Coral Reefs
https://doi.org/10.1007/s00338-025-02756-0

BRIEF REPORT

Spawning times and breeding trials suggest no inbreeding occurs between *Porites* aff. *lutea* and *P.* cf *cylindrica*

Tullia I. Terraneo[1,3] · Alyson Kuba[3] · Michael L. Berumen[1] · Saki Harii[2] · Andrew H. Baird[3]

Received: 19 May 2025 / Accepted: 25 September 2025

Abstract Molecular approaches have advanced our understanding of reef coral biodiversity. However, in some taxa, they have yet to deliver a robust taxonomy that reflects differences in the ecology and biology of species. For example, *Porites* cf. *cylindrica* and *P.* aff. *lutea*, which have different morphologies and life histories, are clustered close together in a complex containing several other *Porites* species when analyzed using both single locus molecular markers and thousands of SNPs from ezRAD data. This suggests the possibility of recent or ongoing speciation and/or hybridization between these species. Here, we used the timing of gamete release and gamete compatibility to test reproductive boundaries between the two. At Orpheus Island (Australia), *P.* cf. *cylindrica* and *P.* aff. *lutea* spawned on the same night, while at Sesoko Island (Japan), they did not. Furthermore, when spawning occurred during in vitro breeding trials, the eggs did not cleave, suggesting that *P.* cf. *cylindrica* and *P.* aff. *lutea* are not interbreeding. Breeding trials can help resolve species identities in taxa for which molecular and morphological data provide unclear results.

Supplementary Information The online version contains supplementary material available at https://doi.org/10.1007/s00338-025-02756-0.

✉ Tullia I. Terraneo
tulliaisotta.terraneo@kaust.edu.sa

[1] Red Sea Research Center, Division of Biological and Environmental Science and Engineering, King Abdullah University of Science and Technology, Thuwal, Saudi Arabia

[2] Sesoko Marine Research Station, Tropical Biosphere Research Center, University of the Ryukyus, 3422 Sesoko, Motobu, Okinawa 905-0227, Japan

[3] College of Science and Engineering, James Cook University, Townsville, QLD, Australia

Keywords Corals · Species boundaries · Barriers to gene flow · Species complexes

Introduction

Molecular methods have revolutionized our understanding of the systematics and taxonomy of scleractinian corals (Romano and Palumbi 1996; Kitahara et al. 2016). Nuclear and ribosomal markers proved highly effective at resolving the evolutionary relationships within many taxa, such as among genera in the families Faviidae and Mussidae (Huang et al. 2011, 2016) and among species in the genus *Pocillopora* (Schmidt-Roach et al. 2014). More recently, next-generation sequencing genomic approaches have provided sufficient resolution to resolve relationships among species in genera that had proved intractable to single marker studies, such as the *Acropora* (Bridge et al. 2024). However, in other species-rich genera, complex evolutionary scenarios are hard to disentangle (Kitahara et al. 2016), and interspecific hybridization is often called into question (e.g., Veron 1995). Using a reduced representation genomic approach (ezRAD), an Indo-Pacific wide study of the genus *Porites* recovered 15 distinct molecular lineages (Terraneo et al. 2025), a small fraction of the nominal species currently listed at the World Register of Marine Species (WoRMS) (Hoeksema and Cairns 2024). These included two lineages (clade V and clade XV sensu Terraneo et al. 2025) that comprise numerous closely related nominal species with vastly different morphologies and life histories. In particular, clade V sensu (Terraneo et al. (2025)) clusters close together common, widespread, and ecologically significant species, such as *P.* cf. *cylindrica, P.* aff. *lutea*, and *P.* cf. *lobata*. This suggests complex evolutionary dynamics, including, for example, recent speciation and incomplete lineage sorting, such

Published online: 03 October 2025

as in species with large effective population sizes. Alternatively, these results might suggest hybridization and introgression, encompassing inbreeding between nominal species. Further research is needed to refine our understanding of species boundaries within this clade.

Conflicts among different lines of evidence to determine species boundaries are not unusual in taxonomy. Indeed, there are dozens of species definitions in the literature (Willis 1990). One influential definition was the Biological Species Concept of Mayr (1963). The definition of species as interbreeding units seems intuitive; however, it cannot help resolve fossil lineages, and it is often impossible to cross individuals to test for breeding compatibility if they occur in geographically disparate locations. De Queiroz (1998) attempted to resolve competing species definitions. He defined a species as an independent evolving metapopulation lineage and suggested that the plethora of species concepts outlined by evolutionary biologists should be regarded as "operational criteria" to provide different lines of evidence to distinguish these lineages. This suggests that multiple lines of evidence are required to produce a robust taxonomy. A recent study, for example, showed how integrating different lines of evidence encompassing coral colony morphology, genomics, and reproductive biology can shed light on species boundaries, also in a challenging genus like *Acropora* (Ramírez-Portilla et al. 2022).

Here, we use reproductive biology as an alternative line of evidence to help clarify complex genomic patterns retrieved for *P.* cf. *cylindrica* and *P.* aff. *lutea* and determine the presence of mating compatibility and breeding barriers by attempting to cross these species at Orpheus Island (Palm Islands–QLD, Australia) and Sesoko Island–(Okinawa, Japan).

Material and methods

Sample collection and morphological identification

A total of 72 fragments of colonies of *P.* cf. *cylindrica* and *P.* aff. *lutea* were collected over three spawning seasons. In the first experiment, 23 fragments (13 *P.* cf. *cylindrica* and 10 *P.* aff. *lutea*) were collected in Pioneer Bay (-18º36′64''N; 146º29′28''E) (Palm Islands, Orpheus Island—QLD, Australia) between one and four days after the full moon in November 2017 (Babcock et al. 1986; Baird et al. 2021). In the second experiment, 28 fragments (14 *P.* cf. *cylindrica* and 14 *P.* aff. *lutea*) were collected in front of Sesoko Island (26º38'0.00''N; 127º51′56.24''E) (Okinawa, Japan) between one and five days after the full moon in May 2018 (Hirose and Hidaka 2006). Finally, in the third experiment, 21 fragments (11 *P.* cf. *cylindrica* and 10 *P.* aff. *lutea*) were collected in Pioneer Bay (Palm Islands, Orpheus Island—QLD, Australia) between one and four days after the full moon in November 2018 (ESM. 1). Fragments > 20cm^2 were collected using a hammer and chisel from colonies that were tagged and then photographed using a Canon G15 camera. Fragments were placed into aquaria at either Orpheus Island Research Station (OIRS) from James Cook University (JCU, Australia) or Sesoko Marine Research Station, University of the Ryukyus (Japan), with running sea water until spawning. Specimens were identified following comparison to type material and original descriptions.

Spawning observations, gamete collection, and ex situ *breeding trials.*

Porites species are generally gonochoric (Baird et al. 2009), meaning each colony is either male or female, although at Sesoko Island *P.* cf. *cylindrica* also showed gonochoric females and male-dominated hermaphrodites (Inoha et al. 2024). Determining the sex of each colony of *Porites* before spawning is not possible without destructive sampling. Therefore, in order to separate gametes of different sexes at spawning, fragments were isolated in individual 20 L buckets 00:30 h prior to sunset. Fragments were observed every 00:10 h from 18:00 to 1:00—the following morning, for spawning. Following spawning, the sex of each colony and the time of gamete release was recorded. The gametes were collected using Pasteur pipettes and brought to a temperature-controlled room for the breeding trials. In particular, sperm was collected directly from colonies using large pipettes during release, and concentrations were monitored visually to produce concentrations of approximately 10^6 sperm/mL (Hagedorn et al. 2009). Around 5000 eggs were mixed with sperm from the two different species in separate containers and allowed to sit for 00:30 h. Notably, no eggs were observed in the *P.* cf. *cylindrica* sperm used at Sesoko Island. After this time, three replicates of around 50 eggs for each treatment (*i.e.,* around 150 eggs for each intraspecific mix) were examined under a dissecting microscope for cleavage, which would indicate successful fertilization. Similarly, eggs and sperm from the same nominal species were crossed and left undisturbed for 00:30 h to allow fertilization and were used to test for viability of the gametes. For each cross, a control of clean eggs was also set up in order to check for sperm contamination or self-fertilization. Observations were repeated every 00:30 h for 2:00 h in triplicates. If cleavage was observed, the crosses were checked until a fertilization success of 80% was reached.

Results and discussion

From the observation and breeding trials, reproductive isolation between *P.* aff. *lutea* and *P.* cf. *cylindrica* was suggested in all three sets of experiments, either due to differences in spawning times between the two species, or

incompatibility of the gametes. The apparent reproductive isolation, along with distinctive morphologies and life histories, might indicate that both nominal species could be evolving independently.

Spawning times

Experiment 1: November 2017 Orpheus Island.

In 2017 at Orpheus Island, the release of eggs and sperm differed by between 2:00 and 2:30 h, with *P.* cf. *cylindrica* always spawning earlier in the evening. In particular, in November 2017, of the 23 colonies collected, eight colonies of *Porites* cf. *cylindrica* and three colonies of *P.* aff. *lutea* spawned between November 7 and November 9. Of the spawning *P.* cf. *cylindrica* colonies, 62% (n = 8) were females, as were 67% (n = 3) of the spawning *P.* aff. *lutea* colonies. Sperm release in *P.* cf. *cylindrica* started between 20:30 and 20:40; egg release started between 21:00 and 22:00. Egg release in the two *P.* aff. *lutea* colonies occurred between 21:50 and 22:30, and the one male colony released sperm at 23:00 (Fig. 1, ESM. 1).

Experiment 2: June 2018 Sesoko Island.

In June 2018, of the 28 colonies collected, three *P.* cf. *cylindrica* and four *P.* aff. *lutea* spawned ex situ at Sesoko Island. All three *P.* cf. *cylindrica* colonies (100%) that spawned were males, compared to two of four (50%) of the spawning colonies of *P.* aff. *lutea*. The time of gamete release differed between the two species by two days. All *P.* cf. *cylindrica* colonies spawned on June 3, and all *P.* aff. *lutea* colonies spawned on June 6. Sperm release for *P.* cf. *cylindrica* started at 21:30, while for *P.* aff. *lutea*, eggs were released between 21:45 and 22:30, and sperm release started at 22:30 (Fig. 1, ESM. 1).

Experiment 3: November 2018 Orpheus Island.

In 2018 at Orpheus Island, *P.* aff. *lutea* and *P.* cf. *cylindrica* did not spawn at the same time (Fig. 1, ESM. 1). In November 2018, of the 21 colonies collected, eight spawned ex situ: three *P.* cf. *cylindrica* and five *P.* aff. *lutea*. Two colonies of *P.* aff. *lutea* spawned two nights in a row. Two colonies of *P.* cf. *cylindrica* released sperm on November 25 and November 26; one colony spawned eggs on November 26. Two colonies of *P.* aff. *lutea* spawned sperm on November 27, while one colony released eggs. Finally, four colonies released sperm on November 28. Both sperm and eggs release in *P.* cf. *cylindrica* started at 20:30, while *P.* aff. *lutea* spawned sperm always at 22:00, and eggs at 22:30 (Fig. 1, ESM. 1).

Differences in spawning times and gamete incompatibility are the main barriers to gene flow among sympatric populations (Knowlton 1993; Knowlton et al. 1997; Dai et al. 2000) and species with as little as 1:30 h difference in spawning times have proven to be genetically divergent (e.g., Knowlton et al. 1997; Fukami et al. 2003). Indeed, differences on the scale of hours are sufficient to maintain species boundaries in *Acropora* and *Orbicella* in relation to the diffusion and dilution of sperm in the ocean (van Oppen et al. 2002; Fukami et al. 2003; Levitan et al. 2011; Wei et al. 2012). In *Acropora*, field-based observations showed a sudden drop in sperm concentration as early as 0:30 h after spawning, decreasing the chances for gametes' encounter and thus hybridization (Fukami et al. 2003). Estimates of fertilization potential for *Orbicella* showed that sperm has maximum fertilization potential for about 1:00 h, after which

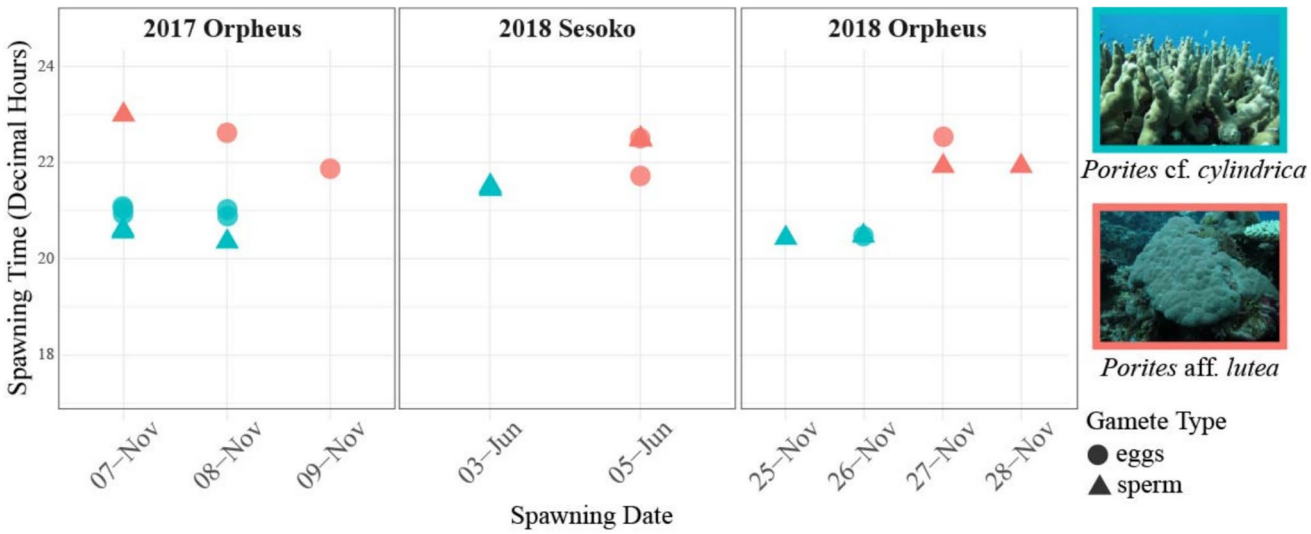

Fig. 1 Ex situ spawning day and time at Orpheus Island (Great Barrier Reef, Australia) and Sesoko Island (Okinawa, Japan) for the years 2017 and 2018. For each colony, the nominal species is highlighted with different colors, and the released gametes (eggs or sperm) with different symbols

dilution and aging decrease the chances of breeding (Levitan et al. 2004). From the data collected in our work and previous spawning observations on *Porites* at the same location (Willis et al. 1985), 1:30 h seems to be the time threshold after which fertilization potential decreases in *Porites*.

Breeding trials

Results are summarized in Table 1. At Orpheus Island in 2017, from the eight colonies of *P.* cf. *cylindrica* that spawned, we were able to successfully cross 50% of the within-species trials (four successful fertilization out of eight trials). The egg controls did not fertilize, indicating that there was no contamination from other sperm. In particular, on November 7, eggs and sperm of *P.* cf. *cylindrica* successfully crossed at 23:20 (TAUSP3 x TAUSP5) and 23:39 (TAUSP1 x TAUSP5), 1:30 h after mixing—84.14% fertilization (n of total eggs = 150), and 73.75% (n of total eggs = 140) fertilization. Finally, 4.6% of the eggs were fertilized at 23:30 (TAUSP3 x TAUSP2—n of total eggs = 152). No other fertilization occurred between *P.* cf. *cylindrica* gametes on that night. On November 8, eggs and

Table 1 Results from the *ex situ* fertilization trials within and between *Porites* aff. *lutea* and *Porites* cf. *cylindrica* at Orpheus Island (Queensland, Australia) in 2017 (**a**) and 2018 (**b**) (Experiment 1 and 2), and at Sesoko Islands in 2018 (**c**) (Okinawa, Japan) (Experiment 3). Proportions of fertilization are shown. DAFM = days after full moon

(a) EXPERIMENT 1				Eggs					
Year	Site	DAFM	Sperm	*P.* cf *cylindrica* TAUSP3	*P.* cf. *cylindrica* TAUSP4	*P.* cf. *cylindrica* TAUSP1	*P.* cf. *cylindrica* TAUSP19	*P.* cf. *cylindrica* TAUSP21	*P.* aff *lutea* TAUSP11
2017	Orpheus Island	3	*P.* cf. *cylindrica* TAUSP2	4.6% (7/152)	0 (0/150)	0 (0/150)	–	–	–
			P. cf. *cylindrica* TAUSP5	84.14% (138/164)	0 (0/150)	73.57% (103/140)	–	–	–
			P. aff. *lutea* TAUSP8	0.66% (1/150)	0 (0/150)	–	–	–	–
		4	*P.* cf. *cylindrica* TAUSP6	–	–	–	0.64% (1/154)	87.2% (75/86)	0 (0/150)
			Egg control	0 (0/156)	0 (0/150)	0 (0/150)	0 (0/150)	0 (0/82)	0 (0/150)

(b) EXPERIMENT 2				Eggs	
Year	Site	DAFM	Sperm	*P.* aff *lutea* TJP8	*P.* aff *lutea* TJP28
2018	Sesoko Island	6	*P lutea* TJP10	80% (120/150)	0 (0/150)
			P. aff *lutea* TJP27	74.66% (112/150)	0 (0/150)
			Egg control	0 (0/156)	0 (0/150)

(c) EXPERIMENT 3				Eggs
Year	Site	DAFM	Sperm	*P. aff lutea* T11
2018	Orpheus Island	3	*P.* aff *lutea* T13	73.85% (113/153)
			P. aff *lutea* T19	64.61% (84/130)
			Egg control	0 (0/150)

sperm of *P.* cf. *cylindrica* TAUSP21 x TAUSP6 crossed at 22:56—87.2% fertilization (n of total eggs = 86), 1:40 h after mixing. Only one egg was fertilized at 22:56 (TAUSP19 x TAUSP6)—0.64%; thus, this was not considered significant. The eggs of one colony (TAUSP4) were never fertilized during the trials, so we cannot exclude infertility for this colony. No interspecific crosses occurred during the non-choice breeding trials experiments between *P.* cf. *cylindrica* and *P.* aff. *lutea*. Similar results are common in closely related *Acropora* species that spawn in synchrony in Taiwan (Wei et al. 2012). Nevertheless, we could not confirm the fertility of *P.* aff. *lutea* colonies using within-species crosses given the lack of synchronous spawning of male and female *P*. aff. *lutea* colonies in 2017.

In Okinawa in June 2018, fertilization was observed within 50% (n = 4) of *P.* aff. *lutea* within species crosses on June 5. Fertilization occurred at 00:30, 1:30 h after gametes mixing—TJP8 x TJP10, 80% fertilization (n of total eggs = 150), and 74.55% TJP8 x TJP27 (n of total eggs = 150 eggs). The eggs of one colony (TJP28) were never fertilized; as such, we cannot confirm the fertility of this colony. No colonies of *P.* cf. *cylindrica* spawned during the same night as *P.* aff. *lutea;* thus, no breeding trial was conducted between the two (Table 1b).

At Orpheus in November 2018, fertilization was observed in 100% (n = 2) of *P.* aff. *lutea* within species crosses—November 26. The egg controls showed no contamination from foreign sperm for all the mixes. Fertilization occurred at 00:15 (T11 x T19) and 00:30 (T11 x T13)—64.61% (*n* of total eggs = 130) and 73.85% fertilization (n of total eggs = 153), 1:40 h after mixing. No colonies of P. cf. *cylindrica* spawned on the same night as *P.* aff. *lutea* in 2018; thus, no species crosses were conducted (Table 1c).

Our results should be carefully interpreted as, overall, only a small proportion of colonies spawned ex situ, particularly in Okinawa, where more than half of the colonies of both *P.* cf. *cylindrica* and *P.* aff. *lutea* failed to release gametes. Recent work focusing on ex situ spawning trials of *Porites* in Palau has similarly documented spawning proportions of just 14–35% in monitored colonies and only 18% in histological sections over different seasons (Bennett et al. 2024). Similarly, Glynn et al. (1994) found relatively low proportions (30–67%) of gametes in *Porites* in the eastern Pacific, suggesting reproductive output in *Porites* populations could be naturally low in any given year. For our study population in Okinawa, it is also highly likely that colonies were sampled before the peak spawning month. In fact, spawning of *Porites* in Okinawa occurs predominantly around the 7th full moon after the winter solstice (Hirose & Hidaka 2006, Baird et al. 2022), whereas our experiments at Sesoko occurred around the 5th full moon. Nevertheless, there is no reason to suggest that gametes will be less fertile earlier in the reproductive season. In addition, coral reproductive output is sensitive to environmental factors (Baird et al. 2009), and prolonged and elevated sea water temperatures at Sesoko in 2017 (Singh et al. 2022) might have affected the reproductive output of the population. These factors, alone or in combination, could account for the reduced spawning observed and might have influenced the outcome of both intra- and interspecific fertilization trials.

Conclusions

Together with genomic and ecological data, controlled breeding experiments can offer insights into reproductive barriers, helping to distinguish between closely related species. Our findings suggest that *P.* cf. *cylindrica* and *P.* aff. *lutea* could be reproductively isolated species in the study locations, maintained by differences in spawning times and gamete incompatibility. The observed time gaps in spawning, ranging from hours to days, could significantly reduce the potential for hybridization, reinforcing their status as independently evolving species. These findings align with previous research on coral reproductive isolation and suggest that temporal separation in spawning is a key mechanism maintaining species boundaries in *Porites*. Additionally, the apparent failure of interspecific crosses in our breeding trials further supports the presence of intrinsic reproductive barriers, although more trials and observations would be needed to provide a definitive conclusion. Building on these results, future studies should prioritize reproductive trials as a crucial tool for refining coral taxonomy and clarifying intricate evolutionary patterns. Integrating reproductive biology with genomic approaches will be essential for resolving taxonomic uncertainties, particularly in species-rich and morphologically variable coral genera. Expanding such studies across diverse geographic regions and environmental conditions will further enhance our understanding of coral speciation, hybridization potential, and the mechanisms maintaining species boundaries.

Conflict of interest

The authors declare no competing interests.

Acknowledgements This project was founded by the Higher Degree Research Enhancement (HDRES) Award 2017 to Tullia I Terraneo (ARC Centre of Excellence for Coral Reef Studies, James Cook University), the David Yellowlees Excellence in Research Award 2018 to Tullia I Terraneo (ARC Centre of Excellence for Coral Reef Studies, James Cook University), the 2018 Red Sea Research Center (RSRC) Travel Grant to Tullia I Terraneo (King Abdullah University of Science and Technology), the Australia Research Council (ARC) funds to Andrew H Baird, and King Abdullah University of Science and Technology (KAUST) baseline funds to Michael L Berumen. This research was undertaken in accordance with the policies and procedures of King

Abdullah University of Science and Technology (KAUST). We thank Catalina Ramírez-Portilla for assistance during fieldwork in Okinawa.

Author contributions Tullia I Terraneo performed conceptualization, sampling, methodology, data curation, formal analysis, investigation, visualization, and writing—original draft. Alyson Kuba performed sampling and methodology. Michael L Berumen performed funding acquisition, resources, supervision, and writing—review and editing. Saki Harii performed resources, supervision, and writing—review and editing. Andrew H Baird performed conceptualization, sampling, methodology, funding acquisition, resources, supervision, and writing—review and editing.

Funding Open access publishing provided by King Abdullah University of Science and Technology (KAUST). This project was founded by the Higher Degree Research Enhancement (HDRES) Award 2017 to Tullia I. Terraneo (ARC Centre of Excellence for Coral Reef Studies, James Cook University), the David Yellowlees Excellence in Research Award 2018 to Tullia I. Terraneo (ARC Centre of Excellence for Coral Reef Studies, James Cook University), the 2018 Red Sea Research Center (RSRC) Travel Grant to Tullia I. Terraneo (King Abdullah University of Science and Technology), the Australia Research Council (ARC) funds to Andrew H. Baird and King Abdullah University of Science and Technology (KAUST) baseline funds to Michael L. Berumen. This research was undertaken in accordance with the policies and procedures of King Abdullah University of Science and Technology (KAUST).

Data availability Data are provided within the manuscript or supplementary information files.

References

Babcock RC, Bull GD, Harrison PL, Heyward AJ, Oliver JK, Wallace CC, Willis BL (1986) Synchronous spawnings of 105 scleractinian coral species on the Great Barrier Reef. Mar Biol 90:379–394. https://doi.org/10.1007/BF00428562

Baird AH, Guest JR, Willis BL (2009) Systematic and biogeographical patterns in the reproductive biology of scleractinian corals. Annu Rev Ecol Evol Syst 40(1):551–571. https://doi.org/10.1146/annurev.ecolsys.110308.120220

Baird AH, Guest JR, Edwards AJ, Bauman AG, Bouwmeester J, Mera H, Yusuf S (2021) An Indo-Pacific coral spawning database. Sci. Data 81:35. https://doi.org/10.6084/m9.figshare.13100552

Baird AH, Edwards AJ, Guest JR, Harii S, Hatta M, Lachs L, Mera H, Sinniger F, Abrego D, Ben-Zvi Or, Bronstein O, Cabaitan PC, Cumbo VR, Eyal G, Eyal-Shaham L, Feldman B, Figueiredo J, Flot J-F, Grinblat M, Heyward A, Hidaka M, Hirose M, Iguchi A, Isomura N, Kinzie RA, Kitanobo S, Kuba A, Levy O, Loya Y, Mezaki T, Mohamed AR, Morita M, Nojima S, Nozawa Y, Prasetia R, Puill-Stephan E, Ramirez-Portilla C, Rapuano H, Rosenberg Y, Sakai Y, Sakai K, Shlesinger T, Terraneo TI, Yakovleva I, Yamamoto HH, Yamazato K (2022) A coral spawning calendar for Sesoko Station, Okinawa, Japan. Galaxea J Coral Reef Stud 24(1):41–49. https://doi.org/10.3755/galaxea.G2021_S10O

Bennett MJ, Grupstra CG, Da-Anoy J, Andres M, Holstein D, Rossin A, Meyer-Kaiser KS (2024) Ex situ spawning, larval development, and settlement in massive reef-building corals (Porites) in Palau. Invertebr Biol 143(4):e12447. https://doi.org/10.1111/ivb.12447

Bridge TC, Cowman PF, Quattrini AM, Bonito VE, Sinniger F, Harii S, Baird AH (2024) A tenuis relationship: traditional taxonomy obscures systematics and biogeography of the '*Acropora tenuis*' (Scleractinia: Acroporidae) species complex. Zool J Linn Soc 202(1):zlad062. https://doi.org/10.1093/zoolinnean/zlad062

Dai CF, Fan TY, Yu JK (2000) Reproductive isolation and genetic differentiation of a scleractinian coral *Mycedium elephantotus*. Mar Ecol Prog Ser 201:179–187. https://doi.org/10.3354/meps201179

Fukami H, Omori M, Shimoike K, Hayashibara T, Hatta M (2003) Ecological and genetic aspects of reproductive isolation by different spawning times in *Acropora* corals. Mar Biol 142:679–684. https://doi.org/10.1007/s00227-002-1001-8

Glynn PW, Colley SB, Eakin CM, Smith DB, Cortés J, Gassman NJ, Guzmán HM, Del Rosario JB, Feingold JS (1994) Reef coral reproduction in the eastern Pacific: Costa Rica, Panamá, and Galápagos Islands (Ecuador). II. Poritidae. Mar Biol 118(2):191–208

Hagedorn M, Carter V L, Hollingsworth L, Leong J C, Kanno R, Borneman E H & Schick M (2009). *Ex Situ* culture of Caribbean and Pacific coral larvae comparing various flow-through chambers. Smithson Contrib Mar Sci, 38

Hirose M, Hidaka M (2006) Early development of zooxanthella-containing eggs of the corals *Porites cylindrica* and *Montipora digitata*: the endodermal localization of zooxanthellae. Zool Sci 23(10):873–881. https://doi.org/10.2108/zsj.23.873

Hoeksema B.W, Cairns S D 2024. World List of Scleractinia. Accessed through: World Register of Marine Species at: http://www.marinespecies.org/ accessed on 2024–05–14

Huang D, Licuanan WY, Baird AH, Fukami H (2011) Cleaning up the "Bigmessidae": Molecular phylogeny of scleractinian corals from Faviidae, Merulinidae. Pectiniidae and Trachyphylliidae BMC Evol Biol 11:1–13. https://doi.org/10.1186/1471-2148-11-37

Huang D, Arrigoni R, Benzoni F, Fukami H, Knowlton N, Smith ND, Budd AF (2016) Taxonomic classification of the reef coral family Lobophylliidae (Cnidaria: Anthozoa: Scleractinia). Zool J Linn Soc 178(3):436–481. https://doi.org/10.1111/zoj.12391

Inoha K, Isomura N, Morita M, Kurihara H (2024) Combination of hermaphroditic and gonochoric sexual modes in the coral *Porites cylindrica*. Coral Reefs 43(6):1831–1841. https://doi.org/10.1007/s00338-024-02580-y

Kitahara M V, Fukami H, Benzoni F & Huang D (2016). The New Systematics of Scleractinia: Integrating Molecular and Morphological Evidence. In: Goffredo, S., Dubinsky, Z. (eds) The Cnidaria, Past, Present and Future. Springer, Cham. https://doi.org/10.1007/978-3-319-31305-4_4

Knowlton N (1993) Sibling species in the sea. Annu Rev Ecol Evol Syst. https://doi.org/10.1146/annurev.es.24.110193.001201

Knowlton N, Mate JL, Guzman HM, Rowan R, Jara J (1997) Direct evidence for reproductive isolation among the three species of the *Montastraea annularis* complex in Central America (Panama and Honduras). Mar Biol 127:705–711. https://doi.org/10.1007/s002270050061

Levitan DR, Fukami H, Jara J, Kline D, McGovern TM, McGhee KE, Knowlton N (2004) Mechanisms of reproductive isolation among sympatric broadcast-spawning corals of the *Montastraea annularis* species complex. Evolution 58(2):308–323. https://doi.org/10.1111/j.0014-3820.2004.tb01647.x

Levitan DR, Fogarty ND, Jara J, Lotterhos KE, Knowlton N (2011) Genetic, spatial, and temporal components of precise spawning synchrony in reef building corals of *the Montastraea annularis* species complex. Evolution 65(5):1254–1270

Mayr E (1963) Animal Species and Evolution. Harvard University Press

De Queiroz K (1998). The general lineage concept of species, species criteria, and the process of speciation. Endless Forms: Species and Speciation

Ramírez-Portilla C, Baird AH, Cowman PF, Quattrini AM, Harii S, Sinniger F, Flot JF (2022) Solving the coral species delimitation conundrum. Syst Biol 71(2):461–475. https://doi.org/10.1093/sysbio/syab077

Romano SL, Palumbi SR (1996) Evolution of scleractinian corals inferred from molecular systematics. Science 271(5249):640–642. https://doi.org/10.1126/science.271.5249.640

Schmidt-Roach S, Miller KJ, Lundgren P, Andreakis N (2014) With eyes wide open: a revision of species within and closely related to the *Pocillopora damicornis* species complex (Scleractinia; Pocilloporidae) using morphology and genetics. Zool J Linn Soc 170(1):1–33. https://doi.org/10.1111/zoj12092

Singh T, Sinniger F, Nakano Y, Nakamura S, Kadena S, Jinza M, Fujimura H, Harii S (2022) Long-term trends and seasonal variations in environmental conditions in Sesoko Island, Okinawa, Japan. Galaxea J Coral Reef Stud 24(1):121–133

Terraneo TI, Benzoni F, Arrigoni R, Berumen ML, Mariappan KG, Antony CP, Harrison HB, Payri C, Huang D, Baird AH (2025) A genomic approach to *Porites* (Anthozoa: Scleractinia) megadiversity from the Indo-Pacific. Mol Phylogenet Evol 203:108238. https://doi.org/10.1016/j.ympev.2024.108238

Van Oppen MJ, Willis BL, Van Rheede T, Miller DJ (2002) Spawning times, reproductive compatibilities and genetic structuring in the *Acropora aspera* group: evidence for natural hybridization and semi-permeable species boundaries in corals. Mol Ecol 11(8):1363–1376. https://doi.org/10.1046/j.1365-294X.2002.01527.x

Veron JEN (1995) Corals in space and time: the biogeography and evolution of the Scleractinia. Cornell University Press

Wei NV, Hsieh HJ, Dai CF, Wallace CC, Baird AH, Chen CA (2012) Reproductive isolation among *Acropora* species (Scleractinia: Acroporidae) in a marginal coral assemblage. Zool Stud 51:85–92

Willis BL (1990) Species concepts in extant scleractinian corals: considerations based on reproductive biology and genotypic population structures. Syst Bot. https://doi.org/10.2307/2419023

Willis B L, Babcock R C, Harrison P L, Oliver J K & Wallace C C (1985). Patterns in the mass spawning of corals on the Great Barrier Reef from 1981 to 1984