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REPORT

A resilient brooding coral in the broadcast spawning *Porites lobata* species complex: a new endemic, introduced species, mutant, or new adaptive potential?

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Abstract With increasing exposure to local and global stressors associated with a rapidly changing climate, corals adapted to thrive within stressful environments are of particular interest to researchers and managers. A bleaching resilient *Porites* coral with an unusual appearance was discovered dominating shallow waters (1–2 m) within Honolulu Harbor, Hawai'i, a heavily sedimented and polluted habitat with high levels of anthropogenic influence. Continuous monitoring of this 'Harbor *Porites*' revealed prolific year-round brooding and release of planula larvae, with no clear seasonal pattern. Furthermore, recruitment and rapid growth were observed in seawater tanks followed by fusing of various sized colonies, indicating brooding of clonal larvae. Genetic markers placed this coral with high similarity (histone and ITS sequences are 99.9% and 99.4% similar, respectively) to corals in the *P. lobata* species complex which are gonochoric broadcast spawning corals. Fixed differences were observed, and F_{ST} values were high and significant, which could either be explained by reproductive isolation or from clonal sampling over a limited area (this coral has not yet been found in other locations). Observations of skeletal microstructure also showed similarity to corals in the *P. lobata* complex, although with a higher proportion of corallites with excavated columella resulting in a cavity similar in size to the brooded larvae. These observations suggest that bleaching resilience and reproductive mode may be more plastic than previously assumed for *Porites* corals. Additional work is needed to determine if these corals represent a very recent endemic species, an introduced coral, the result of reproductive disruption from pollution (e.g., endocrine disruption), or extreme phenotypic variation within the *P. lobata* complex. Prolific growth and production of larvae, combined with observations of resilience to anthropogenic impacts such as bleaching, sedimentation, and pollution, make this coral a good candidate model for the study of adaptation and acclimatization to climate change and other anthropogenic stressors.

Keywords Bleaching resilience · Sediment tolerance · Brooding coral · *Porites* · Hawai'i

Introduction

Coral reefs have declined dramatically in recent years, with at least half of the world's coral populations affected by large-scale bleaching events that resulted in high levels of coral mortality over the past 40 years (Hughes et al. 2018). Increased seawater temperatures can result in bleaching events and are just one of many stressors known to impact coral survival, recruitment, and reproduction (Omori et al. 2001). Compounding the global threats are the increasing levels of local stressors such as sedimentation, toxicant exposure, and increased algal overgrowth, which can have

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cumulative impacts on the ability for corals to successfully reproduce and recruit (Gilmour 1999; Fabricius 2005; Humanes et al. 2017).

Populations of corals that are uniquely adapted to chronically stressful conditions, such as those within Honolulu harbor, Hawai'i, may be better adapted to global stressors expected to impact coral reefs by mid-century (Golbuu et al. 2016; Palumbi et al. 2014). Honolulu Harbor epitomizes a stressful environment with low circulation, high levels of urban runoff and sedimentation, frequent sewage and petroleum spills, heavy metals from antifouling paint leachate and shedding, and intense commercial activities (Andrews and Sutherland 2004; McMurtry et al. 1995; Loh et al. 1979; Wang et al. 2011; Seligman et al. 1989; AECOS Inc 2014). Water quality in Honolulu Harbor is categorized by the Hawai'i Department of Health as 'impaired.' Runoff from the Ke'ehi/Honolulu Harbor area is responsible for approximately 20% of nutrient input into the southern shore of Honolulu, with four freshwater streams emptying into the harbor area (Laws et al. 1999).

In addition to the sedimentation accumulated from stream runoff, sediment resuspension occurs with propeller wash from the frequent ship movement in the area, with observed increases in turbidity resulting in visibility of only inches (< 50 mm). The presence of both suspended sediments and sediment on the surface of available settlement substrata has a demonstrated negative impact on the ability for coral larvae to recruit and can degrade reef systems at the local level (Gilmour 1999; Perez et al. 2014; Fabricius 2005). Divers who responded to a massive molasses spill within the harbor (233,000 gallons) in 2013 reported coral communities were devastated in much of Honolulu Harbor (personal communication J. WA Murphy). Surveys before the 2013 molasses spill described species within the harbor, and a model of the distribution of species based on that data predicted higher coral cover and diversity of species than is currently present (Coles et al. 2009; Franklin et al. 2013).

Corals within the genus *Porites* can display traits that may benefit them in stressful environments, such as the formation of a mucous layer in response to sediment (Bessell-Browne et al. 2017). The reef-building coral *P. lobata* in particular displays high levels of resilience following bleaching events in Hawai'i, with the ability to recover relatively quickly compared to other species (Hughes 2003; Levas et al. 2013). This may be at least partially due to the perforate skeleton, the subsurface porous connection among polyps, in which coral tissue and zooxanthellae are sequestered (Swain et al. 2018). The genus *Porites* includes the most dominant reef-building corals in Hawai'i (Franklin et al. 2013), with at least a dozen species currently described (Richmond and Hunter 1990). Species-level identification is particularly challenging for corals in this genus, due to several unresolved species complexes that may represent phenotypic polymorphism, hybridization, or very recent species (Forsman et al. 2009, 2017). For example, a variety of closely related branching and massive *Porites* morphospecies form an unresolved clade according to genetic and genomic data (including *P. lobata*, *P. compressa*, *P. annae*, *P. solida*, *P. cylindrica*, and *P. duerdeni*) to form the '*P. lobata* species complex' (Forsman et al. 2009, 2017). *P. lobata* is a dominant coral reef species in Hawai'i and across the Pacific, capable of displaying a high degree of phenotypic plasticity of physiological traits in response to local habitat conditions (Smith et al. 2007).

Here, we sought to identify and further characterize an unusual and apparently resilient *Porites* coral, hereafter referred to as 'Harbor *Porites*.' While almost all corals in Honolulu Harbor showed signs of bleaching or paling (i.e., loss of tissue coloration) during the 2014, 2015, and 2019 thermal stress events, the Harbor *Porites* showed no visual signs of bleaching or paling despite growing in a very shallow and highly sedimented environment. Furthermore, we observed prolific recruitment and fusion of juvenile corals in seawater tanks, prompting us to investigate if this coral is reproducing by clonal brooding of larvae as has been observed in *Pocillopora* (Stoddart 1983), *Tubastrea*, *Acropora*, and *Seriatopora* (Ayre and Resing 1986). To test the hypothesis that the Harbor *Porites* is a distinct species, we compiled observations from the field and seawater tanks, we investigated reproduction with larval collection and settlement trials, and we examined microskeletal traits and several genetic markers (the mitochondrial putative control region, the nuclear histone region, and the nuclear ribosomal internal transcribed spacer) to determine relatedness to other *Porites* corals in Hawai'i.

Materials and methods

Field, tank, and planula observations

The Coral Restoration Nursery at Ānuenue Fisheries Research Center (AFRC) is located immediately next to a reef on the edge of Honolulu Harbor with seawater intakes near the 'AFRC' reef at ~ 1.5 m. The Harbor *Porites* was first observed on this reef near the seawater intake pipes in 2013 occurring in shallow water. The colonies were found at depths ranging from 1 m to fully exposed at extreme low tides. These unusual corals were notable during the 2014–15 and 2019 thermal stress events because they were the only corals on the AFRC reef that showed no signs of bleaching or paling, when monitored every 1–2 d throughout the duration of the bleaching event. During construction of the Coral Restoration Nursery, the Harbor

Porites was observed to recruit into the newly established unfiltered seawater tanks; the source of these larvae was either from several Harbor *Porites* colonies in the tank or from the unfiltered seawater intakes on the AFRC reef. In addition, underwater surveys were performed near the Ke'ehi boat harbor, to assess coral abundance and species diversity within the area. A belt transect 7 × 2 m in length was surveyed along an accessible portion of shoreline near the Ke'ehi dinghy dock, and all coral species (including the Harbor *Porites*) within the transect were counted. Photographs with scale were also taken for colony size analysis.

For initial brooding observations, we first collected a single coral colony measuring 80 × 80 mm near Honolulu Harbor (21°18′56.05″ N, 157°53′27.31″ W) under Hawai'i Division of Aquatic Resources (DAR) SAP 2016–66 in September of 2016 which was transported to the Kewalo Marine Laboratory. Upon inspection under a dissecting microscope, planula larvae were observed being released from coral polyps into a beaker, with more than 400 planula larvae released over the course of 5 h. The coral colony was placed in a closed system recirculating tank with 0.5 um filtered seawater flowing over the colony at a rate of approximately one liter per minute. The tank spillover was captured and filtered through No. 400 nylon mesh-lined collection containers, and cleared daily, with trapped larvae counted after each cleaning. The colony was monitored daily for reproductive output for approximately 1 year. Closed system larval collectors were constructed using individual bowls with a spillover spout where larvae were aggregated over a period of 24 h. Four additional colonies were collected in June of 2017 and were monitored in larval collectors for reproductive output. Planula larvae from all colonies were pooled for use in experiments. These new colonies on collectors were monitored daily for 38 d, and larvae were counted once per 24-h period.

Settlement experiments

Larvae from a 24-h collection period were pooled from all five of the collected colonies and used for settlement experiments. Seawater was filtered twice through 0.2-mm filters (Thermo Scientific, Nalgene, Waltham, MA) prior to use in experiments. Biofilm slides were created by suspending clean glass slides in flow through seawater tables for 4 weeks. Either a biofilm-conditioned glass slide (with no Crustose Coraline Algae) or a clean glass slide, and ten planula larvae were added to each well of sterile 6-well polystyrene plates ($n = 6$). The planula larvae were allowed to undergo settlement in the laboratory at 25 °C (± 0.4). Settlement rates were recorded after 24 h, and recruit development was monitored daily for 1-week post-settlement.

Coral skeletal analysis

Small coral fragments measuring 3 × 3 cm were removed from both the tops and edges of six different Harbor *Porites* colonies. Tissue was removed from the skeletons by soaking in a 5% sodium hypochlorite solution, and the dried coral skeletons for each colony were examined under the microscope (Carl Zeiss Stemi SV 11, 0.6–6.6x zoom). Skeletons were dyed using methylene blue, toluidine blue O, and eosin Y for contrast so that skeletal structures could be more easily imaged.

Genetic analysis

DNA was extracted from coral tissue using a Qiagen DNeasy blood and tissue kit (Qiagen, Hilden, Germany), according to kit instructions. Genetic analyses were performed on the *Porites* colonies collected from the Honolulu Harbor area. Three genetic markers, the mitochondrial putative control region (*CR2:* ~ 400-bp coral mitochondrial putative control region (CR) with primers CRf and CO3r; Vollmer et al. 2002), the nuclear histone region spanning from *H2A* to *H4* (*H2:*primers zH2AH4f 5′-GTGTACTTGGCTGCYGTRCT-3′) and zH4Fr (5′-GACAACCGAGAATGTCCGGT-3′), and the nuclear ribosomal internal transcribed spacer (*ITS;* ~ 700 bp coral nuclear ITS1-5.8S-ITS2 region (ITS) with primers ITSZ1 and ITSZ2; Forsman et al. 2009), were amplified by PCR, sequenced, and analyzed for six colonies for H2 and four colonies for ITS and CR2, as described in Tisthammer et al. (2020), briefly; H2 was amplified under the following conditions: 96 °C for 2 min (one cycle), followed by 34 cycles consisting of 96 °C for 20 s, 58.5 °C for 20 s, and 72 °C for 90 s, and a final extension at 72 °C for 5 min. H2 amplifications (25 µl) consisted of 0.5 µl of DNA template, 0.2 µl of GoTaq® DNA Polymerase (Promega, Madison, WI), 5 µl of GoTaq® Reaction Buffer, 1.6 µl of 50 mM $MgCl_2$, 2 µl of 10 mM dNTPmix, 1.6 µl of each 10-mM primer, and nuclease-free water to volume. For samples with multiple bands, approximately 1500-bp PCR products were extracted from agarose gels after electrophoresis and purified using the UltraClean® 15 DNA Purification Kit (MO BIO Laboratories, Carlsbad, CA) according to the manufacturer's instruction. The rest of the PCR products were purified with UltraClean® PCR Clean-Up Kit (MO BIO Laboratories) and sequenced directly in both directions on the ABI 3730xl DNA Analyzer. Clone libraries were created for each amplified ITS region using the pGEM®-Easy Vector System (Promega). Positive inserts were verified by PCR using SP6 and T7 primers, and

plasmids (2–5 per library) were treated with UltraClean® 6 Minute Mini Plasmid Prep Kit (MO BIO Laboratories) and sequenced on an ABI-3130XL Genetic Analyzer sequencer. H2 was amplified and sequenced using the same method as described above.

The analysis of molecular variance (AMOVA) was performed following the method described in Tisthammer et al. (2020). Briefly, global AMOVA with a weighted average over loci with permutation tests was conducted in Arlequin 3.5 (Excoffier and Lischer 2010), using pairwise difference as a distance computation method. For H2, sequences were phased with PHASE 2.1 (Stephens et al. 2001) and SeqPHASE (Flot 2010) prior to AMOVA tests. For ITS sequences, each cloned sequence was treated as a haplotype.

The net average genetic distances between Harbor *Porites, P. lobata*, and *P. evermanni* were calculated using H2 and ITS in MEGA7 (Kumar et al. 2016) by selecting the 'maximum composite likelihood' model and 'pairwise deletion' for H2 and 'partial deletion' for ITS. Standard error estimates were obtained by bootstrap procedure (500 replicates). Because the obtained sequences were highly similar to the Clade I species complex of *Porites* (Forsman et al. 2009), phylogenetic analysis was conducted using the ITS sequences of the Clade I species obtained from GenBank, along with that of two Hawaiian brooding *Porites* corals (*P. brighami*, and *P. hawaiiensis*) in MrBayes 3.6.5 (1.5 million generations, GTR + G model) (Ronquist et al. 2012) and PhyML 3.1 (1000 bootstrap, GTR + G model) (Guindon et al. 2009). All sequences used in the genetic analysis were deposited in GenBank (Table S1).

Results

Field and seawater tank observations

The Harbor *Porites* showed no signs of bleaching or loss of coloration during the 2014–2015, 2016, and 2019 thermal stress events, while most other corals in Honolulu Harbor were noticeably pale or white. We also observed this coral recruiting, spreading, and fusing over the walls of seawater tanks at the Coral Restoration Nursery at Ānuenue Fisheries Research Center, located immediately next to Honolulu Harbor, over a period of approximately 2 years (Fig. 1). From the field surveys performed in Honolulu Harbor, O'ahu, Hawai'i, following the 2013 Molasses spill, we observed an abundance of Harbor *Porites* coral growing in clusters on highly sedimented substrate often in very shallow waters (from 1 m to a few cm depth). The Harbor *Porites* has a distinctive dark brown and slightly purple hue, with a bumpy surface morphology, which contrasts to other *Porites* observed in Honolulu Harbor and Ke'ehi lagoon such as *P. compressa* (cream or brown colored with finger sized branches) and *P. lobata* (yellow or cream and massive with lobes), or *P. evermanni* (brown and massive with smooth mounds); (Coles et al. 2009).

Among the surveys performed at areas affected by the molasses spill in 2013, only three brooding corals (Harbor *Porites*, *Leptastrea purpurea*, and *Pocillopora damicornis*) have been documented in areas we were able to survey. Surveys performed at two areas within Honolulu Harbor revealed that Harbor *Porites* is the dominant species at these sites; it is not known to occur outside the Harbor or at any other location. Our underwater survey along the shoreline at Ke'ehi counted 734 Harbor *Porites* colonies and 88 *L. pupurea* colonies within the 7 m × 2 m surveyed area. Harbor *Porites* colonies ranged in shape from small encrusting forms to lumpy mounding colonies. The longest diameter measurements for 104 of the 743 *Porites* colonies were recorded, ranging from 0.4 to 12 cm with an average colony diameter of 3.6 cm. Harbor *Porites* was present in our study site at a population density of 52 colonies per square meter.

Planulation and larval recruitment

As our collection permit was initially limited to a single colony, the single Harbor *Porites* coral colony collected in 2016 was placed in a planula collector. On September 2016, the Harbor *Porites* coral was observed to brood planula larvae (Fig. 2a). It averaged approximately 200 larvae per day from a 7.5 cm × 9 cm colony for the first month. Brooding was subsequently confirmed by staff at the State of Hawai'i Department of Land and Natural Resources (DLNR) Ānuenue Coral facility on Sand Island, using additional Harbor *Porites* colonies at their facility. Collection of additional colonies throughout 2017 confirmed that colonies release planula larvae continuously, with no distinct pattern yet observed under closed system laboratory conditions. The larvae contain algal symbionts and are dark brown in color, with an average length of 0.1 mm. Planula larvae readily settled on biofilms, with settlement rates of 80% (SD ± 22.00) on the biofilm slides and 0% on the clean glass slide controls.

Colony characteristics and coral skeletal analysis

This harbor *Porites* colony morphology is superficially similar to *P. brighami*, but its corallite morphology is very distinct from *P. brighami*'s highly excavated structure. A preliminary identification of this species as *Porites cf. studeri* was based on skeletal analysis (D. Fenner, pers. comm). *Porites studeri* was first described in 1907 by Vaughan and is highly similar to *P. lobata* with no discrete identifying characteristics other than small colony size and

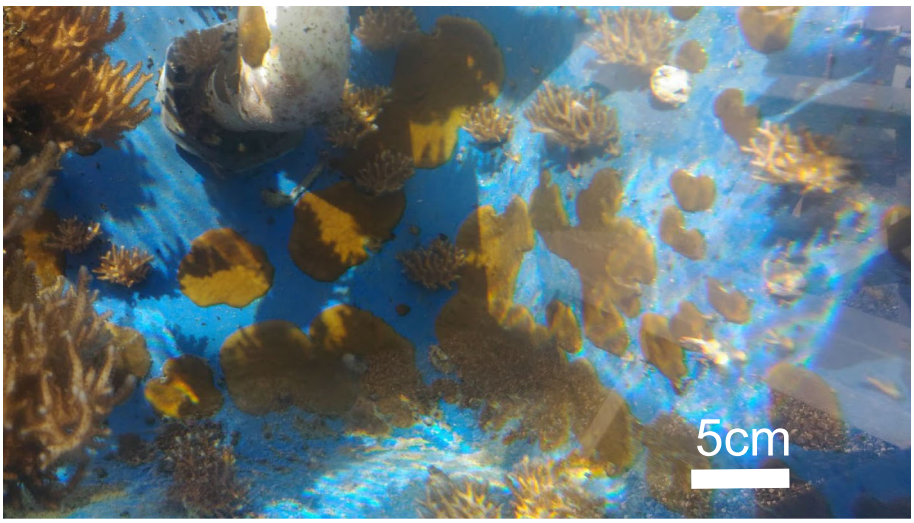

Fig. 1 Coral recruitment, tissue growth, and fusion of the 'Harbor *Porites*' in an unfiltered seawater tank on October 2016. The tank was installed in 2014

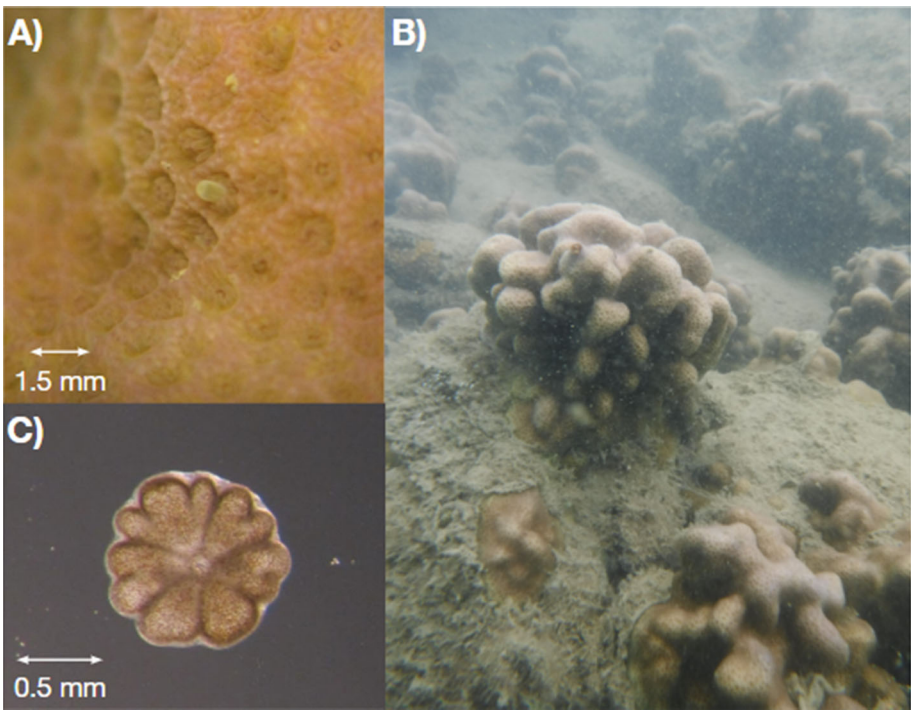

Fig. 2 Harbor *Porites* (**A**) planula larva being released from polyp (**B**) colonies in Honolulu Harbor (**C**) larval recruit 24 h post-settlement

a tendency to occur in deeper waters (Vaughan 1907; Veron 2000). Samples identified as *P. cf. studeri* were genetically identical to *P. lobata*; therefore, it is not clear if *P. studeri* is a junior synonym of *P. lobata* (ZHF unpublished data). The Harbor *Porities* colony color can range from purple to green or brown, or darker brown in the more shaded areas of the seawater tanks, which contrasts with yellow or crème color of *P. brighami* or *P. studeri*. In some instances, all three colors were observed simultaneously in a single coral colony, which is atypical for corals in the genus *Porites*. Average polyp size was approximately 1 mm, with very little variation among polyps within a colony and between different colonies. Asexual extratentacular budding of polyps was observed in several colonies collected.

Skeletal analysis after tissue removal revealed that the corallite skeletal structure of the Harbor *Porites* colonies has a variable columella structure. The original description of *Porites studeri* by Vaughan in 1907 notes the pali as small, crowded down around the columella. The columella of the Harbor *Porites* is absent in most corallites, with the highest number of columella visible in corallites collected from the edge of colonies (Fig. 3A). A very small columella projection was visible in 54% of corallites examined on fragments collected from the edge of colonies (Fig. 3B). In fragments collected from the tops of colonies,

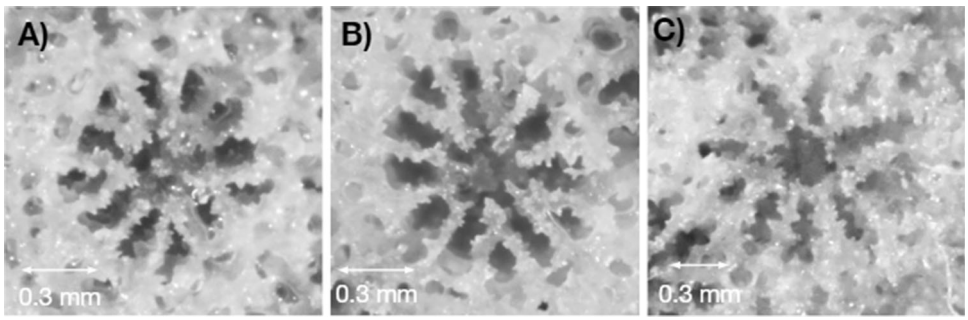

Fig. 3 Columella skeletal structure varies among corallites. The presence of a columella within the corallite ranges from (**A**) prominent (**B**) present but diminished in height (**C**) and completely absent

the columella is absent in nearly all corallites (Fig. 3C). There is an intermediate morphology where the columella is present but diminished in height and flattened (Fig. 2B). The columella is altered, or absent which may provide a brooding chamber for planula larval development within the corallite. Missing columella has also been observed in *P. lobata*, *P. compressa*, and *P. cylindrica*, and the structure of the calyx is well within the range of variation observed in these corals (ZHF personal observation). Corallite size of the 1907 *P. studeri* specimen was recorded as a 1.5–2 mm diameter, while Harbor *Porites* corallite diameter measures 1.1 mm on average (Forsman et al. 2015). We saw minimal variability in corallite sizes among the dozen different colony skeletons we have analyzed, which contrasts with variable corallites of *Porites cf. studeri* samples collected from over 50 m off Maui described by Fenner (2005). The Harbor *Porites* colonies, on the other hand, are found in high abundance in shallow, murky waters, representing the opposite end of the coral reef environmental spectrum.

Genetic analysis

All three genetic markers place the Harbor *Porites* in the *P. lobata* species complex (Clade I from Forsman et al. 2009), which includes *P. lobata*, *P. compressa*, *P. solida*, *P. annae*, and *P. duerdeni* from Hawai'i (Forsman et al. 2009, Fig. 4).

Out of the total 21 ITS sequences obtained from four colonies, there were 15 unique sequence haplotypes (three to five unique sequence haplotypes per colony). Comparing our data with 288 *P. lobata* ITS sequences (Table S1), one fixed SNP locus and one indel ($\sim$ 10 bp) were found exclusively in the harbor *Porites* coral. H2 sequences also showed several potentially fixed SNP loci, when compared to 87 *P. lobata* sequences. However, sequence variability of H2 among the six colonies was extremely low (sequence similarity was $>$ 99.5%), suggesting that the coral individuals in the Honolulu Harbor area may be clones or very closely related (Fig. 5). The mitochondrial marker CR2 had one fixed single-nucleotide polymorphic (SNP) locus in the Harbor *Porites* samples (n = 5), which was not found existing in over 60 *P. lobata*, *P. compressa*, *Porites lutea*, *Porites rus*, *Porites evermanni*, and *P. hawaiiensis* sequences (Table 1).

An AMOVA was performed to assess population molecular variance. The AMOVA results showed significant F_{ST} values between the Harbor *Porites* samples and *P. lobata* sequences from the Pacific. F_{ST} was 0.4965 (P = 0.0000) for ITS and 0.22,019 for H2 (P = 0.0000) Table 1. Since the numbers of samples between the two groups differed substantially, we randomly resampled *P. lobata* sequences to match the total number of Harbor *Porites* sequences and tested with AMOVA ten times. The results did not change the analysis outcomes (ITS average F_{ST} = 0.57782 $\pm$ 0.021, P = 0.0000; H2 F_{ST} = 0.23564 $\pm$ 0.071, P = 0.00005). These high and significant F_{ST} values suggest that the Harbor *Porites* could be a genetically distinct species from *P. lobata*, although closely related or clonal samples coupled with the small sample size could also yield this result and the Harbor *Porites* had higher than 99% similarity to corals in the *P. lobata* species complex (Table 2). It is not clear if these genetic differences represent population structure or species-level differences. The ITS tree supported the finding of genetic similarity of the Harbor *Porites* and *P. lobata* (and other Clade I species), while highlighting a distinct subgroup formed by the harbor *Porites* nested within Clade I (Fig. 4). The two known brooding *Porites* species from Hawai'i and the Harbor *Porites* all belong to separate clades, indicating that reproductive mode in *Porites* is not a conserved trait, even between closely related corals. The estimated genetic distances using ITS and H2 were approximately an order of magnitude larger between the Harbor *Porites* and *P. evermanni* (a non-Clade I Hawaiian species with a similar colony morphology to that of *P. lobata*) than those between *P. lobata* and the Harbor *Porites* (Table 2), confirming the genetic proximity of the Harbor *Porites* to the *P. lobata* species complex.

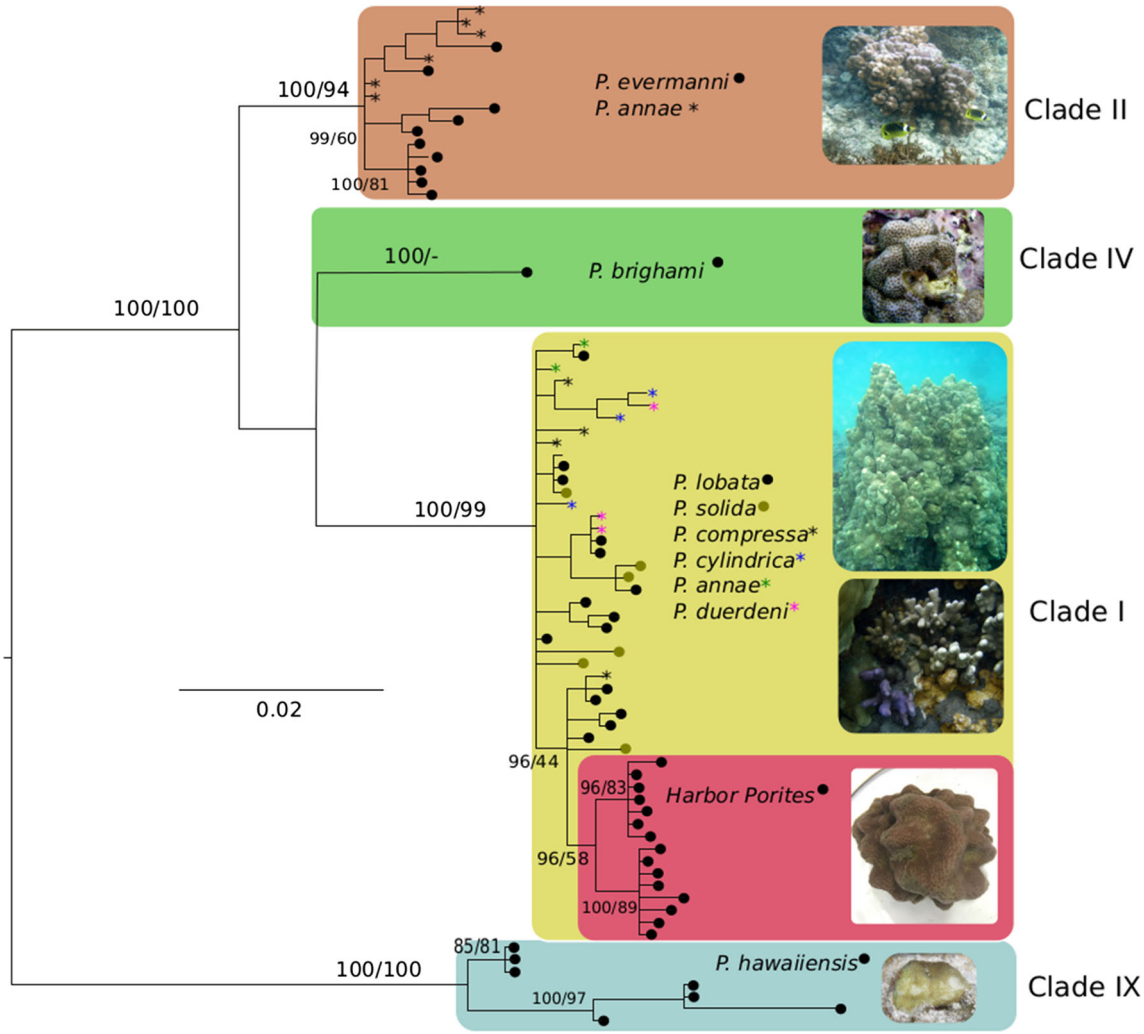

Fig. 4 Bayesian phylogenetic tree of select *Porites* corals based on the ITS region. Each sequence is named with its species names along with the GenBank accession number. The clade numbers are based on Forsman et al. (2009). Numbers near branches represent Bayesian posterior probability values (left) and maximum likelihood bootstrap values (right)

Discussion

Hawaiian corals in the genus *Porites* are among the most resilient species in Hawai'i, yet the characterization of species using morphological or genetic traits remains challenging (Forsman et al. 2009). In Hawai'i, two other species of *Porites* (*P. hawaiiensis,* and *P. brighami*) are brooders, while the rest of the species in this genus are either broadcast spawners or remain uncharacterized (Richmond and Hunter 1990). Outside of Hawai'i, *P. cylindrica* is a gonochoric broadcast spawning species also in the *P. lobata* species complex; however, it was also observed to brood larvae in the Philippines (Abecia et al. 2016); therefore, it may be possible that corals in the *P. lobata* species complex are capable of mixed modes of reproduction under certain conditions. Another brooding coral in the genus *Porites*, *P. hawaiiensis*, was identified as a distinct species discovered in the same area as the Harbor *Porites* (Forsman et al. 2010). The harbor may be an area where hardy coral species can locally adapt to high selective pressures present within the area, and brooding may be part of this strategy. Previous survey work performed in Honolulu Harbor lumped several *Porites* into a single category; therefore, these corals may have been overlooked if they were present (Coles et al. 2009).

Our genetic data found fixed differences and strong genetic structure between the Harbor *Porites* and the *P. lobata* species complex; however, given the high levels of genetic variation within the *P. lobata* complex and the high similarity and low sample size of the Harbor *Porites* (which has not been found outside of Honolulu Harbor), we cannot determine if it is a new species since it falls within the large range of variation found associated with *P. lobata* species complex. The observation that the Harbor *Porites*

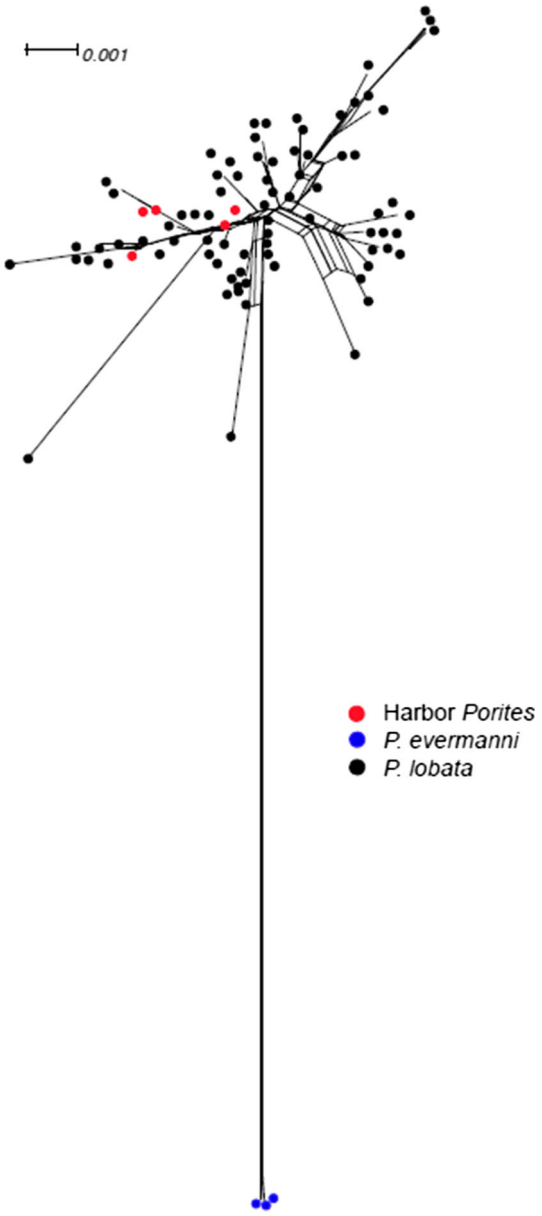

Fig. 5 Diagram of neighbor-net tree network generated by SplitsTree v.4.14.2 for Hawaiian *Porites* corals based on phased H2 sequences

broods planula larvae, yet is very closely related to annual broadcast spawners, indicates that reproductive mode is not conserved and may be more flexible than previously understood for these corals. Reproductive mode therefore is of limited utility for determining if this coral is a new species. Microskeletal observations also do not provide evidence of new species status, since missing columella also occurs (although at a lower frequency) for other corals in the *P. lobata* species complex.

The Harbor *Porites* population within Honolulu Harbor appears to be growing and highly reproductive, despite high levels of anthropogenic stress and multiple unprecedented coral bleaching events. Brooded planula larvae from colonies freshly collected in Honolulu Harbor display high rates of settlement on a biofilm substrate, and recruits developed normally under laboratory observation. This unique coral with its notable ability to survive, grow, and reproduce while exposed to numerous stressors makes it an important species for studying resilience in corals. Its distribution pattern and ability to recruit to substrata in marginal/stressful habitats make it a good target for more focused studies that address key ecological concepts including isolation by adaptation (Nosil 2007) and evolutionary rescue (Bell 2017). Other harbors examined for healthy coral communities include the Port of Miami, where a number of coral species were detected including colonies of *Porites astreoides* (Miller et al. 2016). Interestingly, *P. asteroids* is also a brooding coral, whose symbionts are maternally inherited (Serrano et al. 2016). The recent reports from the National Academies of Science, Engineering and Medicine committee on Interventions to Increase the Persistence and Resilience of Coral Reefs (National Academies of Sciences, Engineering, and Medicine 2019) identified the need to study corals that exhibit the very characteristics exhibited by Harbor *Porites*.

Additional investigation is needed to understand the origin and geographic range of this coral and to understand the basis for its resilience. Genomic and or reproductive

Table 1 Average genetic distances between the Harbor *Porites*, *P. lobata,* and *P. evermmani*, estimated using H2 and ITS sequences. The sample size used in each analysis is shown in parentheses after the genetic marker names

		Harbor *Porites* H2 (5) ITS (21)	*P. lobata*
P. lobata	H2 (85)	0.00118 (± 0.0052)	
	ITS (288)	0.005826 (± 0.00248)	
P. evermanni	H2 (3)	0.02203 (± 0.00475)	0.02120 (± 0.00425)
	ITS (30)	0.04211(± 0.00881)	0.03923 (± 0.00835)

Table 2 GenBank accession numbers for the DNA sequences used in this study

Species	Gene	GenBank accession number	Reference
P. lobata	ITS	FN564941–FN565160	Barshis et al. (2010), Tisthammer et al. (2020)
		KY493091–KY493160	
	H2	KY502280–KY502347	
		KY502354–KY502370	
		MF629151–MF629152	
P. evermanni	ITS	LT558179–LT558182	Forsman et al. (2009)
		LT558196–LT558210	
		FJ416559–FJ416563	
		FJ416586–FJ416590	
	H2	KY502351–KY502353	Tisthammer et al. (2020)
Harbor *Porites*	ITS	Pending	This study
	H2	Pending	This study

work is needed to determine if this coral represents a new species, or extreme phenotypic polymorphism and local adaptation that has been previously overlooked. If this harbor *Porites* is recognized as a distinct species in the future, we propose *Porites halupaensis* as an appropriate name. The Hawaiian word 'Hālupa' means to 'flourish, nevertheless,' which accurately describes the resilience of this cryptic coral within the highly degraded environment of Honolulu Harbor. This coral has the potential to be an important organism for providing insights into resilience to climate change, and local adaptation and acclimatization in a stressful environment.

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Compliance with ethical standards

Conflict of interest On behalf of all authors, the corresponding author states that there is no conflict of interest.

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