Measuring success for Caribbean acroporid restoration: key results from ten years of work in southern Belize

Lisa Carne, Les Kaufman, Karina Scavo

Abstract Coral restoration efforts have become accepted widely as an active management tool but still lack a realistic sense of scale, achievable goals and success indicators. Since Caribbean acroporids are listed by the IUCN as 'Critically Endangered', the general goals of restoration efforts for these taxa are to prevent localized extinction and to promote recovery of self-sustaining populations. Genetic diversity of restored populations is an important consideration regardless of propagation methods (sexual versus asexual), and recent data suggest natural levels of genetic diversity are higher than previously assumed. However, there are few guidelines on the optimal transplant density or spatial arrangement needed to trigger natural regenerative processes at larger scales. Presented here are results from ten years of *Acropora* restoration efforts at Laughing Bird Caye National Park, Belize, where over 59,000 nursery-grown fragments (inclusive of all three Caribbean acroporids) have been out-planted in over one hectare of degraded reef. Data were acquired on host and algal clade diversity, coral growth rates (in nurseries) and survival (for outplants), bleaching history and *in-situ* temperature, reproductive indicators, change in live coral cover, and in fish biomass. All of the above metrics, as well as no-take zones and community involvement, are integral components of successful *Acropora* recovery efforts. Discussed here are three quantifiable indicators: scale (absolute increase of coral coverage), longevity of outplants, and sexual reproduction of nursery-grown, out-planted corals. We suggest realizable goals and success indicators and offer guidance for expanding restoration efforts to new sites as well as suggestions for future monitoring needs.

Keywords: Caribbean acroporids, genetics, restoration, resilience, photo-mosaics

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Lisa Carne
Fragments of Hope, General Delivery, Placencia, Belize

Les Kaufman, Karina Scavo
Boston University Marine Program, 5 Cummington Mall, Boston, MA 02215

Communicating author: Lisa Carne, lisasinbelize@gmail.com

Introduction

As coral health and cover continues to decline globally (Hoegh-Guldberg et al. 2007), reef restoration efforts are becoming more widespread (Jaap 2000; Rinkevich 2005; 2014). Since the Caribbean acroporids were listed on the US Endangered Species Act in 2006 (National Marine Fisheries Service 2015 and the IUCN Red list in 2008 (Aronson et al. 2008), species recovery efforts with these corals have increased dramatically throughout their range (Lirman et al. 2010a; Johnson et al. 2011; Young et al. 2012; Lirman and Schopmeyer 2016). In 2015, NOAA released a recovery plan for these species (National Marine Fisheries Service 2015) that outlines the first attempt at regional goals and success indicators, such as recommended genetic diversity and amount of live coral coverage targets. Weaknesses identified with Caribbean acroporid recovery work to date include a lack of standardized methods for assessing growth rates, survivorship and coral coverage. There are also few published data demonstrating long-term survivorship of outplanted corals (Young et al. 2012; Rinkevich 2014; Mercado-Molina et al. 2015) and a paucity of information about the effects of coral replenishment activities on coral reef ecology.

Much work has been completed on the population genetics of *Acropora palmata* (Baums et al 2005a, Baums 2008, Baums et al 2010) and new results indicate even *Acropora cervicornis* may have higher historical genetic diversity than once thought (Hemond and Vollmer 2010; Drury et al. 2016). However, many questions remain regarding the genetics of the holobiont, inclusive of the corals' symbionts and microbiome community, especially as this relates to thermal stress and/or disease resilience (Baums et al. 2014). The NOAA recovery plan suggests a target genetic diversity ratio of 0.5 for both *A. cervicornis* and *A. palmata* but does not address symbiont or microbiome diversity or function. Acroporids naturally reproduce asexually through fragmentation, so the recommended genetic diversity ratio reflects the proportion of unique genotypes per number of colonies sampled in a specific stand or thicket (National Marine Fisheries Service 2015).

Measurement of the growth and survival of acroporid outplants is complicated by their complex branching patterns, three-dimensional growth and their natural tendency to reproduce asexually by fragmentation (Kiel et al. 2012; Walker et al. 2012). Both *A. cervicornis* and *Acropora* x *prolifera* (a frequent natural hybrid of *A. palmata* x *A. cervicornis*) quickly create thickets that expand and shift over time (Walker et al. 2012, Griffin et al. 2015). It is hard to differentiate colonies from clones without spatially explicit genotyping. This complicates traditional point intercept transect methods where discrete colonies need to be defined. However, digital photo and video tools are now used

frequently to assess benthic cover (Lirman et al. 2007) and can be employed, in conjunction with software like CPCe, to track coral cover before and after replenishment efforts, and/or sequentially to measure coral cover changes over time (Lirman et al. 2010b).

Volunteer sexual reproduction by outplanted coral colonies should be a reef replenishment success indicator. Since cross-fertilization of acroporids (broadcast, hermaphrodite spawners) can be limited by distance between populations, the host genetics of the outplanted corals is required to ensure that multiple genets of each species are placed in proximity to each other, to facilitate heterozygosity in mass spawning events (Baums 2008, Young et al. 2012).

Other measures of success of acroporid reef replenishment work may be increased reef rugosity/structural complexity, higher biodiversity of other corals, fishes, invertebrates and other sessile benthic biota, and increased coral reef community resilience both locally and regionally (through source-sink dynamics). All of these parameters, which collectively are closer to the true intent of coral reef restoration rather than only increased staghorn or elkhorn coral coverage, are as yet largely unexplored and difficult to monitor. Here, we share long-term results from repopulation efforts of the three Caribbean acroporids (inclusive of *A.* x *prolifera*) at Laughing Bird Caye National Park (LBCNP) in southern Belize, with our best efforts at identifying and quantifying success indicators such as longevity, genetic diversity, increased coral coverage and sexual reproduction.

Materials and methods

Coral transplants (2006) and nurseries (2009-2015)

The original *A. palmata* transplants (2006) were naturally broken fragments (or corals of opportunity) averaging ~30cm in at least one dimension and were transferred in seawater-soaked sheets by boat from Gladden Spit and the Silk Cayes Marine Reserve (GSSCMR) to Laughing Bird Caye National Park (LBCNP) (Carne 2008). Fragments were affixed to the reef with a mixture of Portland II cement and Plaster of Paris (as per H. Hudson pers. comm.).

Six A-frame and two table nurseries, with three different culture methods, were established in 2009 with 354 corals representing eight different genotypes (see next section for genetics) of *A. cervicornis*, seven *A. palmata* and two *A. prolifera*, sourced from several different nearby (<20km) locations/reefs (Bowden-Kerby and Carne 2012). Corals were propagated and then fragmented to create second generation colonies. The first massive outplanting began in 2010-2011 after a total of 4,168

second generation corals had been propagated within the coral nurseries. Overall, 3320 *A. cervicornis*, 808 *A.* x *prolifera* and 40 *A. palmata*, were outplanted into 16 shallow (1-2.5 m) subsites, spaced roughly 1- 10 m apart around Laughing Bird Caye. Outplant locations are described in the following way: reef site (here only LBCNP results are discussed), a subsite refers to smaller areas around the reef site, (n=30, Fig. 1) and plots (n=6, Table 3) are even smaller, accurately measured areas where photo-mosaics were conducted within subsites. Each subsite was planted with between 41 and 1,000 second generation fragments, with one to eight genotypes and one to three species per subsite. Different genets of each species were outplanted in proximity to each other with recommended distances of 50 cm-10 m for *A. cervicornis* and 1m-10 m for *A. palmata* to allow successful cross fertilization (I Baums, pers. comm.). Subsite areas ranged from a few meters square to 1600 m^2. In 2012-2013, an additional 1,048 fragments were out-planted.

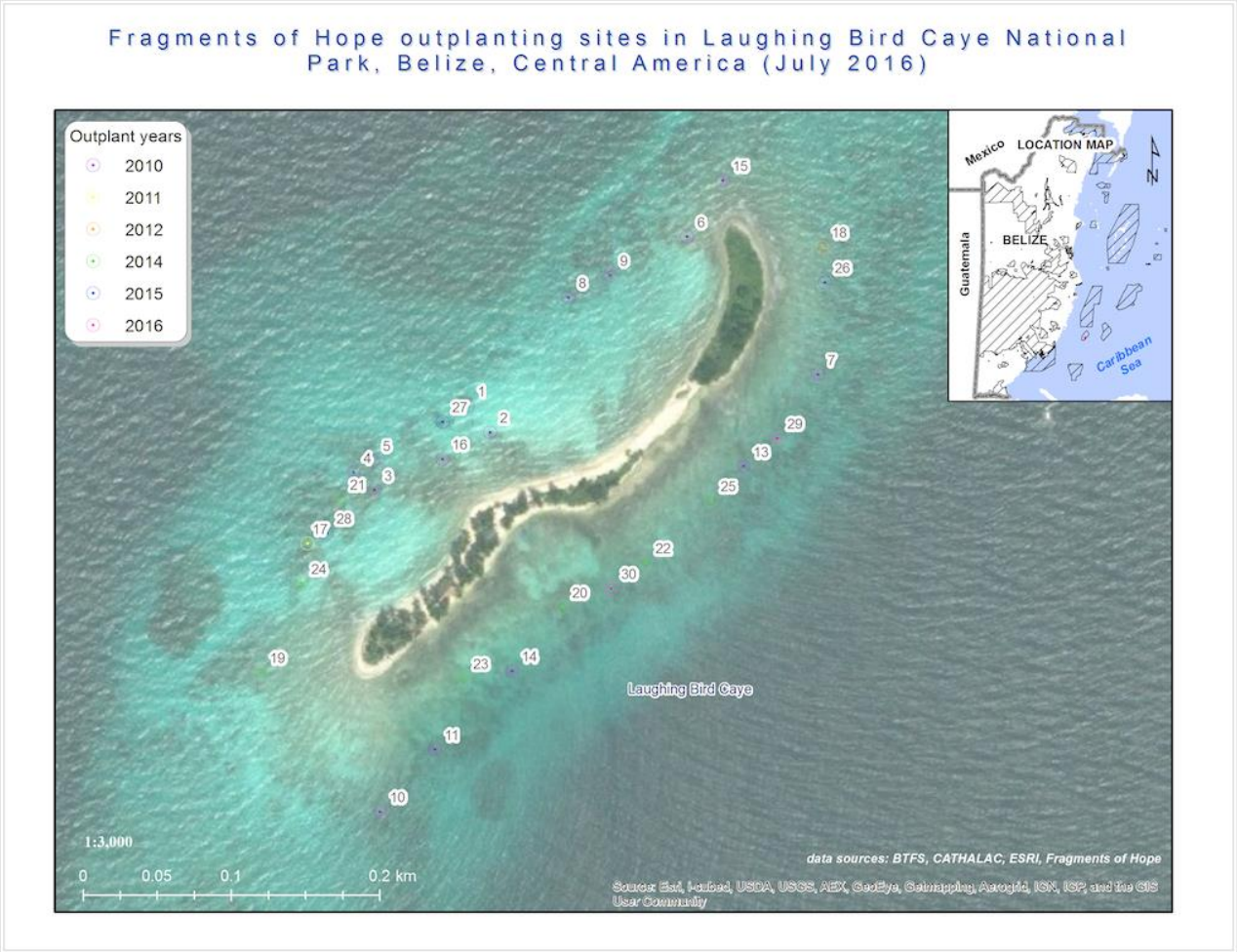

Fig. 1 Map of out-planted subsites at LBCNP with accurate coordinates

Outplanted reef and subsites are chosen with the following criteria: presence of some extant acroporids and/or identifiable dead acroporids, presence of grazers such as parrotfishes, surgeonfishes and *Diadema*, low macro-algal cover and the presence of crustose coraline algae and clear water.

Additional table nurseries were installed in 2013 (two) and 2014 (eleven) with 1,495 additional fragments including both existing and new genotypes (n=11), which allowed subsequent outplanting of 54,069 fragments in 2014-2016 to existing and new subsites (n =30) around Laughing Bird Caye National Park (Fig. 1 and Table 1). The outplanting methods used from 2010-2016 were primarily cement, with some ropes and wedging, and are described in Bowden-Kerby and Carne (2012). For *A. palmata*, single colonies are outplanted while for *A. cervicornis* and *A.*x *prolifera* branches are outplanted in clusters of 5-30 per cement 'rosette'.

Table 1 Summary of the 59,285 acroporids outplanted at LBCNP by year: 2006-2016

Year	Number fragments outplanted	Percent Outplanted
2006	19	N/A
2010	3,816	6%
2011	346	1%
2012	1,026	2%
2013	22	N/A
2014	12,045	20%
2015	9,445	16%
2016	32,566	55%
Total	59,285	100%

Genetics

The original *A. palmata* donor reef site, GSSCMR, was assessed in 2007 using Baums et al. (2005a) methods. All subsequent host samples (2009-2015) were genotyped at five previously published, polymorphic microsatellite loci with Mendelian inheritance as shown by experimental crosses (Baums et al. 2005b). Symbiont genetics 2009-2010 were completed by Baker et al. using ITS-2 and qPCR methods. Symbiont samples in 2015 anlayzed by Baums were genotyped at 13 previously published, polymorphic microsatellite loci following Baums et al. (2014). Unique clonal IDs were assigned to samples that have exact matching multilocus genotypes (host and symbiont considered separately). Symbiont type in *A. cervicornis* was identified by M.A. Coffroth in 2015 (Table 2) using length variation in the Domain V of the chloroplast 23 rDNA (Santos et al 2003).

Table 2 Summary of host and symbiont genetics results and years sampled. The five *A. cervicornis* that housed *S. trenchi* (D1a) in 2009/2010 were rerun in 2015 and all found to now house *S. fitti* (A3); one new *A. cervicornis* genet was added in 2015 and found to house *S. trenchi*. While none of the original *A. palmata* were resampled, 15 new genets were sampled in 2015 and of these, four housed *S. trenchi*

Species	Symbiont type/number with type	Year sampled	No. of colonies sampled	No. genets included in nurseries and outplants
A. palmata	A3/40 D1a/8	2009, 2010, 2015	48	23
A. cervicornis	A3/33(38) D1a/5 (1)	2009, 2010, 2015	40	16
A. prolifera	A3/6	2009, 2010	6	2

Measuring coral and acroporid coverage with photo mosaics

In order to explore the utility of photo-mosaics for monitoring reef surface ontogeny at outplant sites, six plots within out-planted subsites around LBCNP were created, ranging in area from 35.5 m^2 to 180m^2; three are on the windward side of LBCNP, and three to the lee (Fig.1, Table 3). Mosaics of each of the six plots were acquired in August 2014 and again in August 2015. All of the subsites had 0% live acroporid cover before out-planting. At the time of the first photo-mosaic, however, only subsites 23 and 24 were un-planted; subsites 9 and 13 had been outplanted in 2010, with no additional out-planting since that time. Subsites 20 and 21 had been outplanted earlier in 2014 before acquiring imagery for the mosaics (Table 3).

Mosaics were acquired using a dual camera set up as described by Gintert et al. (2008). Processing from raw images to mosaics followed the approach described by Lirman et al. (2007). Changes in total live coral and acroporid coverage over time were quantified using Coral Point Count (CPCe; http://cnso.nova.edu/cpce/) with 400 randomly positioned points per image (Lirman et al. 2007; Lirman et al. 2010b).

Sexual reproduction

The ability of the nursery-grown *A. cervicornis* to sexually reproduce was initially confirmed by histological examination and then by field observations. In July 2012, four fragments were removed from different *A. cervicornis* genets that had been outplanted less than two years before and fixed in 10% seawater-formalin solution, then shipped to the Histology Laboratory at George Mason University. The samples were photographed, decalcified using a 5% formic acid solution, then

Table 3 Summary of mosaic plots, areas, locations and status at LBCNP

Site/ plot name	Location (windward or leeward)	Area (m^2)	% Live acroporid cover 2014	SE	% Live acroporid cover 2015	SE	Change in acroporid cover	Out-plant date/status	Species out-planted
13	windward	182	13.75	0.8	37.14	1.1	23.39	Dec. 2010	ACER, APAL, APRO
9	leeward	110	18.49	0.3	27.02	0.5	8.53	April 2010	ACER
20	windward	144	1.82	0.1	8.07	0.6	6.25	Feb. 2014	ACER, APAL
21	leeward	109.3	3.2	0.4	11.77	0.5	8.57	Feb. 2014	ACER, APAL
23 (UP2)	windward	112	0	N/A	4.43	0.3	4.73	Nov. 2014	ACER, APAL
24 (UP1)	leeward	35.5	0	N/A	7.94	0.5	7.94	Nov. 2014	ACER, APAL

processed and embedded in paraffin, sectioned at 5-micrometers thickness, and mounted on glass microscope slides. Sections stained with Harris's hematoxylin and eosin and Giemsa procedures were examined with an Olympus BX43 compound light microscope and photographed with a DP-72 camera (Peters et al. 2005; Miller et al. 2014).

Visual monitoring of nursery-grown out-planted acroporids (all three taxa) was conducted 2014-2016 at LBCNP over the August full moons: from the full moon date until ~ Day 6 after the full moon from ~20:00-22:30 each night. Dates and times chosen were based on recommendations from natural acroporid spawning data collected at Carrie Bow Caye in Belize from past several years (N. Fogerty pers. comm.). Weather and safety dictated the subsites monitored at LBCNP each night. Photographs and/or video were taken each night.

Results

Genetics

The original *A. palmata* donor reef, at Gladden Spit, was assessed in 2007 and found to have a baseline clonal diversity of 0.7 (of 24 colonies sampled 17 were different genotypes). The host and symbiont genetics were analyzed on 23 acroporid colonies in 2009 (Bowden-Kerby and Carne 2012), another 50 acroporids were sampled for their symbionts only (2010), and 21 more were analyzed for both host and symbiont genetics in 2015, results are shared in Table 2. In 2009/2010, five *A. cervicornis* genets

housed *Symbiodinium trenchi* (D1a), but when these were resampled (from the mother colony, the nurseries and out-plants) in 2015, all housed *S. fitti* (A3). The single *A. cervicornis* that housed *S. trenchi* in 2015 was a new genet, not included previously (Table 2).

Longevity of outplanted corals

Since only *A. palmata* have discreet colonies (versus forming thickets), longevity results are only shared on the first 19 *A. palmata* transplants (2006) and nursery-grown *A. palmata* (n=187, outplanted 2010-2016). The first 19 *A. palmata* transplants at LBCNP were natural fragments sourced from Gladden Spit and the Silk Cayes Marine Reserve in November 2006. Monitoring of these *A. palmata* outplants was conducted through May 2016. Two of the 19 original *A. palmata* fragments transplanted in 2006 were lost (one was dislodged, one unknown mortality) and the simple survivorship is $17/19 * 100 = 89\%$ after nine and half years. This figure cannot account for coverage however, since *A. palmata* also asexually reproduces by fragmentation (from disturbances), yet unlike *A. cervicornis* or *A. prolifera* the 'satellite colonies' can be easily counted. 'Satellite colonies' refers to fragments broken off but surviving separately, from the original colony. For example, a recent (May 2016) count including 'satellite' colonies from the surviving *A. palmata* colonies (17) transplanted in 2006 yielded 48 distinct colonies. Similarly the 187 nursery-grown *A. palmata* out-plants (2010-2016) were assessed in June 2016, and even with one mortality, one missing, and two (same genets) fused, the total number of distinct *A. palmata* colonies was 234. Again, the standard survivorship calculated is $185/187 * 100 = 99\%$, but this does not reflect the increased coral coverage from asexual fragmentation.

Coverage of outplanted acroporids

Mass out-planting of all three Caribbean nursery-grown acroporids began in 2010 and continued through 2016 at LBCNP (1). To date there are 59, 285 fragments/colonies outplanted in 30 subsites at reef site LBCNP, inclusive of the first *A. palmata* transplants from 2006. Table 1 shows the number of out-plants by year, and percent of total, at LBCNP. There is a disproportionate amount of *A. cervicornis* outplanted (51,595 fragments[1]), compared to *A.*x *prolifera* (7,483) and *A. palmata* (207) due to their ease of propagation and fast growth.

[1] "Fragment" is defined for all acroporids as anything large enough to be a 'starter' fragment in the nursery/separate branches and they are counted the day of outplanting.

Since individual tracking of long-term survivorship (and growth) of thousands of outplanted *A. cervicornis* and *A.* x *prolifera* is unrealistic, in 2014 the use of photo-mosaics was employed in six plots within the outplanted subsites, and repeated one year later in 2015. CPCe analyses of the six plots at Laughing Bird National Park revealed changes in total coral and acroporid coverage from 2014 to 2015. The characteristics of the different plots, including size, location, and species and date outplanted are given in Table 3. Only subsites 23 (UP2) and 24 (UP1) were unplanted when they were surveyed in 2014, but all subsites had zero acroporid cover before outplanting. None of the plots measured with photo-mosaics had any additional outplants added, therefore all increased coral coverage is a reflection of natural growth and dispersion from asexual fragmentation.

Total live coral cover increased from 2014 to 2015 in all six sub-sites at Laughing Bird Caye National Park (Fig. 2). All of the total coral coverage increases in 2015 are due to increased acroporid cover, which ranged from 6.5- over 23% in one year from natural asexual regeneration processes (Table 3). The percent increase of total live acroporid cover was the highest in the older (2010) outplanted plot 13 (23%) and the lowest in the newest (2014) outplanted plot, 23 (4.7%) (Table 3, Fig. 2).

Figure 3 is a graph illustrating by species the coral cover on subsite 13 in 2014 and 2015. The coral cover on subsite 13 in 2014 and 2015 increased from a baseline of zero acroporid cover before outplanting in December 2010 to an excess of 35% in August 2015 in the 182m^2 plot (Fig. 3). This large change in acroporid cover can be attributed mostly to *A. cervicornis* that increased from 11.9% in 2014 to 33.2% in 2015 and *A.* x *prolifera*, which increased from .86% in 2014 to 3% in 2015 (Fig. 3).

Sexual reproduction

Gamete development was documented in three different, nursery-grown, *A. cervicornis* genets (and one unknown genet) collected in July 2012, less than two years after out-planting at LBCNP (Table 4) using histology methods.

Only one single *A. cervicornis* colony from one genet spawned on the night of the full moon in 2014. Monitoring continued through Day 6 after the full moon but no other spawning was observed in 2014, and data from wild acroporids at Carrie Bow Caye in Belize recorded spawning unusually late

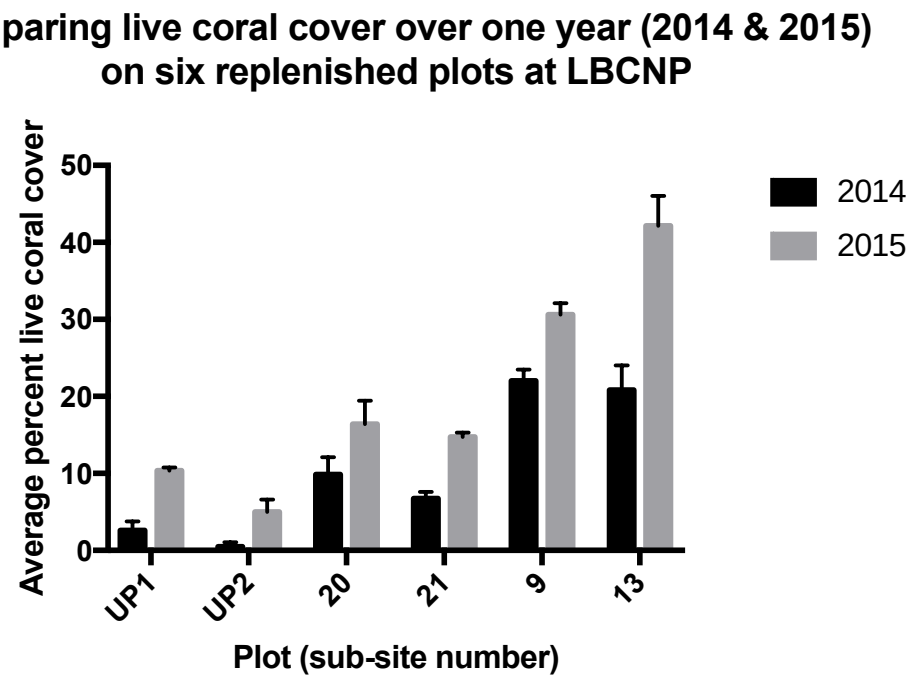

Fig. 2 Live coral at six replenished plots at LBCNP, in 2014 and 2015, assessed with photo-mosaics and CPCe

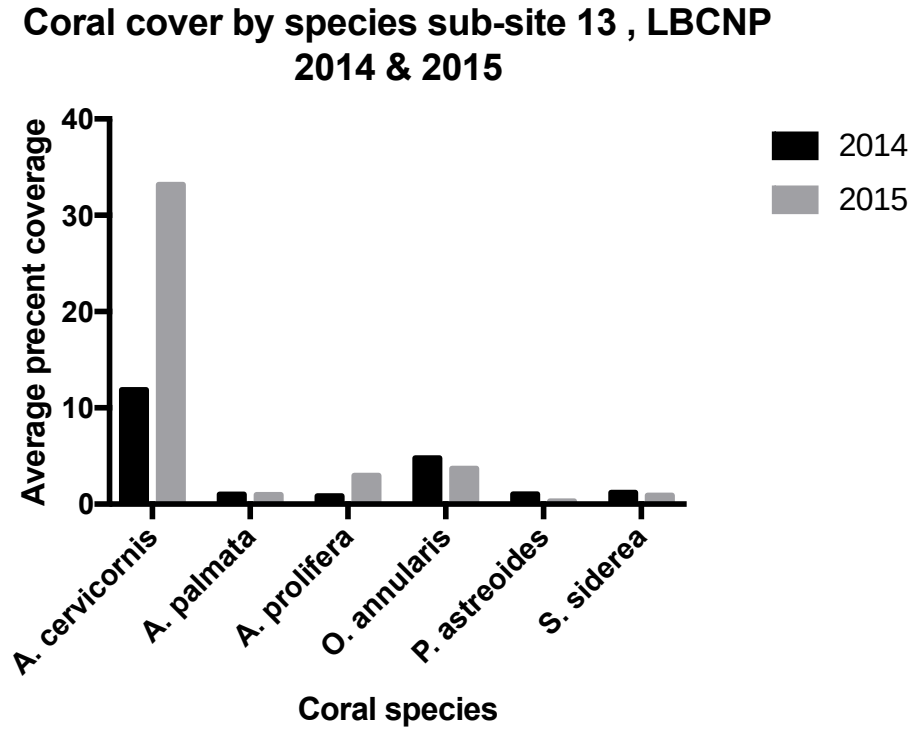

Fig. 3 Live coral cover at mosaic plot 13 (2015) by species

Table 4 Summary of documented sexual reproduction of nursery-grown, outplanted acroporids at LBCNP

Species	Mother/genet	Sub-site	Date outplanted	Spawning 2012*	Spawning 2014	Spawning 2015	2016
Acer	Whipray	9	Apr-10	gametes formed			
Acer	Lazy	13	Dec-10	gametes formed	Y	Y	
Acer	Lazy	28	Apr-15				Y
Acer	Tarpon	15	Dec-10	gametes formed			
Acer	Tarpon	28	Apr-15				N
Apal	Loggerhead 1	13, 5	Dec-10			Y	
Apal	Loggerhead 2	17	Jul-11				Y
Apal	Bugle	13, 5	Dec-10			Y	
Apro	French Louie	13	Dec-10			Y	
Apro	Gladden	6	Dec-10				Y

(Day 8 and Day 9 after the full moon in 2014). In 2015, all three nursery-grown acroporid species spawned (visually documented) on 6 August, which represents Day 6 after the full moon (31Jul 2015). These included two distinct *A. palmata* genets, one *A. cervicornis* and one *A.*x *prolifera* genet at subsite 13 and 5, all of which had been outplanted in 2010. All three nursery-grown acroporids spawned again 23 August 2016, representing Day 5 after the full moon, (18 August 2016) but different subsites were monitored due to weather conditions. The same *A. cervicornis* genet spawned, 2014-2016, but the spawning colonies monitored in 2016 were only 14 months outplanted (subsite 28, Table 4). A different *A. palmata* genet (and age) and *A.* x *prolifera* (genet) was observed spawning in 2016 versus 2015.

In summary, three different nursery-grown *A. palmata* genets, two *A.* x *prolifera* genets and one *A. cervicornis* genet have been visually documented spawning, with outplant ages ranging from 14 months to 5.5 years, and two additional *A. cervicornis* genets showed gamete formation at 19 months outplanted (Table IV). Spawning times for all three years (2014-2016) were ~20:50-21:20 (Belize time) and spawning dates and time coincided with the wild acroporids' spawning at Carrie Bow Caye, Belize (N. Fogerty, pers. comm).

Discussion

Genetics

We consider host genotyping to be a mandatory component of acroporid replenishments efforts for several reasons. First, it is important to ensuring that multiple genets of each species are outplanted in close enough proximity to allow cross-fertilization during mass spawning events, and to aim for targeted genetic diversity ratios in replenished sites. Additionally, as others have shown primarily for *A. cervicornis* (Lohr and Patterson 2016), we note differences in growth rates between genotypes of all three Caribbean acroporids (Bowden-Kerby and Carne 2012), as well as variation in growth pattern (morphology), bleaching and disease resistance and resilience and overall survival (long-term data collection is on-going).

Regarding the symbiont genotyping, it was originally undertaken in order to examine the correlation between bleaching (Bowden-Kerby and Carne 2012) and temperature data collected and even growth and survivorship as a function of genotype (none of which has been presented here). However, based on results obtained between 2009/2010-2015, which reflected *Symbiodinium* spp. shifts (from *S. trenchi* to *S. fitti*) in that time frame, we suggest this should be included at regular time intervals (annually) in replenishment efforts for better understanding of what components may contribute to long-term resiliency. This also affords an opportunity to better understand the shifting relationships between the host and symbiont taxa in the field.

Longevity of outplants

Our results are unusual in documenting an extremely high survival rate of 89% for *A. palmata* transplants from one reef to another (November 2006-June 2016). This should be compared to similar efforts in the USVI that found only 3% survivorship (*A. palmata*) after 12 years (Garrison and Ward 2012), or in Japan, where investigators observed a 20% survival in outplanted corals after four years (Omori 2011). We also document a 99% survival rate for nursery-grown, out-planted *A. palmata* (December 2010-June 2016). While tracking thousands of individual outplanted branches of *A. cervicornis* and *A.* x *prolifera* survivorship over the long-term is practically impossible, a small amount (159 fragments) were tracked six months (April-November) in 2010 and found to have 97% survivorship (best case), and in one isolated worst case, 224 fragments had only 38% survivorship in the same six months. All observed mortality was attributable to predation. With only those two short-term quantitative examples, we refer instead to the mosaic results for tracking these two species with

coverage overtime, and equate increased coverage to survivorship. Anecdotally, we observe far less predation on acroporids at LBCNP than on reef sites outside of protected areas. In Belize, licensed tour guides are trained in coral 'gardening' techniques that include removal of predatory snails on a regular basis. Tourists are required by law to be accompanied by a licensed tour guide in all MPAs in Belize, even for snorkeling. We suggest that choosing outplant sites in well-managed No-Take (Replenishment) Zones that have regular visitation by trained tour guides was a key factor in achieving high outplant survivorship over the long term. Furthermore, by using the local guide community in our reef replenishment efforts, they transfer knowledge, build awareness, and share their sense of ownership and pride in restored sites to international visitors.

Coverage of outplanted acroporids

Coral coverage more than doubled at sites less than one year after outplanting (subsites 23=UP2 and 24=UP1, Fig. 2, Table 3). Even more encouraging results were from the other four plots (in subsites 9, 13, 20, 21) where previously outplanted corals were documented to continue to increase in number (due to fragmentation) and percent cover as they grew in size (diameter and branch complexity) over time, with no additional coral outplanted. The highest increases of acroporid cover were at plot 13: from zero (2010) to over 35% less than five years after the initial (and only) outplanting effort. Several factors may explain these results: the location is on the windward side, with higher water flow and more fish than in the lee; their presence may contribute beneficial nutrients to coral growth (Holbrook et al. 2008). Another contributing factor to the high coral coverage increases at plot 13 may be the *A.* x *prolifera* in larger amounts than any other measured plots (although only a small percentage was captured with CPCe), since this coral fragments and spreads rapidly, creating thickets similar to those formed by *A. cervicornis*.

Although debates continue about realistic restoration goals, the NOAA recovery plan has suggested a target of 5% for *A. cervicornis* and 10% for *A. palmata* in their respective historical habitats (depth and reef types), for at least 20 years. We have surpassed the targeted coverage for *A. cervicornis* for six years albeit in one replenished reef site (LBCNP) at shallow depths, and while we have not yet reached the targeted coverage for *A. palmata*, we have demonstrated high survivorship of this outplanted species for almost ten years. With continued replenishment efforts at LBCNP planned, we propose that these targets can be approached in a reasonable amount of time.

Sexual reproduction

The NOAA recovery plan also refers to the presence of sexual recruits surviving as a success indicator, but in the absence of protocols for documenting this, we offer evidence of genetic diversity (multiple genets of each acroporid species) and documented sexual reproduction of these genets in multiple subsites as crucial steps for measuring evidence of reef replenishment success (Baums et al. 2005a, Vollmer and Palumbi 2007, Young et al 2012). We plan to continue to monitor the outplanted corals' spawning events and recommend that such monitoring be included in replenishment protocols.

Our study is unusual in its relatively long period of observation and high rate of success. The bulk of publications refer to nurseries only, or short term (~one year) monitoring results of outplanted corals. We are in the process of designing protocols and collecting data on acroporid-associated fish abundance and diversity, as well as data on other associates such as crustaceans and sponges that contribute to overall increased biodiversity on replenished reef sites and may also profoundly influence community dynamics. We are continuing to collect temperature and bleaching data and suggest that corals selected for replenishment efforts be sourced from shallow, warmer sites, with the assumption that they have an inherent thermal tolerance. Our replenishment efforts have focused to date in the shallow, fringing reefs around cayes. Not only is this the historical depth range for *A. palmata* in particular, but studies also show that living shallow reefs provide the most shoreline protection (Beck et al. 2014). Furthermore, by choosing to focus on replenishment in these depths, logistically we are unlimited by restricted bottom time when using SCUBA for outplanting, allowing for more corals to be outplanted per person per dive/day. We welcome further collaborations to explore these and other factors that could contribute to the relative success at LBCNP, such as its protected (no-take) status, and local community involvement.

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