 4 (2)	500bp, or 400bp, 600bp, 900bp	—	C
Madagascar	9 (4) / 4 (2)	500bp, or 400bp, 600bp, 900bp	—	C
St. Brandon's (Mauritius)	7/3	500bp	—	C
Chagos Archipelago	7/3	500bp	Yes	C
Australia	7/3	500bp	—	*C
Clade 3 <i>Stylophora madagascarensis</i>				
Tanzania	7 (3) / 4 (2)	500bp	—	C and D
Madagascar	9 (4) / 4 (2)	500bp, or 400bp, 600bp, 900bp	—	C
South Africa	9 (4) / 4 (2)	500bp, or 400bp, 600bp, 900bp	—	C
Yemen	Morph S 6/2	900bp	No	A and C
Clade 4 Red Sea				
Saudi Arabia (Gulf of Aqaba)	—	900bp	No	A and A+C
Eilat (Red Sea)	—	900bp	No	A and A+C
Djibouti	—	900bp	No	A and A+C
Yemen	Morph M, L	900bp	No	A and C
Oman	—	900bp	No	A and A+C

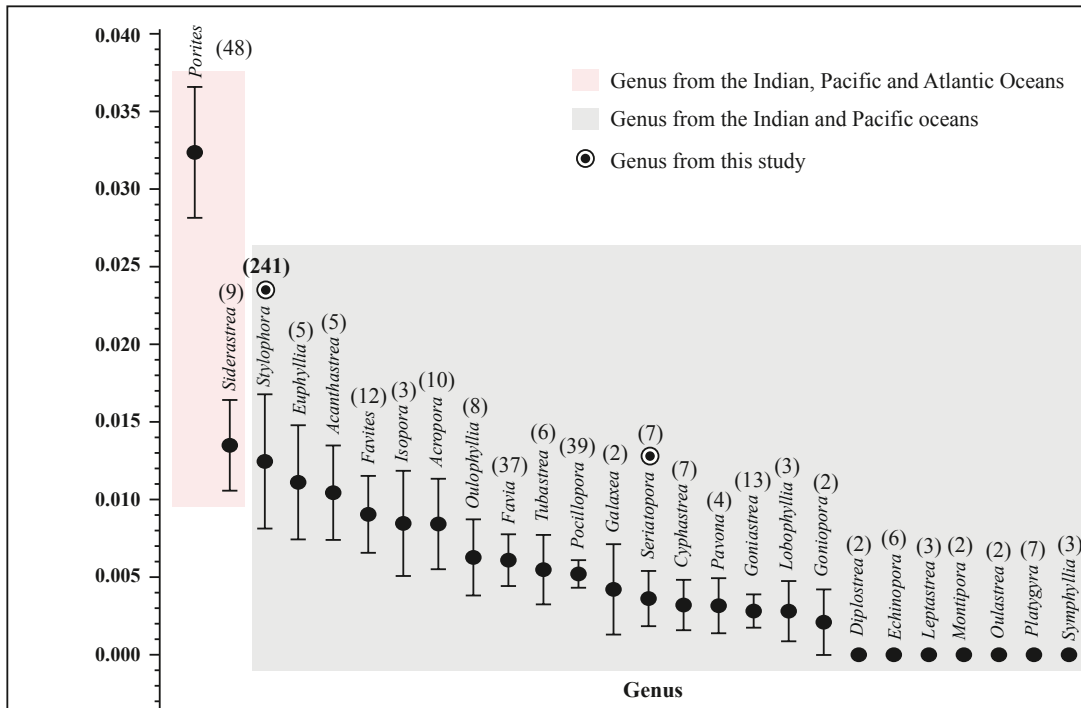


Figure S1. Interspecific variation in COI [sequence diversity (p-distance)] for *Stylophora* samples compared with other coral groups. Numbers in the brackets represent the number of sequences for each genus obtained from the NCBI GenBank database, except for *Stylophora* and *Seriatopora* sequences for which the samples were obtained in this study.

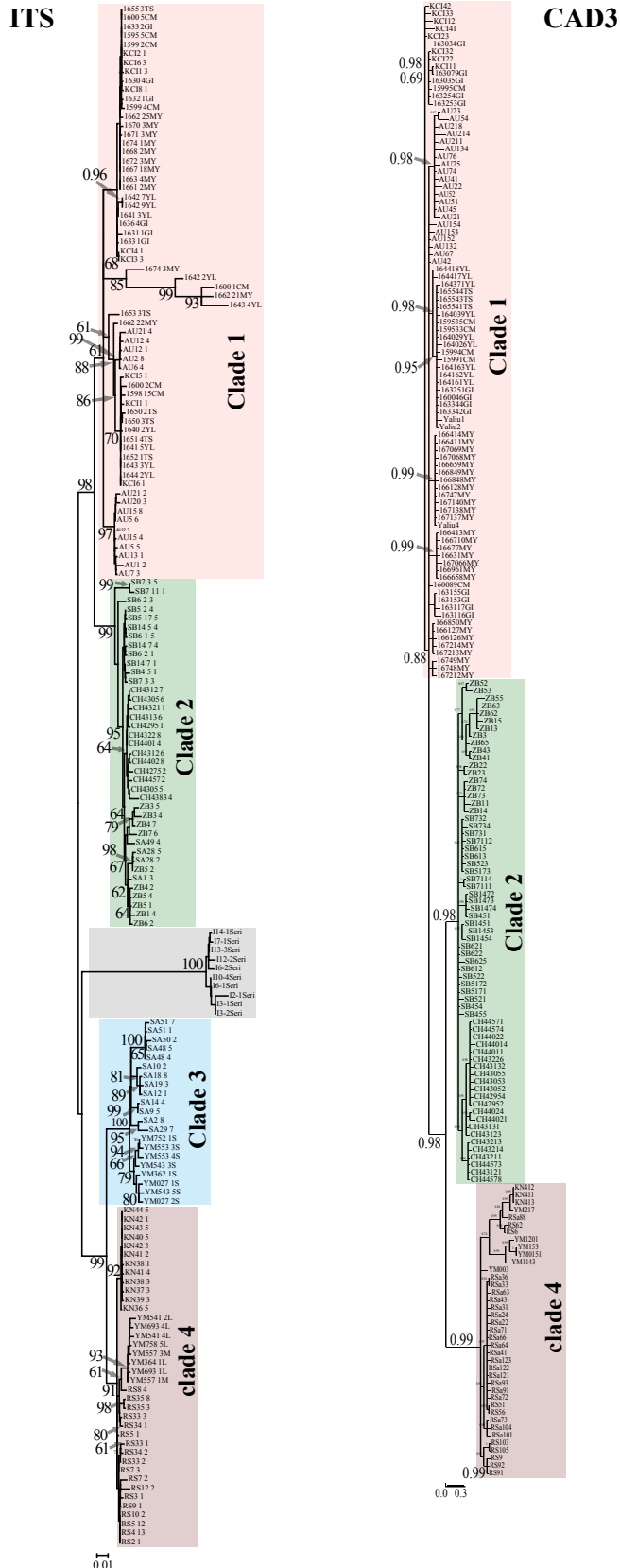


Figure S2. Phylogenetic analysis of ITS (whole ITS1 and ITS2 corresponding to 1138bp) and CAD3 (corresponding to 500bp) sequences of *Stylophora* samples. The absence of clade 3 and *Seriatoproa* groups in CAD3 phylogenetic tree was due to non-amplification of the corresponding samples. Bootstrap values are based on ML/Baysian analysis.

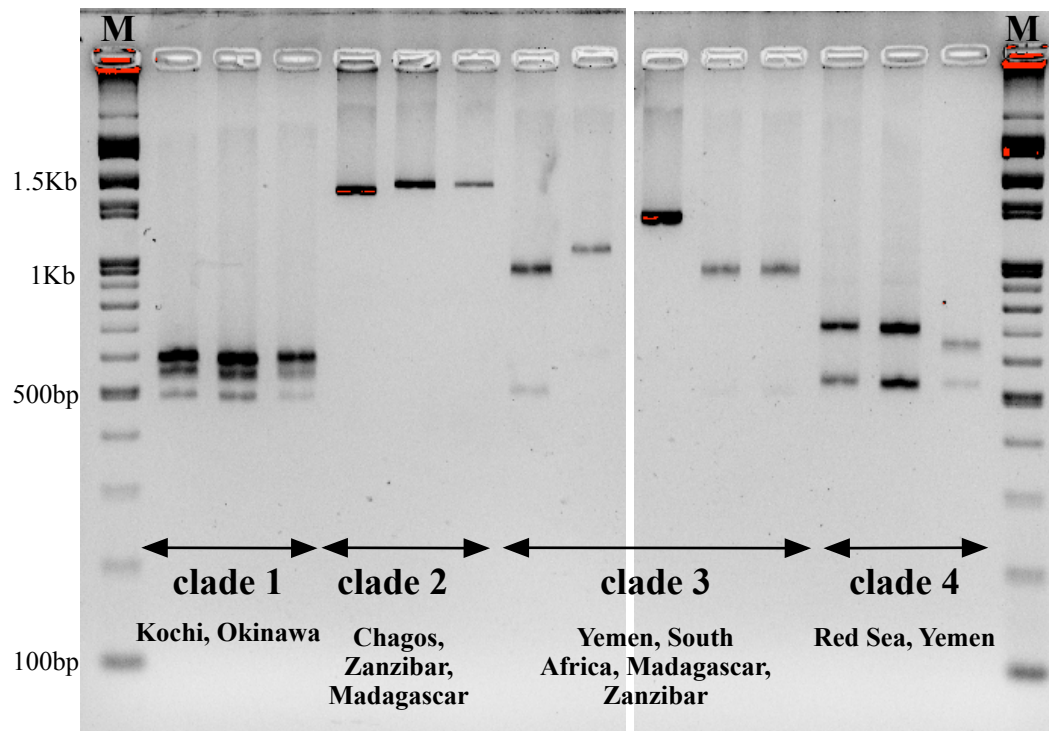


Figure S3. Representative tRNAW pattern for clade 1, clade 2, clade 3 and clade 4 of *Stylophora* sp. obtained by restriction enzyme (RSAI) digestion of PCR products.