Supplementary Material

Consensus guidelines for advancing coral holobiont genome and specimen voucher deposition

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Supplementary Tables

Supplementary Table S1. Scleractinian corals and their associated Symbiodiniaceae microalgae targeted for sequencing for the "Coral symbiosis sensitivity to environmental change hub" under the "Aquatic Symbiosis Genomics Project".

Species	Type Locality (where available and collected)
Acropora cytherea	Tahiti
Acropora formosa	Fiji
Acropora palmata	
Acropora pulchra	Cocos <u>Keeling</u> Islands
Acropora spathulata	Solomo Islands
Agaricia agaricites	
Alveopora japonica	Shizuoka, Japan
Balanophyllia europaea	
Blastomussa merleti	New Caledonia
Blastomussa wellsi	
Colpophyllia natans	
Dendrogyra cylindrus	
Diploria labyrinthiformis	
Dipsastraea pallida	Fiji
Echinopora horrida	Fiji
Galaxea fascicularis	Indian Ocean
Goniastrea retiformis	Seychelles
Montastraea cavernosa	
Montipora capitata	Hawaii
Montipora digitata	Fiji
Mussismilia hispida	
Oculina arbuscula	
Orbicella franksi	
Pavona decussata	Fiji
Platygyra daedalea	Fiji
Pocillopora damicornis	Indo-Pacific
Porites cylindrica	Fiji
Pseudodiploria strigosa	
Seriatopora hystrix	Fiji
Siderastrea radians	
Siderastrea siderea	
Siderastrea stellata	
Stylophora subseriata	Red Sea
Turbinaria reniformis	Palm Islands, GBR
Mussismilia hispida	Brazil
Mussismilia braziliensis	Brazil
Oculina patagonia	Spain
Orbicella arbuscula	
Madrepora oculata	
Paragorgia sp.	
Javania sp.	

	Marker	Таха	Resolution (i.e., ability to differentiate); where available/applicable	Reference
mtDNA	Cyt-B (Cytochrome B)	Acropora spp.	Limited	(van Oppen et al., 1999)
	Cyt-B (Cytochrome B)	Scleractinia	Good	(Fukami et al., 2004b)
	Cyt-B (Cytochrome B)	Montipora spp.	Good	(Forsman et al., 2010)
	CR (Control region)	Acropora spp.	Limited (multi-specific subclades could be distinguished)	(van Oppen et al., 2001)
	CR (Control region)	Montipora spp.	Limited	(van Oppen et al., 2004)
	CR (Control region)	Pocillopora spp.	Good (Resolved 5 <i>Pocillopora</i> species)	(Flot et al., 2008)
	CR (Control region)	Seriatopora spp.	Good (Resolved 5 Seriatopora species)	(Chen et al., 2008)
	CR (Control region)	Montipora spp.	Good	(Forsman et al., 2010)
	CR (Control region)	Stylophora spp	Good	(Flot et al., 2011)
	CR (Control region)	Acropora spp.	Good	(Richards et al., 2008, 2013)
	CR (Control region)	Porites spp.	Good	(Terraneo et al., 2019)
	COI (Cytochrome c oxidase subunit I)	Orbicella annularis spp.	None	(Medina et al., 1999)
	COI (Cytochrome c oxidase subunit I)	Scleractinia	Good	(Fukami et al., 2004b)
	COI (Cytochrome c oxidase subunit I)	Montipora spp.	Good	(Forsman et al., 2010)
	COI (Cytochrome c oxidase subunit I)	Caulastraea spp.	Limited (multi-specific subclades could be distinguished)	(Arrigoni et al., 2021)
	COI (Cytochrome c oxidase subunit I)	Oulophyllia spp.	Limited (multi-specific subclades could be distinguished)	(Arrigoni et al., 2021)
	COI (Cytochrome c oxidase subunit I)	Astraeosmilia spp.	Limited (multi-specific subclades could be distinguished)	(Arrigoni et al., 2021)
	COI (Cytochrome c oxidase subunit I)	Lobophylliidae	Good	(Arrigoni et al., 2019)
	COI (Cytochrome c oxidase subunit I)	Coscinaraeidae	Good	(Benzoni et al., 2012)
	Non-coding region	Orbicella annularis spp.	Good	(Fukami et al., 2004a)
	Non-coding region	Pocillopora spp.	Good (resolved 5 species)	(Flot et al., 2008)
	nad5 (NADH dehydrogenase subunit 5)	Madracis spp.	none	(Frade et al., 2010)
	ATP6 (ATP synthase membrane subunit 6)	Montipora spp.	Good	(Forsman et al., 2010)
	ATP6-NAD4	Lobophylliidae	Good	(Arrigoni et al., 2019)
	NAD3-NAD5 (NADH dehydrogenase subunits 3- 5)	Lobophylliidae	Good	(Arrigoni et al., 2019)

Supplementary Table S2. Mitochondrial and nuclear markers used in coral taxonomy studies.

	(ORF) Open Reading Frame	Stylophora spp	Good	(Flot et al., 2011)
	125	Lobophylliidae	Good	(Arrigoni et al., 2019)
	16S	Scleractinia	Limited (broad, general patterns of evolution among Anthozoans)	(Romano and Palumbi, 1996, 1997)
	16S	Montipora spp.	Good	(Forsman et al., 2010)
	IGR (intergenic region)	Acropora hyacinthus and A. cytherea	None	(Marquez et al., 2002)
	IGR	Cyphastrea spp.	Good	(Arrigoni et al., 2017a)
	IGR	Pachyseris spp.	Good	(Terraneo et al., 2014)
	IGR	Agariciidae	Good	(Terraneo et al., 2017)
	IGR	Caulastraea spp.	Limited	(Arrigoni et al., 2021)
	IGR	<i>Oulophyllia</i> spp.	Limited	(Arrigoni et al., 2021)
	IGR	Astraeosmilia spp.	Limited	(Arrigoni et al., 2021)
	COI+IGR+mtMutSI	Octocorallia	Limited to good for species-level resolution	(McFadden et al., 2011)
	CO3+IGR-COI	Antipatharia	Limited	(Brugler et al., 2013)
	COI+16S	Zoantharia	Limited to good for species-level resolution	(Sinniger et al. 2008)
nDNA	28S	Anthozoa	Limited (broad, general patterns of evolution among anthozoans) to good for species-level resolution	(Chen et al., 1995; Veron et al., 1996)
	ITS (Internally Transcribed Spacer)	Porites spp.	Good (all 5 species could be distinguished)	(Hunter, 1997)
	ITS (Internally Transcribed Spacer)	Orbicella annularis spp.	None	(Lopez and Knowlton, 1997)
	ITS (Internally Transcribed Spacer)	Acropora spp (GBR)	Limited	(Odorico and Miller, 1997; Márquez et al., 2003)
	ITS (Internally Transcribed Spacer)	Orbicella annularis spp.	None	(Medina et al., 1999)
	ITS (Internally Transcribed Spacer)	<i>Acropora</i> spp (Caribbean)	Limited	(Oppen et al., 2000; Vollmer and Palumbi, 2004)
	ITS (Internally Transcribed Spacer)	<i>Madraci</i> s spp (Caribbean)	Good	(Diekmann et al., 2001)
	ITS (Internally Transcribed Spacer)	<i>Madracis</i> spp (Caribbean, Mediterranean, Indo- Pacific)	Good (resolved 4 morphs)	(Benzoni et al., 2018)
	ITS (Internally Transcribed Spacer)	Orbicella annularis spp.	Good	(Fukami et al., 2004a)
	ITS (Internally Transcribed Spacer)	Siderastrea spp	Limited	(Forsman et al., 2005)
	ITS (Internally Transcribed Spacer)	Scleractinia (8 different genera)	Good	(Wei et al., 2006)
	ITS (Internally Transcribed Spacer)	Pocillopora spp.	Good (resolved 2 species incl. hybridization)	(Combosch et al., 2008)
	ITS (Internally Transcribed Spacer)	Porites spp.	Good	(Forsman et al., 2009, 2015)

ITS (Internally Transcribed Spacer)	Montipora spp.	Good	(Forsman et al., 2010)
ITS (Internally Transcribed Spacer)	Stylophora spp	Good	(Flot et al., 2011)
ITS (Internally Transcribed Spacer)	Lobophylliidae	Good	(Arrigoni et al., 2019)
ITS (Internally Transcribed Spacer)	Caulastraea spp.	Good	(Arrigoni et al., 2021)
ITS (Internally Transcribed Spacer)	<i>Oulophyllia</i> spp.	Good	(Arrigoni et al., 2021)
ITS (Internally Transcribed Spacer)	Astraeosmilia spp.	Good	(Arrigoni et al., 2021)
ITS (Internally Transcribed Spacer)	Coscinaraeidae	Good	(Benzoni et al., 2012)
β-tubulin	Orbicella annularis spp.	None	(Lopez and Knowlton, 1997)
β-tubulin	Scleractinia	Good	(Fukami et al., 2004b)
β-tubulin	Psammocora spp.	Good	(Stefani et al., 2008)
PaxC intron	Acropora spp.	Good (Allele-frequency differences in Caribbean, multi-species subclades distinguishable at GBR)	(Oppen et al., 2000; van Oppen et al., 2001; Marquez et al., 2002; Richards et al., 2008, 2013)
PaxC intron	Montipora spp.	Limited	(van Oppen et al., 2004)
MiniCollagen intron	Acropora spp	Good	(Hatta et al., 1999; Vollmer and Palumbi, 2002)
Calmodulin intron	<i>Acropora</i> spp (Caribbean)	Limited (A. cervicornis vs. A. palmata)	(Vollmer and Palumbi, 2002)
SRP54 (Signal Recognition Particle 54- kDa)	Madracis spp.	None (Hybridization hypothesized)	(Frade et al., 2010)
ATPSα (ATP synthase subunit a/alpha)	Madracis spp.	Nnone (Hybridization hypothesized)	(Frade et al., 2010)
ATPs β (ATP synthase subunit b/beta)	Montipora spp.	Limited	(Forsman et al., 2010)
ATPs β (ATP synthase subunit b/beta)	Pocillopora spp.	Limited	(Flot et al., 2010)
SSU rRNA	Scleractinia	Resolves families and species within some families, esp. Acroporidae	(Arrigoni et al., 2017b)
H3 (Histone 3)	Lobophylliidae	Limited	(Arrigoni et al., 2019)
H3 (Histone 3)	Caulastraea spp.	Limited	(Arrigoni et al., 2021)
H3 (Histone 3)	<i>Oulophyllia</i> spp.	Limited	(Arrigoni et al., 2021)
H3 (Histone 3)	Astraeosmilia spp.	Limited	(Arrigoni et al., 2021)
H3 (Histone 3)	Cyphastrea spp.	Limited	(Arrigoni et al., 2017a)

Supplementary Table S3. Summary of information available from coral genomes, published between 2011-2021.

		Metadata							
Species	Sample collection site	Sample collection GPS	Colony picture	Skeleton picture	Phylogenetic information	Raw seq. data available	Transcriptome available	Notes	Reference
Scleractinia	1								
Acropora acuminata	Okinawa, Japan	no	yes	no	no	yes	no		(Shinzato et al., 2021)
Acropora awi	Okinawa, Japan	no	yes	no	no	yes	no		(Shinzato et al., 2021)
Acropora cervicornis	Florida, USA	yes	yes	no	yes	yes	yes	Coral colony tagged in the Florida Keys	(Kitchen et al., 2019)
Acropora cytherea	Okinawa, Japan	no	yes	no	no	yes	no		(Shinzato et al., 2021)
Acropora digitifera	Okinawa, Japan	no	yes	no	no	no	yes	Colony fragment maintained at Sekoto Station aquarium	(Shinzato et al., 2011)
Acropora digitifera	Okinawa, Japan	no	yes	no	no	yes	yes		(Shinzato et al., 2021)
Acropora echinata	Okinawa, Japan	no	yes	no	no	yes	no		(Shinzato et al., 2021)
Acropora florida	Okinawa, Japan	no	yes	no	no	yes	no		(Shinzato et al., 2021)
Acropora gemmifera	Okinawa, Japan	no	yes	no	no	yes	no		(Shinzato et al., 2021)
Acropora hyacinthus	Okinawa, Japan	no	yes	no	no	yes	no		(Shinzato et al., 2021)
Acropora intermedia	Okinawa, Japan	no	yes	no	no	yes	no		(Shinzato et al., 2021)
Acropora microphthalma	Okinawa, Japan	no	yes	no	no	yes	no		(Shinzato et al., 2021)
Acropora millepora	Magnetic Island, Australia	yes	yes*	no	yes (mt genome)	yes	yes		(Ying et al., 2019)
Acropora millepora	Central GBR, Australia	yes	yes*	?	no	yes	no		(Fuller et al., 2020)
Acropora muricata	Okinawa, Japan	no	yes	no	no	yes	no		(Shinzato et al., 2021)
Acropora nasuta	Okinawa, Japan	no	yes	no	no	yes	no		(Shinzato et al., 2021)
Acropora palmata	Florida, USA	yes	yes	no	yes	yes	yes	Coral colony tagged in the Florida Keys	(Kitchen et al., 2019)
Acropora selago	Okinawa, Japan	no	yes	no	no	yes	no		(Shinzato et al., 2021)
Acropora tenuis	Okinawa, Japan	no	yes	no	no	yes	yes		(Shinzato et al., 2021)
Acropora yongei	Okinawa, Japan	no	yes	no	no	yes	no		(Shinzato et al., 2021)
Astreopora myriophthalma	Okinawa, Japan	no	yes	no	no	yes	no		(Shinzato et al., 2021)
<i>Fungia</i> sp.	Orpheus Island, Australia	no	yes*	yes*	no	yes	yes		(Ying et al., 2018)
Galaxea fascicularis	Orpheus Island, Australia	no	yes*	yes*	no	yes	yes		(Ying et al., 2018)

Goniastrea aspera	Orpheus Island, Australia	no	yes*	yes*	no	yes	yes		(Ying et al., 2018)
Montipora cactus	Okinawa, Japan	no	yes	no	no	yes	no		(Shinzato et al., 2021)
Montipora capitata	Hawaii, USA	no	no	no	no	yes	yes		(Shumaker et al., 2019)
Montipora capitata	Hawaii, USA	yes	yes*	no	no	yes	no		(Helmkampf et al., 2019)
Montipora efflorescens	Okinawa, Japan	no	yes	no	no	yes	no		(Shinzato et al., 2021)
Orbicella annularis	Puerto Morelos, MX / Bocas del Toro, PA	yes	no	no	no	yes	no		(Prada et al., 2016)
Orbicella faveolata	Florida, USA; Puerto Morelos, MX; Bocas del Toro, PA	yes	no	no	no	yes	yes	Colony fragment maintained at RSMAS coral husbandry facility	(Prada et al., 2016)
Orbicella franski	Puerto Morelos, MX; Bocas del Toro, PA	yes	no	no	no	yes	no		(Prada et al., 2016)
Pocillopora damicornis	Saboga Island, PA	no	no	no	no	no	no		(Cunning et al., 2018)
Pocillopora verrucosa	Central Red Sea, Saudi Arabia	yes	yes	yes	yes (mtORF)	yes	yes	Skeletal voucher picture on github	(Buitrago- López et al., 2020)
Porites lutea	Orpheus Island, Australia	yes	no	no	no	yes	yes		(Robbins et al., 2019)
Porites rus	Indonesia	no	no	no	no	yes	yes	Colony fragment is maintained at JLU Giessen	(Celis et al., 2018)
Stylophora pistillata	Gulf of Aqaba, Jordan	no	yes	yes	yes (ITS and COI)	yes	yes	Colony maintained at CSM	(Voolstra et al., 2017)
Tubastraea tagusensis	Angra dos Reis, Brazil	yes	yes	yes	yes	yes (once published)	yes (once published)		(Soares- Souza et al., 2020)
Tubastraea coccinea	Angra dos Reis, Brazil	yes	yes	yes	yes	yes (once published)	yes (once published)		(Soares- Souza et al., 2020)
<i>Tubastraea</i> sp.	Arrail do Cabo, Brazil	yes	yes	yes	yes	yes (once published)	yes (once published)		(Soares- Souza et al., 2020)
Octocorallia	•				•				•
Dendronephthya gigantea	Jeju Island, South Korea	yes	no	no	no	yes	yes		(Jeon et al., 2019)
Paramuricea clavata	Catalunya, Spain	yes	no	no	no	yes	yes		(Ledoux et al., 2020)
Renilla muelleri	Gulf of Mexico, USA	no	no	no	no	yes	yes	Specimen obtained from aquarium shops. Collection site unknown.	(Jiang et al., 2019)
<i>Trachythela</i> sp.	Xisha Trough, South China Sea	yes	yes	no	no	yes	no	deep-sea	(Zhou et al., 2021)
<i>Xenia</i> sp.	local coral aquarium shop called CTE Aquatics, USA	no	yes	no	yes	yes	yes	Colony maintained in aquaria; frozen and fixed coral colonies, gDNA, RNA deposited at the Smithsonian National Museum of Natural History (catalogue no., USNM 1613385).	(Hu et al., 2020)

*The provided photograph is not from the colony used for genome assembly.

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Supplementary Table S4. Summary of information available from 15 Symbiodiniaceae genomes, published between 2013-2021.

Species	Strain	Sample collection GPS	Isolation source	Voucher image [light, SEM/TEM microscopy	Phylogenetic information	Raw seq. data available	Transcriptome available	Reference
Symbiodinium microadriaticum	CCMP2467	no	no	no, no	yes	yes	yes	(Aranda et al., 2016)
Symbiodinium microadriaticum	CCMP2467- SC	no	no	no, no	no	yes	no	(Nand et al., 2020)
Symbiodinium microadriaticum	04- 503SCI.03	no	no	no, no	no	yes	yes	(González-Pech et al., 2021)
Symbiodinium microadriaticum	CassKB8	no	no	no, no	no	yes	yes	(González-Pech et al., 2021)
Symbiodinium necroappetens	CCMP2469	no	no	no, no	no	yes	no	(González-Pech et al., 2021)
Symbiodinium linucheae	CCMP2456	no	no	no, no	no	yes	yes	(González-Pech et al., 2021)
Symbiodinium tridacnidorum	CCMP2592	no	no	no, no	no	yes	yes	(González-Pech et al., 2021)
Symbiodinium natans	CCMP2548	no	no	no, no	no	yes	yes	(González-Pech et al., 2021)
Symbiodinium pilosum	CCMP2461	no	no	no, no	no	yes	yes	(González-Pech et al., 2021)
Symbiodinium sp. Y-106, type A3	NIES-4076	no	yes	yes, no	yes	yes	yes	(Shoguchi et al., 2018)
Breviolum minutum	Mf1.05b.01	no	yes	yes, yes*	no	no	yes	(Shoguchi et al., 2013)
<i>Cladocopium</i> sp. Y103, clade C	NIES-4077	no	yes	yes, no	yes	yes	yes	(Shoguchi et al., 2018)
Cladocopium goreaui	SCF055-01	no	yes	no, no	yes	yes	no	(Liu et al., 2018)
Fugacium kawagutii	CCMP2468	no	yes	yes, yes		yes	yes	(Lin et al., 2015)
Fugacium kawagutii	CCMP2468	no	no	no, no	yes	yes	no	(Liu et al., 2018)

*SEM image for the purpose of showing surface contamination

Supplementary Table S5. Overview of 74 coral bacterial isolates with their genomes sequences available in public databases (NCBI; IMG/DOE-JGI). Data taken from (Sweet et al., 2020).

Isolate name	Bacterial family (class)	Host scientific name (coral type)	Project Accession	Sample Accession	Assembly Accession
<i>Janibacter corallicola</i> NBRC 107790	<i>Intrasporangiaceae</i> (Actinobacteria)	Acropora gemmifera (hexacoral)	PRJDB231	SAMD00046480	GCA_001570965.1
<i>Dietzia</i> sp. WMMA184	<i>Dietziaceae</i> (Actinobacteria)	<i>Montastraea faveolata</i> (hexacoral)	PRJNA400578	SAMN07571474	GCA_002441635.1
<i>Cytophaga</i> sp. FL35	<i>Cytophagaceae</i> (Cytophagia)	<i>Montastraea cavernosa</i> mucus (hexacoral)	PRJNA343499	SAMN05805725	JACSOH000000000
Aquimarina megaterium EL33	<i>Flavobacteriaceae</i> (Flavobacteriia)	<i>Eunicella labiata</i> (octocoral)	PRJEB14417	SAMEA4034450	GCA_900089995.1
Paracoccus isoporae DSM 22220	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	Isopora polifera (hexacoral)	PRJNA329835	SAMN05421538	GCA_900101865.1
Paracoccus sp. R12_1	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	Acropora cytherea (hexacoral)	PRJNA698462	SAMN18440239	JAGHQZ000000000
Paracoccus sp. R12_2	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	Acropora hemprichii (hexacoral)	PRJNA698462	SAMN18440240	JAGHRA000000000
<i>Shimia</i> sp. R9_1	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	<i>Acropora hemprichii</i> (hexacoral)	PRJNA698462	SAMN18440253	JAGHRN000000000
Shimia sp. R9_2	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	<i>Acropora cytherea</i> (hexacoral)	PRJNA698462	SAMN18440254	JAGHRO000000000
Shimia sp. R9_3	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	<i>Acropora humilis</i> (hexacoral)	PRJNA698462	SAMN18440255	JAGHRP000000000
<i>Shimia</i> sp. R10_1	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	<i>Acropora cytherea</i> (hexacoral)	PRJNA698462	SAMN18440237	JAGHQX000000000
<i>Shimia</i> sp. R11_0	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	<i>Acropora hemprichii</i> (hexacoral)	PRJNA698462	SAMN18440238	JAGHQY000000000
<i>Ruegeria</i> sp. EL01	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	<i>Eunicella labiata</i> (octocoral)	PRJEB25531	SAMEA104692609	GCA_900313035.1
<i>Ruegeria</i> sp. R8_1	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	<i>Acropora humilis</i> (hexacoral)	PRJNA698462	SAMN18440251	JAGHRL000000000
<i>Ruegeria</i> sp. R8_2	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	<i>Acropora cytherea</i> (hexacoral)	PRJNA698462	SAMN18440252	JAGHRM000000000
<i>Ruegeria</i> sp. R13_0	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	<i>Acropora cytherea</i> (hexacoral)	PRJNA698462	SAMN18440241	JAGHRB000000000
<i>Ruegeria</i> sp. R14_0	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	<i>Acropora cytherea</i> (hexacoral)	PRJNA698462	SAMN18440242	JAGHRC000000000
Sulfitobacter sp. EL44	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	<i>Eunicella labiata</i> (octocoral)	PRJEB25531	SAMEA104692611	GCA_900313045.1
Sulfitobacter sp. R18_1	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	<i>Acropora humilis</i> (hexacoral)	PRJNA698462	SAMN18440246	JAGHRG000000000
Sulfitobacter sp. R18_2	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	<i>Acropora humilis</i> (hexacoral)	PRJNA698462	SAMN18440247	JAGHRH000000000
<i>Tropicibacter</i> sp. R15_0	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	<i>Acropora hemprichii</i> (hexacoral)	PRJNA698462	SAMN18440243	JAGHRD000000000
<i>Tropicibacter</i> sp. R16_0	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	<i>Acropora cytherea</i> (hexacoral)	PRJNA698462	SAMN18440244	JAGHRE000000000
<i>Nautella</i> sp. R17_0	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	<i>Acropora humilis</i> (hexacoral)	PRJNA698462	SAMN18440245	JAGHRF000000000
<i>Roseovarius</i> sp. EL26	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	<i>Eunicella labiata</i> (octocoral)	PRJEB25531	SAMEA1059311	GCA_900327775.1
Pseudophaeobacter sp. EL27	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	<i>Eunicella labiata</i> (octocoral)	PRJEB25531	SAMEA104692610	GCA_900313025.1
Rhodobacteraceae bacterium	Rhodobacteraceae	Eunicella labiata	PRJEB25531	SAMEA104692612	GCA_900313015.1

EL53	(Alphaproteobacteria)	(octocoral)			
<i>Rhodobacteraceae</i> bacterium EL129	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	<i>Eunicella labiata</i> (octocoral)	PRJEB25531	SAMEA104692613	GCA_900313005.1
Stappia indica EBBD 17.2	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	black-band-diseased hexacoral	PRJNA328867	SAMN05382895	GCA_001696545.1
Stappia indica SBBC49	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	black-band-diseased hexacoral	PRJNA328866	SAMN05382876	GCA_001696535.1
Pseudovibrio sp. FO-BEG1*	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	undefined hexacoral	PRJNA73563	SAMN02261085	GCA_000236645.1
Labrenzia alba EL143	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	<i>Eunicella labiata</i> (octocoral)	PRJEB24678	SAMEA104555632	GCA_900258415.1
<i>Labrenzia</i> sp. R4_1	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	<i>Acropora hemprichii</i> (hexacoral)	PRJNA698462	SAMN18440248	JAGHRI000000000
<i>Labrenzia</i> sp. R4_2	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	Acropora cytherea (hexacoral)	PRJNA698462	SAMN18440249	JAGHRJ000000000
<i>Labrenzia</i> sp. R5_0	<i>Rhodobacteraceae</i> (Alphaproteobacteria)	Acropora cytherea (hexacoral)	PRJNA698462	SAMN18440250	JAGHRK000000000
Sphingorhabdus sp. EL138	<i>Sphingomonadaceae</i> (Alphaproteobacteria)	<i>Eunicella labiata</i> (octocoral)	PRJEB24502	SAMEA104500216	GCA_900258045.1
<i>Erythrobacter</i> sp. A6_0	<i>Erythrobacteraceae</i> (Alphaproteobacteria)	<i>Acropora humilis</i> (hexacoral)	PRJNA698462	SAMN18440231	JAGHQR000000000
<i>Erythrobacter</i> sp. A7_0	<i>Erythrobacteraceae</i> (Alphaproteobacteria)	<i>Acropora cytherea</i> (hexacoral)	PRJNA698462	SAMN18440232	JAGHQS000000000
<i>Nitratireductor aquibiodomus</i> EBB 35.1	<i>Phyllobacteriaceae</i> (Alphaproteobacteria)	black-band-diseased hexacoral	PRJNA327597	SAMN05348542	GCA_001696575.1
<i>Kiloniella</i> sp. EL199	<i>Kiloniellaceae</i> (Alphaproteobacteria)	<i>Eunicella labiata</i> (octocoral)	PRJEB25501	SAMEA104692614	GCA_900313065.1
Thalassospira sp. A3_1	<i>Rhodosprilillacaeae</i> (Alphaproteobateria)	<i>Acropora cytherea</i> (hexacoral)	PRJNA698462	SAMN18440230	JAGHQQ000000000
<i>Brevundimonas</i> sp. A19_0	<i>Caulobacteraceae</i> (Alphaproteobacteria)	<i>Acropora cytherea</i> (hexacoral)	PRJNA698462	SAMN18440229	JAGHQP000000000
<i>Endozoicomonas acroporae</i> Acr-14	<i>Endozoicomonadaceae</i> (Gammaproteobacteria)	Acropora spp. (hexacoral)	PRJNA422318	SAMN08174373	GCA_002864045.1
Endozoicomonas montiporae CL-33	Endozoicomonadaceae (Gammaproteobacteria)	Montipora aequituberculata (hexacoral)	PRJNA66389	SAMN04155633	GCA_001583435.1
Endozoicomonas sp. G2_1	<i>Endozoicomonadaceae</i> (Gammaproteobacteria)	Acropora cytherea (hexacoral)	PRJNA698462	SAMN18440233	JAGHQT000000000
Endozoicomonas sp. G2_2	<i>Endozoicomonadaceae</i> (Gammaproteobacteria)	Acropora hemprichii (hexacoral)	PRJNA698462	SAMN18440234	JAGHQU000000000
<i>Marinomonas fungiae</i> AN44 (JCM 18476)	Oceanosprilliaceae (Gammaproteobacteria)	<i>Fungia echinata</i> (hexacoral)	PRJNA289015	SAMN03840741	GCA_001418205.1
Cobetia marina BMC 6	<i>Halomonadaceae</i> (Gammaproteobacteria)	Pocillopora damicornis (hexacoral)	PRJNA638634	SAMN15198645	GCA_013350055.1
Halomonas taeanensis BMC 7	<i>Halomonadaceae</i> (Gammaproteobacteria)	Pocillopora damicornis (hexacoral)	PRJNA638634	SAMN15198646	GCA_013349955.1
Halomonas meridiana R1t3	<i>Halomonadaceae</i> (Gammaproteobacteria)	<i>Acropora palmata</i> mucus (hexacoral)	PRJNA269585	SAMN03255752	GCA_000943375.1
Alteromonas sp. BZK5	<i>Alteromonadaceae</i> (Gammaproteobacteria)	O <i>rbicella annularis</i> mucus (hexacoral)	PRJNA343499	SAMN05805728	JASCOE000000000
Pseudoalteromonas luteoviolacea HI1**	Pseudoaltermonadaceae (Gammaproteobacteria)	Pocillopora damicornis (hexacoral)	PRJNA257109	SAMN02947394	GCA_000814765.1
Pseudoalteromonas sp. BMC 1	<i>Pseudoaltermonadaceae</i> (Gammaproteobacteria)	Pocillopora damicornis (hexacoral)	PRJNA638634	SAMN15198644	GCF_013350085.1
Pseudoalteromonas sp. BMC 2	<i>Pseudoaltermonadaceae</i> (Gammaproteobacteria)	Pocillopora damicornis (hexacoral)	PRJNA638634	SAMN15198640	GCA_013349945.1
Pseudoalteromonas sp. BMC 3	Pseudoaltermonadaceae	Pocillopora damicornis	PRJNA638634	SAMN15198643	GCA_013349995.1

	(Gammaproteobacteria)	(hexacoral)			
Pseudoalteromonas sp. BMC 4	Pseudoaltermonadaceae (Gammaproteobacteria)	Pocillopora damicornis (hexacoral)	PRJNA638634	SAMN15198641	GCA_013350045.1
Pseudoalteromonas sp. BMC 5	Pseudoaltermonadaceae (Gammaproteobacteria)	Pocillopora damicornis (hexacoral)	PRJNA638634	SAMN15198642	GCA_013349985.1
Pseudoalteromonas sp. BZK2	Pseudoaltermonadaceae (Gammaproteobacteria)	Orbicella annularis mucus (hexacoral)	PRJNA343499	SAMN05805727	JACSOG000000000
Thallassotalea euphylliae H1	Colwelliaceae (Gammaproteobacteria)	Montipora sp. (hexacoral)	PRJNA484286	SAMN09762563	GCA_003390335.1
Thallassotalea sp. G20_0	Colwelliaceae (Gammaproteobacteria)	Acropora hemprichii (hexacoral)	PRJNA698462	SAMN18440235	JAGHQV000000000
Salinisphaera sp. G21_0	Salinisphaeraceae (Gammaproteobacteria)	Acropora cytherea (hexacoral)	PRJNA698462	SAMN18440236	JAGHQW000000000
Luteimonas sp. JM171	<i>Xanthomonadaceae</i> (Gammaproteobacteria)	Porites lobata (hexacoral)	PRJNA340268	SAMN05711034	GCA_001717465.1
Photobacterium sp. BZF1	<i>Vibrionaceae</i> (Gammaproteobacteria)	O <i>rbicella annularis</i> mucus (hexacoral)	PRJNA343499	SAMN05805726	JACSOF000000000
Aliivibrio sp. EL58	<i>Vibrionaceae</i> (Gammaproteobacteria)	<i>Eunicella labiata</i> (octocoral)	PRJEB25451	SAMEA104674031	GCA_900312675.1
Vibrio crassostreae Evh12	<i>Vibrionaceae</i> (Gammaproteobacteria)	<i>Eunicella verrucosa</i> (octocoral)	PRJEB10717	SAMEA3532819	GCA_001486525.1
<i>Vibrio</i> sp. Evh13	<i>Vibrionaceae</i> (Gammaproteobacteria)	<i>Eunicella verrucosa</i> (octocoral)	PRJEB26006	SAMEA4610035	GCA_900382745.1
<i>Vibrio</i> sp. Evd3	<i>Vibrionaceae</i> (Gammaproteobacteria)	<i>Eunicella verrucosa</i> (octocoral)	PRJEB26006	SAMEA4591840	GCA_900379685.1
<i>Vibri</i> o sp. Evd11	<i>Vibrionaceae</i> (Gammaproteobacteria)	<i>Eunicella verrucosa</i> (octocoral)	PRJEB26006	SAMEA4610036	GCA_900382755.1
Vibrio fortis S7-72	<i>Vibrionaceae</i> (Gammaproteobacteria)	<i>Fungia</i> sp. (hexacoral)	PRJNA565475	SAMN12748540	GCA_008756975.1
Vibrio fortis AN60	<i>Vibrionaceae</i> (Gammaproteobacteria)	Fungia sp. (hexacoral)	PRJNA565152	SAMN12736039	GCA_008756925.1
Vibrio harveyi 1DA3***	<i>Vibrionaceae</i> (Gammaproteobacteria)	<i>Phyllogorgia dilatata</i> (hexacoral)	PRJNA40361	SAMN02470250	GCA_000182685.1
Vibrio alginolyticus 40B***	<i>Vibrionaceae</i> (Gammaproteobacteria)	<i>Mussismilia hispida</i> (hexaoral)	PRJNA40359	SAMN02470249	GCA_000176055.1
Vibrio shilonii AK1***	<i>Vibrionaceae</i> (Gammaproteobacteria)	Oculina patagonica (hexacoral)	PRJNA19397	SAMN02436099	GCA_000181535.1
<i>Vibrio coralliilyticus</i> ATCC BAA-450***	<i>Vibrionaceae</i> (Gammaproteobacteria)	Pocillopora damicornis (hexacoral)	PRJNA40491	SAMN02393819	GCA_000176135.1
Vibrio coralliilyticus OCN008***	<i>Vibrionaceae</i> (Gammaproteobacteria)	<i>Montipora capitata</i> (hexacoral)	PRJNA214563	SAMN02304133	GCA_000461895.1

Supplementary Methods

I. Coral collection

Prior to collection in the field, ensure to consult any available type-specific information. This may include original taxonomic descriptions and illustrations, or morphological taxonomic information based on the re-examination of type material. To assist sample collection while SCUBA diving, you may print and plastify reference images or save them on the SD card of your underwater camera and use them as references for identification *in situ*.

For each coral, select a colony or an individual that is representative of an adult of its species. This can be challenging and variable by species, but in general younger/smaller colonies tend to lack diagnostic features.

If possible, it is advised to tag the colony in the field or, if extracted from the field, to keep the individual in an aquarium facility. This ensures re-sampling of the originally selected coral in case of need; in the case of field-tagging, it provides the opportunity for long-term monitoring and evaluation.

If sampling multiple corals of the same species at the same reef site, it is best to minimize the chance of collecting clones by adhering to sufficient distance between colonies (min. 5m, ideal > 20m between colonies).

In situ pre- and post- collection images

For all taxa collected during SCUBA diving:

- 1. Wide shots to provide a reference of the coral appearance in the field, with 0.5 m or 1 m square size reference. These types of images should ideally include the whole selected coral as well as the surrounding areas to provide environmental context and provide support reference for potential retrieval.
- 2. Whole colony shot or full view of an individual in case of solitary taxa (Figure S1 A-C).
- 3. Documentation of the coral colour *in vivo*. If possible, capture a coral colour reference card in the photo (Siebeck et al. 2006).
- 4. Close-up of the coral surface to show detail of polyp(s) and coenosarc (Figure S1 D-F).
- 5. Shot of dive computer close to the base of the coral to record unambiguously the collection depth (inset Figure S1 G-I).
- 6. Shot of the polyp of fragment(s) (Figure S1 G-I). This is important in case more than one coral is sampled during the same dive to ensure correct post-dive match of *in situ* images with the collection code.

For Acropora and other branching colonial taxa:

- 7. Coral base (make sure it is attached and growing *in-situ*, secondarily free-living corals could attain aberrant morphologies)
- 8. Close-up of the branch tips (Figure S1 F).
- 9. Especially if table or plate, shots of corallites around edge of the colony
- 10. Skeletal specimens should be 20-30 cm in diameter.

Metadata

In general, the more information is consistently available about the collection, the better. The minimum required information include:

- 1. Latitude and longitude of the collection site, any toponym or reef name is also useful.
- 2. Collection date and time.
- 3. For SCUBA diving collection, collector name. For ROV or SUB collection, pilot and scientist names.
- 4. Depth at the collection point. For shallow specimens, time of collection to know tide level will also be important.
- 5. Water temperature at collection point.
- 6. Reef zone (e.g. flat, lagoon, inner or outer crest), or site profile/geomorphology for mesophotic and deep corals.

Skeletal Voucher Images

After labelling with a uniquely identifying collection code (see next section for details), the voucher should be documented via a set of shots.

For all taxa:

- 1. Voucher shot including the whole corallum, its label and a reference scale (Figure S1 J-L). This can be a ruler or a soft tailor meter that more easily follows the contour of very three-dimensional vouchers.
- Close-up of the voucher surface to show the corallite(s) and other superficial macro-structures (e.g. costosepta, coenosteum structure) (Figure S1 M-O). A digital camera with a decent macro setting can provide good results, however in case of highly three-dimensional or spiky coralla a stack of images can help obtain a final image with better depth of field (Figure S1 O).

For taxa with small corallites (<3mm in diameter, e.g. *Montipora*, *Porites*, *Pavona*, *Madracis*) Scanning Electron Microscopy (optional):

- 3. Grind, mount on stub and coat with Au or Au-Pd a fragment of the voucher.
- 4. Save high resolution images of corallites and coenosteum ornamentation (Figure S1 Q).

For Acropora:

- 5. Lateral (Figure S1 O) and top shots of the radial corallites.
- 6. Axial corallite from the top.
- 7. Coenosteum ornamentation (Figure S1 R).

Skeletal Voucher Curation

 In the case of stony corals (Scleractinia), skeletal vouchers should be stored as whole or partially dried skeletons, cleaned of tissue, either in 70% EtOH or formalin (4% aldehyde in water). Octocorals (Octocorallia) and black corals (Antipatharia) should be preserved in 70% EtOH or dried, depending on size. All skeletons should be labelled with a uniquely identifying collection code, securely affixed or attached to the skeleton.

Collection metadata will be linked to this code. The specimen will receive a museum registration number once it is registered in a collection, but the original field code can be included in the metadata to link the field data to the specimen. The collection code then becomes part of the specimen history, together with any previously assigned label, and remains in the metadata. Likewise, all metadata associated with the specimen should be included when registered in the Museum.

- 2. Skeletal vouchers with collection code should be kept in a dust-free environment or in a plastic ziplock bag at room temperature.
- 3. Skeletal vouchers can be deposited in natural history museums and the registration number made available and added to the metadata to ensure further re-examination. Some potential locations are listed hereafter. This list is by no means comprehensive and includes museums where historical coral collections, including type material, are housed, curated and accessible for further re-examination. Where possible, attempts should be made to ensure specimens or at least duplicates remain in-country. In some countries permits will include requirements that any type material is deposited in-country. At the same time, the need to decolonize research must be balanced by the need to ensure that valuable specimens will be housed in properly curated facilities that minimize the chance that valuable specimens will be lost, damaged or destroyed.

List of potential voucher repositories

- National Museum of Natural History, Smithsonian Institution, Washington, DC, USA: https://naturalhistory.si.edu/research/invertebrate-zoology
- American Museum of Natural History, New York, NY, USA: https://www.amnh.org/research/invertebrate-zoology
- Natural History Museum, London, UK: <u>https://www.nhm.ac.uk/our-science/collections/accessing-collections.html</u>
- Naturalis Biodiversity Center, Leiden, The Netherlands: https://www.naturalis.nl/en/collection
- Muséum national d'Histoire naturelle, Paris, France: <u>https://www.mnhn.fr/en/collections/collection-groups/marine-invertebrates</u>
- Museum for Naturkunde, Berlin, Germany: https://www.museumfuernaturkunde.berlin/en/science/infrastructure/collection/onlinecollections
- Natural History Museum Vienna, Vienna, Austria: <u>https://www.nhm-</u> wien.ac.at/en/research/3_zoology_invertebrates/collection/collection_evertebrata_varia
- Marine Science Institute, University of the Philippines, Quezon City: http://www.msi.upd.edu.ph/library

- National Museum in Tokyo, Japan: https://www.kahaku.go.jp/english/research/index.html
- UOG Biorepository, University of Guam, Guam: https://specifyportal.uog.edu/
- Museum of Tropical Queensland, Townsville, Australia: https://collections.qm.qld.gov.au/explore
- Bishop Museum, Hawaii USA: https://www.bishopmuseum.org/invertebrate-zoology



Supplementary Figure S1. Example of *in situ* (A-I) and post-collection (J-R) images of corals and corresponding vouchers for taxa with large polyps and corallites (*Platygyra daedalea*), small (*Pavona varians*), and for the genus *Acropora* having a distinctive and diagnostic dimorphism of polyps and corallites (O). A-C whole colony shots; D-F close ups; G-I fragments post collection and collection depth (inset); J-L vouchers with collection code and scale; M-O close ups of the corallum showing corallites and other macro-skeletal features; P-R meso skeletal features images at the stereo (P, R) or Scanning Electron Microscope (Q).

Coral tissue/sperm preservation and handling

Degradation of DNA can be problematic for corals (some species have high nuclease activity). Preservation straight after sampling and prior to freezing can help prevent DNA degradation. When extended periods post sampling and prior to freezing occur, it is recommended that coral fragments be immediately placed in DESS solution (also known as DMSO/EDTA/NaCl preservation buffer) prior to freezing. In addition, if only -20°C storage is possible then DESS can help prevent DNA degradation. This buffer 'pickles' samples and is used to preserve tissue for later DNA extraction (Yoder et al., 2006; Mulcahy et al., 2016; Ziegler et al., 2017; Robbins et al., 2019). DESS buffer is very convenient in the field and for sample transport as it is not toxic, does not require special treatment, or special sample storage. Nevertheless, we recommend reviewing chemical handling procedures and permits as relevant per country. Prepare the buffer under a fume hood, as both EDTA and DMSO are irritants. Wear rubber (or at least nitrile) gloves as DMSO penetrates the skin. For octocorals and black corals, quick snap freezing in liquid nitrogen immediately after collection is the best method to preserve DNA integrity. If this is not possible, then cold immersion in 95-100% EtOH and storage at -20°C is recommended.

DESS solution/DMSO/EDTA/NaCl preservation buffer

- 500 mL 0.5 M EDTA
- 200 mL Dimethyl Sulfoxide (DMSO)
- 300 mL milliQ water
- in a beaker or flask on a heating plate using a magnetic stir bar.
- Bit by bit and under constant stirring add 150 200 g NaCl until saturation (the salt does not dissolve any longer).
- Stir for at least 1 hour after pouring the salt. You can heat up the plate a little to increase solubility.
- Fill buffer into storage (PE) bottle and label.

Coral tissue preservation

 Immerse collected coral fragment(s) in the DESS buffer for storage. Short-term storage of samples can be maintained at room temperature. Long-term storage should be maintained at -20°C.

Protocol for flash-frozen sperm sampling

- Place sperm in 15 mL falcon tubes, about 5 mL per tube
- Centrifuge sperm suspension at 3,000 g for 10 minutes
- Wash the sperm pellet by resuspending it in 10 mL of TE buffer and centrifuging it again at 3000 *g* for 10 minutes
- Remove the supernatant and use a small spatula to transfer the pelleted sperm to cryo-vials.
- Snap freeze in liquid nitrogen and store at -80°C.

II. Symbiodiniaceae collection

Host-associated Symbiodiniaceae collections

This protocol has been developed for the isolation of Symbiodiniaceae cells from coral tissue.

- 1. Cut a "finger size" coral branch from the colony. Add the branch to a plastic bag.
- 2. Add 5 ml of filtered seawater (0.2 µm) and (air) blast tissue using an "air gun"
- 3. Pour the 5 ml of tissue slurry from the plastic bag into a 15 ml tube.
- 4. Top up with 5 ml of DMSO for a total of 10 ml slurry. Homogenize slurry.
- 5. Spin the slurry down in a fume hood for 20 min at 1000 2200 g at 4°C.
- 6. Pour off the top host fraction and any remaining liquid into DMSO waste.
- Add 5 ml of new DMSO to the tube that now contains the symbiont pellet. Multiple washes and centrifugations are recommended with DMSO to clean the pellet of any host material. This is the symbiont fraction. Snap freeze in liquid nitrogen or keep in DMSO at 4°C.

Free-living Symbiodiniaceae collections

This protocol is provided to account for the ability to obtain free-living forms of symbiotic microalgal species. Originally, the culturing protocol has been developed for obtaining cultures of free-living Symbiodiniaceae from epipsammic and endolithic habitats associated with reef sands and has been employed to establish >100 Symbiodiniaceae strains from Heron Reef, Great Barrier Reef, Australia (Nitschke et al., 2020).

- 1. Record exact latitude/longitude of collection site, date, sampling depth, reef zone (flat, lagoon, inner or outer crest etc).
- 2. Sample reef sands by scooping up surface sediments, or preferentially by using a sediment corer for sampling of a defined sediment layer. Note: Microbial phototrophs are typically concentrated close to the surface, at the first 1–2 cm (Werner et al., 2008).
- Sieve sediment cores on site or shortly after collection using stainless steel, woven-wire sieves to select sediment grains of 250–1000 μm (adjust size range as required) and wash out loosely attached organisms and detritus. Protect collected sediments from direct sunlight and desiccation.
- Transfer small amounts of prewashed sediments to sterile Petri dishes and add sterile-filtered (0.2 μm) and autoclaved seawater (ASW). Alternatively, break up sediments gently using a sterile mortar and pestle prior to transfer to Petri dishes.
- 5. Seal Petri dishes with parafilm to limit evaporation and incubate at 26°C and 130–150 µmol photons m⁻²s⁻¹ under a 12:12 h light: dark cycle over several days. Survey daily on an inverted microscope for Symbiodiniaceae-like cells based on size, shape, coloration and swimming behavior. Note: Broken-up sediment from Heron Reef consistently yielded larger numbers of Symbiodiniaceae-like cells than intact sediment (Nitschke et al., 2020).
- 6. Target candidate cells for isolation using glass Pasteur pipettes, thinned out into capillaries over a gas flame. Transfer picked single cells into a droplet of ASW on a glass microscope slide. Re-pick the cell into a second droplet of ASW, microscopically confirm that only a single cell has been picked. For single-cell culturing, transfer the droplet into a 96-well plate (Sarstedt, Nürnbrecht, Germany), filled with 100 μL of the following growth media: a 1:1:1

combination of 0.22 µm-filtered seawater from the Petri dish, fresh f/2 medium (Guillard, 1975), and pre-conditioned f/2 medium. Obtain the latter by sterile-filtering growth medium from an established mid to late exponential Symbiodiniaceae culture.

- 7. Confirm successful isolation of single cells by visually examining the wells on an inverted microscope. Seal plates with parafilm to limit evaporation and incubate at 26°C and 130–150 µmol photons m⁻²s⁻¹ under a 12:12 h light: dark cycle. Continue surveying the wells twice weekly and record the establishment of cultures and the presence of any contaminants.
- 8. When cultures reach several hundred cells, add another 100 μL of f/2 medium and then gradually increase culture volume to 2 mL on 24-well plates for suspended cell cultures and then 10 mL in glass reaction tubes with aluminum caps. At this stage cultures are typically stable and can be identified genetically and morphologically.

III. Prokaryote collection

Coral host colony photos

Please refer to I. Coral collections above, same guidelines/recommendations apply.

Metadata

Collect all the biophysical parameters of the collection site possible. The minimum information required includes geographical coordinates, water temperature, depth, and salinity. Additional information includes light environment, current measurements, light intensity, DOC concentration.

Preservation

Please refer to I. Coral collections above, same guidelines/recommendations apply.

Coral sampling for molecular analyses and bacterial isolation

- For all procedures, wear gloves and use clean pliers or chisel for sampling and handling the coral fragments. Optimal is sterilized pliers/chisel, recommended is to use separate pliers/chisel, minimal recommendation is to at least wash tools in seawater vigorously between each coral fragment collected. Fragments measuring 5 cm up in diameter are sufficient. Avoid damaging the coral colony or fragment, as the damage could affect the colony and/or the associated microbes.
- 2. Place the coral fragment in a sterile conical tube or bag (e.g., ZipLoc). It is important to avoid contact between fragments by using separate collection containers for each fragment; minimize exposure to surrounding water along the dive.
- 3. Once reaching the surface, remove the water from the conical tube or bag and immediately process the samples (either by flash-freezing or extracting genetic material). For voucher preparation and deposit please refer to section I. Coral collections above.
- 4. If genetic material is not immediately extracted or if unable to flash freeze, samples used for DNA analysis or bacterial isolation should be kept on ice until processing. Samples used for other analyses should be stored in the appropriate buffer until processing (e.g., RNAlater for samples targeted for RNA analysis). Samples that are flash-frozen as a means of storage, cannot be used for cultivation. For cultivation, fresh tissue from samples stored in the cold and dark is preferable.
- 5. For sample processing, homogenize the coral samples via maceration with a mortar and pestle or airblasting with an airbrush and a sterile seawater solution followed by centrifugation to isolate the tissue from the airblasted solution. Samples should be labeled with the same unique identifier used for the field photo of the coral (see above), so molecular data can be traced back to the source coral.

Sampling coral mucus for molecular analyses

As above, for all procedures, use gloves and sterile collection tools and containers. Mucus samples can be obtained using sterile syringes underwater and stored in sterile conical tubes using the same unique identifier used for the field photo of the coral (see above). An alternative way to collect coral mucus is exposing the whole colony to air for 10 minutes. Mucus will be released and collected with a conical tube as it drops (invert the coral fragment to facilitate this).

Sampling the surrounding seawater for molecular analyses

As above, for all procedures, use gloves and sterile collection tools and containers. To obtain water for molecular analysis that is potentially relevant to coral physiology, collect water close to the target colony but not too close to avoid sampling the 'halo' of the coral colony (~1 m distance to the coral colony). For molecular analyses, filter at least 1 L of water through a 0.22 µm PVDF membrane filter (e.g., Sterivex filter unit). Filter the same volume for each of the samples. After filtering, label the filter with the same unique identifier used for the field photo of the coral (see above) and store the filter at -20°C for DNA extraction.

Collection/Processing Technique	Pros	Cons	Selected References
Coral fragment maceration	- Microbes can be traced to the coral host	- The coral skeleton cannot be separated from the tissue	(Röthig et al., 2017; Rosado et al., 2019; Villela et al., 2019)
Coral tissue airblast	 Obtain coral tissue without coral skeleton Microbes can be traced to the coral tissue 	- It does not allow culturing microbes associated with the coral skeleton	(Glasl et al., 2016; Neave et al., 2017; Pogoreutz et al., 2017; Robbins et al., 2019; Rädecker et al., 2021)
Mucus	- The coral mucus can be used to answer specific questions about the boundary between the coral and the surrounding seawater	- Mucus layers are contaminated and diluted by surrounding seawater	(Leite et al., 2018; Glasl et al., 2019; Hadaidi et al., 2019; Osman et al., 2020)
Surrounding seawater	 Easy to sample and process Can provide information on reef microbial communities 	- Not easily traced to specific coral hosts	(Neave et al., 2017; Glasl et al., 2019; Osman et al., 2020)

Supplementary Table S4. Common sample collection and processing techniques for molecular analysis of coral-associated microbes.

DNA Extraction and Sequencing

- 1. Commercially available soil, plant, and blood tissue kits are suitable for many molecular extraction procedures resulting in metagenomes, amplicon sequences, or transcriptomes. Many kits may also be used to extract genetic material from the frozen filters used for sampling the surrounding seawater (see above). Manual extractions (e.g., phenol-chloroform) are suitable for extracting high molecular weight DNA. Irrespective of the kit/method used, it is essential to have a negative extraction control that will be processed alongside the actual samples to monitor the contamination caused by the extraction, a clean-up step using a commercial clean up kit, might be necessary to remove inhibitors (particularly in black corals, some deep-water stony corals, and octocorals) that affect downstream sequencing library preparations.
- 2. Different sequencing protocols and/or kits can be used to either target specific genes (in most cases the 16S rDNA gene) or the whole genetic material from the microbial fraction of the coral community (e.g., for metagenomic sequencing). For many procedures, PCR amplification may be necessary to ensure sufficient material for generation of sequencing libraries. A null template DNA PCR control should be run alongside the actual samples to monitor the contamination caused by PCR reagents or procedures. A positive control can be included to monitor successful amplification and sequencing procedures.

Side note: A common concern is that DNA isolation procedures have inherent biases. Since typically the interest lies in comparing differences between samples (that are all subject to the same methods and inherent biases), this should be a minor issue of concern and consistency/standardization is more important than choice of extraction (Han et al., 2019). Further to the alleviation of such concerns, the targeted 16S region and the analysis pipeline seem to exert a bigger influence than the DNA extraction method (Rintala et al., 2017).

Bacterial Isolation and Storage

- 1. For isolation of bacteria associated with coral tissue, serial dilute coral samples (macerate, mucus, or airblasted tissue) down to 10⁻⁹ and water samples down to 10⁻⁶ in sterile saline solution.
- Apply 100 µL of each of the dilutions to an agar plate containing the desired medium; a range of bacterial media is available (Röthig et al., 2016; Pogoreutz and Voolstra, 2018; Villela et al., 2019; Sweet et al., 2020). Perform this step in triplicate for each dilution. Incubate the agar plates at the desired temperature for 1-3 days, checking the plates every day.
- 3. To increase diversity of cultured colonies, aim to select distinct colonies from the growth plate and replate on new plates using the streak plate technique (notably, many bacterial species exhibit rather uniform color, border, shape). Repeat as many times as necessary to obtain pure colonies growing on the plates. Pure colonies can then be regrown at the desired temperature or stored for later use.
- 4. To store the bacteria isolates, after obtaining pure cultures, either store the agar plate at 4°C (for short-term storage) or prepare a glycerol stock by homogenizing a pure colony with a sterile medium and adding glycerol (20% v/v); store glycerol stocks at -80°C.

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