

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

Shashank Keshavmurthy<sup>1</sup>✉ Sung-Yin Yang<sup>1,2</sup>✉ Ada Alamaru<sup>3</sup>✉ Yao-Yang Chuang<sup>1,23</sup> Michel Pichon<sup>4,5</sup>  
David Obura<sup>6</sup> Silvia Fontana<sup>1,7,8</sup> Stephane De Palmas<sup>1</sup> Fabrizio Stefani<sup>9</sup> Francesca Benzoni<sup>7</sup> Angus  
MacDonald<sup>10</sup> Annika M. E. Noreen<sup>11</sup> Chienshun Chen<sup>12</sup> Carden C Wallace<sup>4</sup> Ruby Moothien Pillay<sup>13</sup>  
Vianney Denis<sup>1</sup> Affendi Yang Amri<sup>14,15</sup> James D. Reimer<sup>16</sup> Takuma Mezaki<sup>17</sup> Charles Sheppard<sup>18</sup>  
Yossi Loya<sup>3</sup> Avidor Abelson<sup>3</sup> Mohammed Suleiman Mohammed<sup>19</sup> Andrew C. Baker<sup>20</sup> Pargol  
Ghavam Mostafavi<sup>21</sup> Budiyanto A. Suharsono<sup>22</sup> Chaolun Allen Chen<sup>1,8,23\*</sup>

<sup>1</sup>Biodiversity Research Centre, Academia Sinica, Nangang, Taipei 115, Taiwan

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

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

<sup>4</sup>Museum of Queensland, Townsville 4811, Australia

<sup>5</sup>James Cook University, Australia

<sup>6</sup>Coastal Ocean Research and Development of Indian Ocean (CORDIO), Mombasa, Kenya

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

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

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

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

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

<sup>12</sup>Taiwan Ocean Research Institute, National Applied Research Laboratories, Kaohsiung 852, Taiwan

<sup>13</sup>Mauritius Oceanography Institute, France Centre, Victoria Avenue Quatre Bornes, Mauritius

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

<sup>15</sup>The University of Western Australia, Crawley, 6009 Western Australia

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

<sup>17</sup>Biological Kuroshio Institute, Otuski, Kochi, Japan

<sup>18</sup>School of Life Sciences, University of Warwick, CV4 7AL, United Kingdom

<sup>19</sup>Institute of Marine Science, University of Dar es Salaam, Zanzibar, Tanzania

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

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

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

<sup>23</sup>Institute of Oceanography, National Taiwan University, Taipei 106, Taiwan

✉SK, SY and AA contributed equally to this work

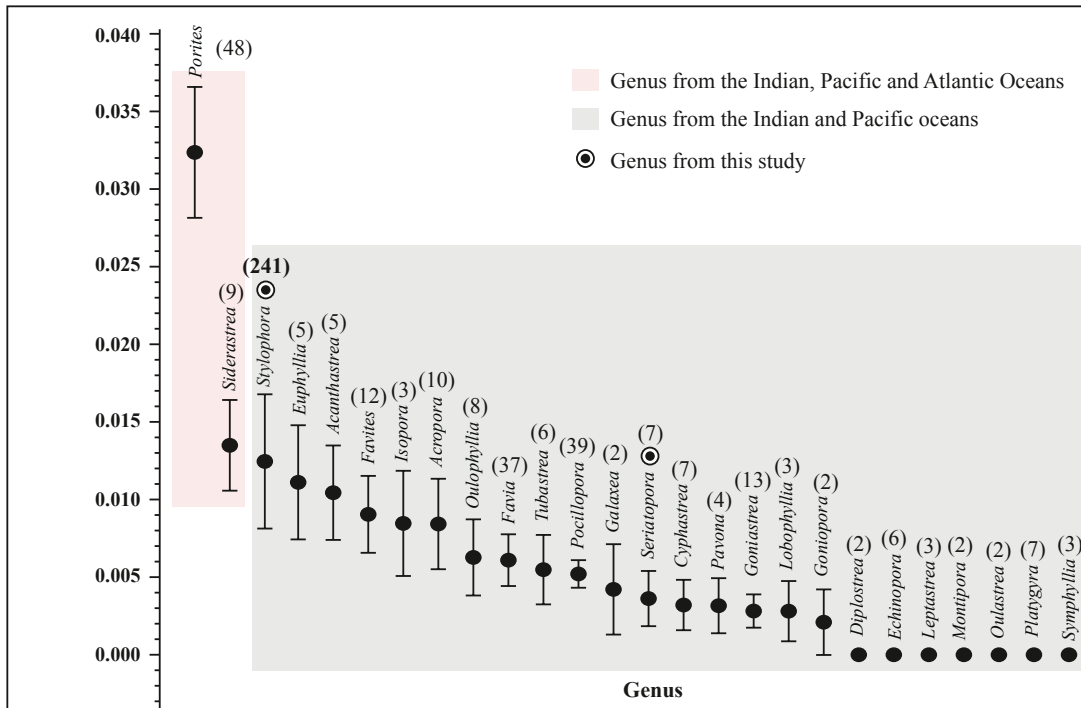
\*Correspondence to - cac@gate.sinica.edu.tw

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

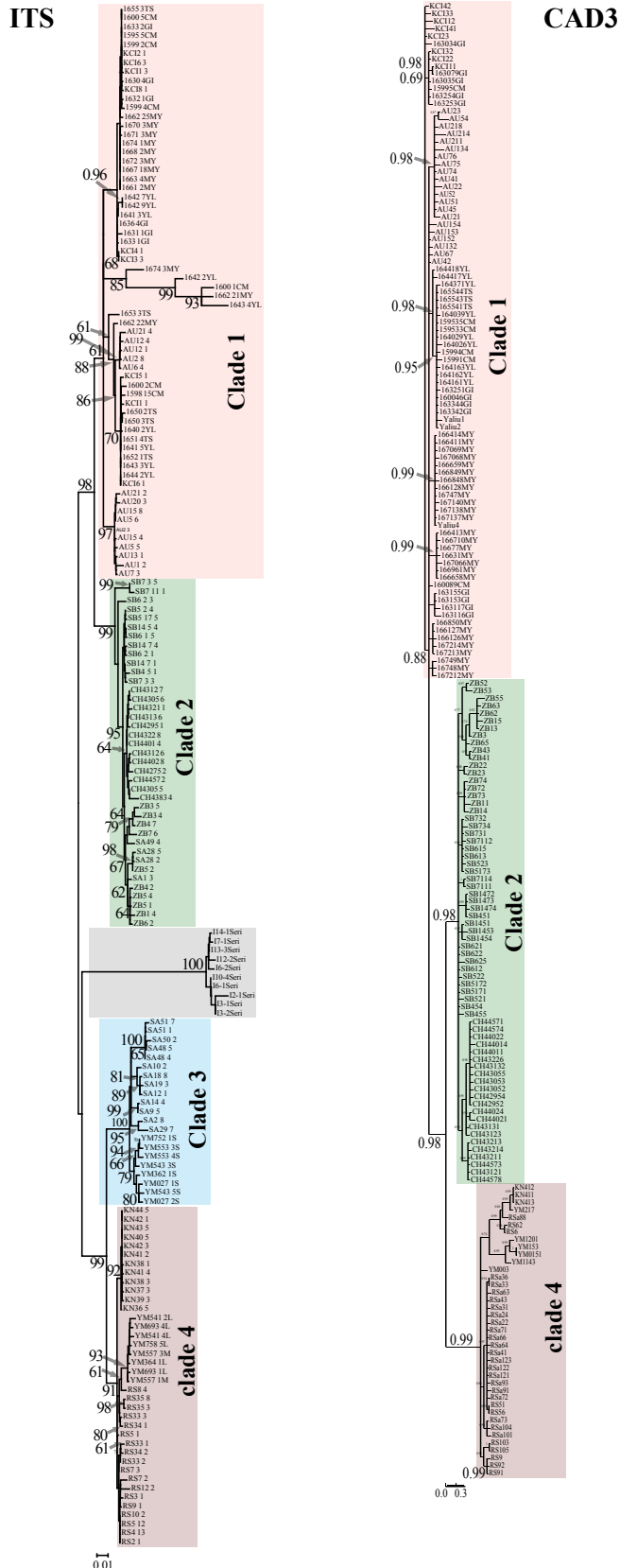
<b>p-distance</b>	Clade 1	Clade 2	Clade 3	Clade 4
Clade1	0.00017503± 0.00009578			
Clade 2	0.00378 ± 0.00248	0 ± 0		
Clade 3	0.01592 ± 0.00492	0.01584± 0.00495	0.00462174 ± 0.001932581	
Clade 4	0.01326 ± 0.00470	0.01317 ± 0.00465	0.01274 ± 0.00436	0.000175716 ±0.000117208

**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.

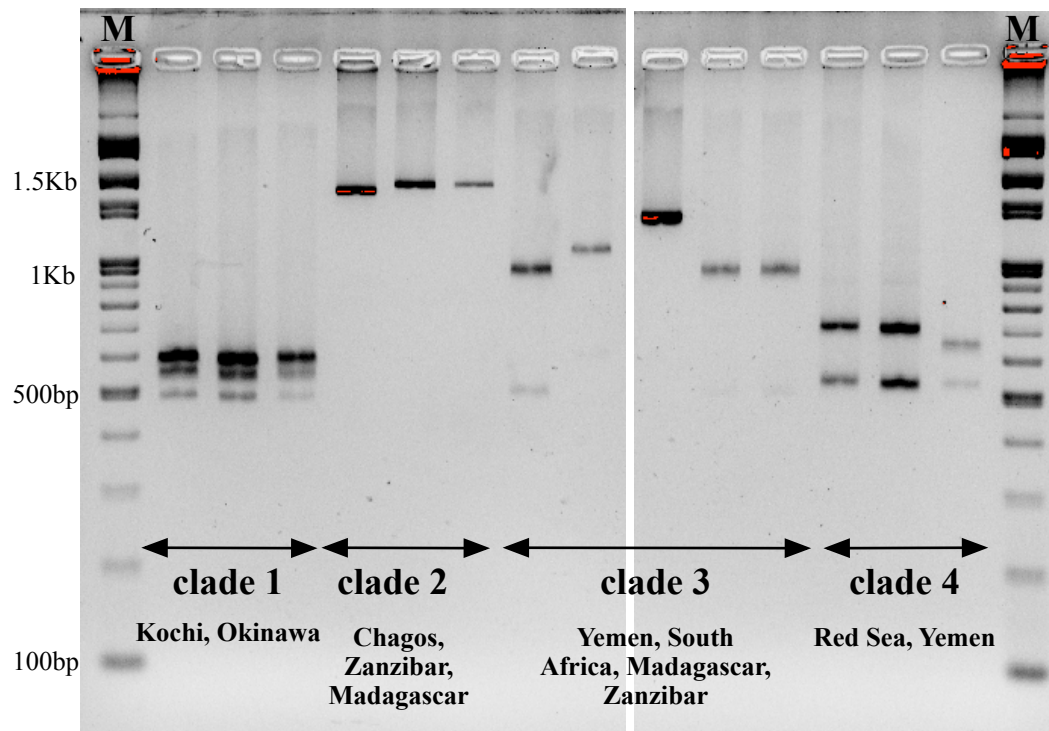
Location	COI bp changes / Amino acid changes	Length of CAD 3	tRNAW duplication	<i>Symbiodinium</i> clades
<b>Clade 1 Pacific Ocean</b>				
Australia (GBR)	7/3	500bp	—	*C
Taiwan	7/3	500bp	Yes	C
Japan	7/3	500bp	Yes	C
Malaysia	7/3	500bp	Yes	C
Indonesia	7/3	500bp		C
New Caledonia	7/3	500bp		C
<b>Clade 2 Indian Ocean</b>				
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
<b>Clade 3 <i>Stylophora madagascarensis</i></b>				
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
<b>Clade 4 Red Sea</b>				
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.