

# Using photomosaics to monitor *Acropora cervicornis* thickets created by outplanting nursery-grown corals

S. Griffin, T. Moore, M. Nemeth, A.C.R. Gleason, B. Gintert

**Abstract** In the last decade, coral nursery operations in the Caribbean have expanded from just a few locations to over 50 programs in over 20 countries. These programs have proven to be effective at increasing the abundance of local populations of *Acropora cervicornis* where outplanting is conducted. Typical monitoring of outplanted corals normally focuses on individual colonies. Frequently, monitoring at the level of an individual colony becomes more complicated after 1-2 years as corals grow and intertwine, new colonies are created through fragmentation, tags become hard to relocate as they are overgrown, etc. Longer-term monitoring goals (3-5 years) should look at a broader scale that considers the health of thickets created by outplanting, coral and thicket size (expansion or reduction), asexual recruitment, and changes in the structure and health of the whole reef community. Photomosaics are a useful tool for this type of community monitoring. The research presented here used photomosaics collected annually and analyzed with Coral Point Count with Excel Extensions (CPCE) to monitor the growth and expansion of thickets that were outplanted at 2 grounding sites in Puerto Rico, USA.. Using photomosaics, significant increases in coral cover by *A. cervicornis* and significant decreases in the presence of bare substrate were recorded. Several years after initial outplanting, the outplanted corals at one of the sites have developed into self-sustaining thickets that are expanding through asexual reproduction and have withstood impacts from multiple hurricanes and swells.

**Keywords:** *Acropora cervicornis*, coral propagation, restoration, outplants, photomosaics

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## Introduction

*Acropora* used to be a dominant reef building coral genus in the Caribbean (Jackson, 1994). Over the last few decades, there has been a severe decline in populations of the staghorn coral *A. cervicornis* throughout the Caribbean (Aaronson and Precht, 2001; Bruckner, 2002), which led to the listing of this species, as well as its congenetic *A. palmata* as “Threatened” under the Endangered Species Act in 2005. Current adult populations of Acroporids in the Caribbean typically have low densities and genetic diversity, resulting in a reduction in genetic connectivity (Vollmer and Palumbi, 2007; Baums 2008). As populations continue to decline, proactive intervention is becoming increasingly warranted (Edwards and Clark, 1998; Vollmer and Palumbi, 2007). The life history traits of this genus (fast growth rates and highly successful asexual propagation through fragmentation) make this species a prime candidate for coral propagation efforts in the Caribbean (Highsmith, 1982; Lirman, 2010a). In 2015, NOAA published a recovery plan for this species which includes ramping up coral propagation efforts to help enhance populations and increase chances for sexual reproduction (NOAA, 2015). Coral nurseries are being used to increase population densities of *Acropora cervicornis* on degraded or impacted reefs throughout the Caribbean as well as increase the genotypic diversity of this species on various reefs to enhance successful sexual reproduction (Quinn and Kojis, 2006; Bowden-Kirby, 2008; Johnson et al., 2011, Young et al., 2012; Griffin et al, 2015). When outplanting from nurseries, corals with different genotypes are clustered together to increase the chances for sexual reproduction. The establishment of “reproductive thickets” may help increase connectivity in some areas (Lirman et al., 2010a), and outplanting efforts have already succeeded at creating self-sustaining thickets (Griffin et al., 2015).

This work was conducted at two grounding sites (T/V Margara and the LNG-C Matthews) off the south coast of Puerto Rico, USA near Guayanilla (Fig. 1). In 2006, the T/V Margara damaged 7,500m<sup>2</sup> of coral reef impacting several species of coral including *A. cervicornis* (NOAA, 2015). In 2009, the LNG-C Matthews damaged over 3,000m<sup>2</sup> of reef. Photomosaics (Gleason et al., 2011; Gintert et al., 2012) and diver surveys were used to monitor the success of outplanting activities by focusing on benthic cover, thicket expansion, and percent mortality as key metrics. Here we tested whether the use of photomosaics would work as a tool to monitor these metrics beyond just the individual colony or outplant. In this study, photomosaics were used to estimate benthic cover and thicket expansion while divers collected data in the field on sizes of clusters and percent mortality. The performance of different genotypes was monitored at the Matthews site to allow for comparisons between genotypes.



**Fig. 1** Location of LNG-C Matthew and T/V Margara grounding sites (red circles) off the south coast of Puerto Rico near Guayanilla

## **Materials and methods**

### **Photomosaics**

Photomosaics are composite images formed by aligning multiple overlapping images then blending them together to form a single image. Mosaics allow images of large objects or areas to be collected closer than would otherwise be necessary to fit the entire scene in a single frame. Thus, mosaics permit large areas to be captured at high spatial resolution. In addition, capturing images at close distances minimizes scattering and attenuation of light between the subject and camera, which is particularly important underwater. Underwater mosaic images provide a unique landscape-scale view of the seabed

due to their combination of high spatial resolution (mm or less) with large area coverage (100s to 1000s of m<sup>2</sup>). Marine archaeologists have used photomosaics extensively (*e.g.* Ballard et al. 2002; Brennan et al. 2010; Foley et al. 2009 among many others). Examples of underwater photomosaics used for coral reef science include assessing hurricane damage (Gleason et al. 2007), vessel groundings (Lirman et al. 2010b), cold-water corals (Ludvigsen et al. 2007), mesophotic reefs (Armstrong, 2007; Gleason et al. 2010), and monitoring recovery following disturbance (Cantwell, 2013).

The photomosaics used in this study were constructed with data from GoPro Hero 3 cameras set to progressive-scan 1280x960 resolution mode. The depth (~10m) and visibility (~5-10m) combined for a relatively low-light environment, so the video mode of the GoPro was used. Video mode for the GoPros has produced better image quality in low light than the still image mode, at the expense of about a factor of 2 in resolution (Gintert et al., 2012). Divers swam over the plots of interest with nadir-viewing cameras in a lawn-mower pattern. The divers swam a series of parallel transects in one direction followed by a series of parallel transects in a perpendicular orientation to ensure that the entire plot was captured using images with high overlap. The raw images of the plots used to create photomosaics using software described by Gracias et al. (2003) and Lirman et al. (2007)

### **Margara Site 120**

One of the impacted sites at Margara, designated Site 120, had about 70m<sup>2</sup> of damage to the top of the patch reef where all corals were removed by the grounding. Initial restoration efforts (2006–2008) at Site 120 focused on stabilizing rubble and reattaching dislodged corals. Prior to restoration, no *A. cervicornis* was present on this particular patch reef (Site 120), but there was *A. cervicornis* present on adjacent reefs and within other impacted areas at the Margara site. Approximately 277 fragments (10 - 20cm in diameter) of *Acropora cervicornis* that had been impacted in other parts of the grounding site were reattached at Site 120. Corals were only reattached within the impacted area. Between 2009 and 2011, another 400 colonies of *A. cervicornis* with maximum diameters of 20 – 40 cm were outplanted from a coral nursery that had been set up at the Margara site (Griffin, 2012). These colonies were attached to the substrate using epoxy, cement nails and/or cable ties. Multiple genotypes were outplanted from the nursery to Site 120, but because of how intertwined all of the corals had become over the years, it was not possible to identify individual genotypes anymore. Photomosaics were collected at Site 120 during 2013, 2014, and 2015 to monitor percent cover of benthic organisms within the original impact along with measuring the growth and expansion of the thicket that had been created.

## **Matthews Clusters**

At the LNG-C Matthews grounding site, clusters of *A. cervicornis* were outplanted in 2014. Clusters were approximately 1-2 m apart. Staghorn colonies (6-8) with average maximum diameters of 20-40 cm were planted within each cluster, and only one genotype was planted within each cluster. A total of 5 genotypes were used in this experiment (n=8 clusters/genotype). The genotypes were identified as described in Griffin et al. (2012) following Baums et al. (2005) and (2009) and included loci 166, 181, 182, and 207. This was conducted in 2 different areas at the Matthews site with 20 clusters planted in each area (40 clusters total). Photomosaics and field data (size, survival, health, percent tissue mortality) were collected annually from 2014 - 2016 to monitor growth and survival of clusters. ANOVAs were used to look for differences in percent tissue mortality and Live Area Index (LAI) between genotypes ( $p < 0.05$ ). The LAI is a measure of the percent of live coral tissue per area as described in Williams and Miller (2011).

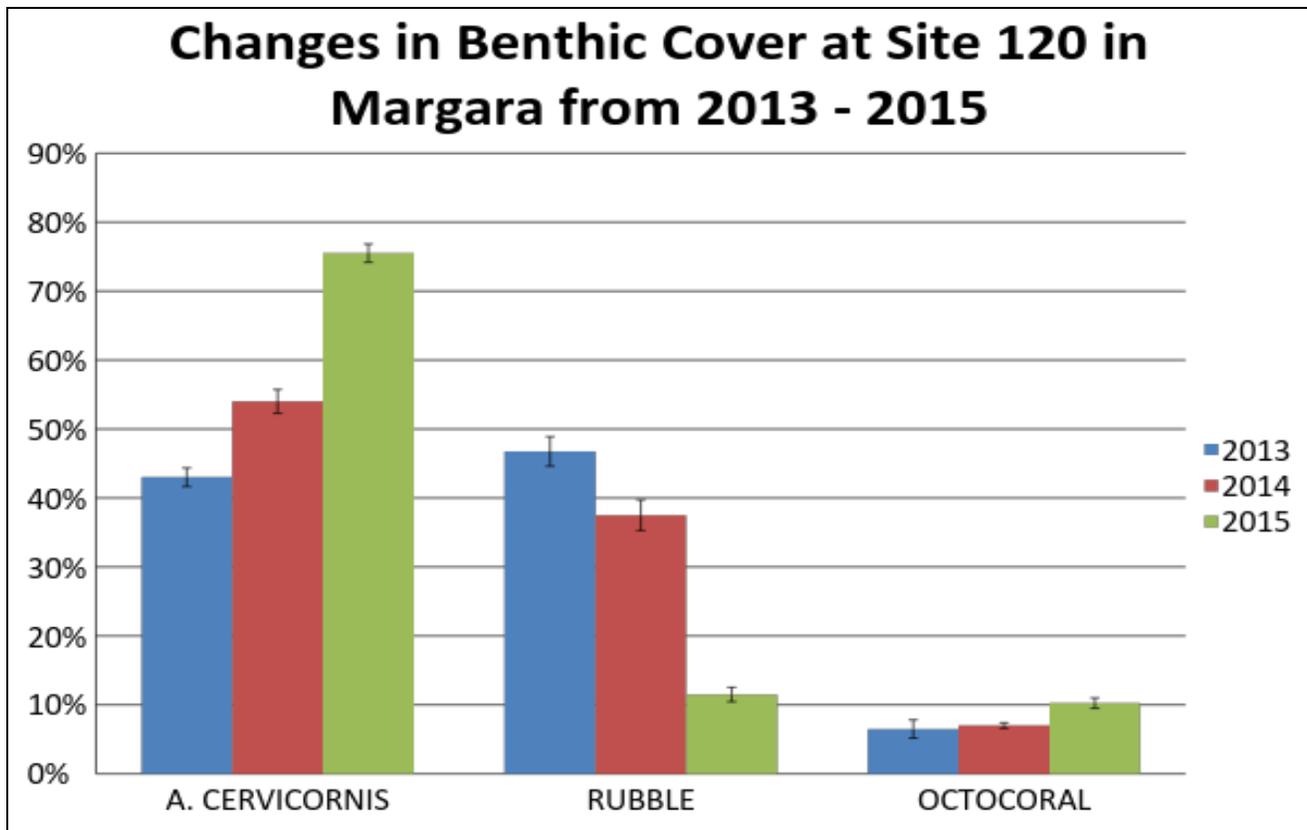
## **Coral Point Count with Excel extensions**

The mosaic images were analyzed using Coral Point Count with Excel extensions (CPCe) software. CPCe was used to estimate percent cover and expansion (Kohler and Gill, 2006). To measure benthic composition, 100 random points were selected by CPCe within the sites where restoration was performed. This was repeated 4 times per mosaic. The mosaics at the Matthews Cluster sites covered approximately 100m<sup>2</sup>, and points were selected within the entire mosaic. The mosaics at Site 120 in Margara covered about 500m<sup>2</sup>, but random points were only selected within the original 70m<sup>2</sup> impact site to determine changes in percent cover within the original impact. The area where *A. cervicornis* was present was also estimated in the Margara Site 120 images using CPCe. Meter sticks that were placed at the site while the images were recorded served as a reference within the images to determine area (Lirman et al., 2007). Using the program, a perimeter was drawn around all of the colonies of *A. cervicornis* at Site 120 to estimate the extent of reef where *A. cervicornis* was present.

## **Results**

### **Margara Site 120**

Percent cover of *A. cervicornis* at Site 120 increased from 43%  $\pm$  1.3 SE in 2013, to 54%  $\pm$  1.7 SE in 2014, and to 76%  $\pm$  1.3 SE in 2015 (Fig. 2). At the same time, there was a decrease in percent cover of

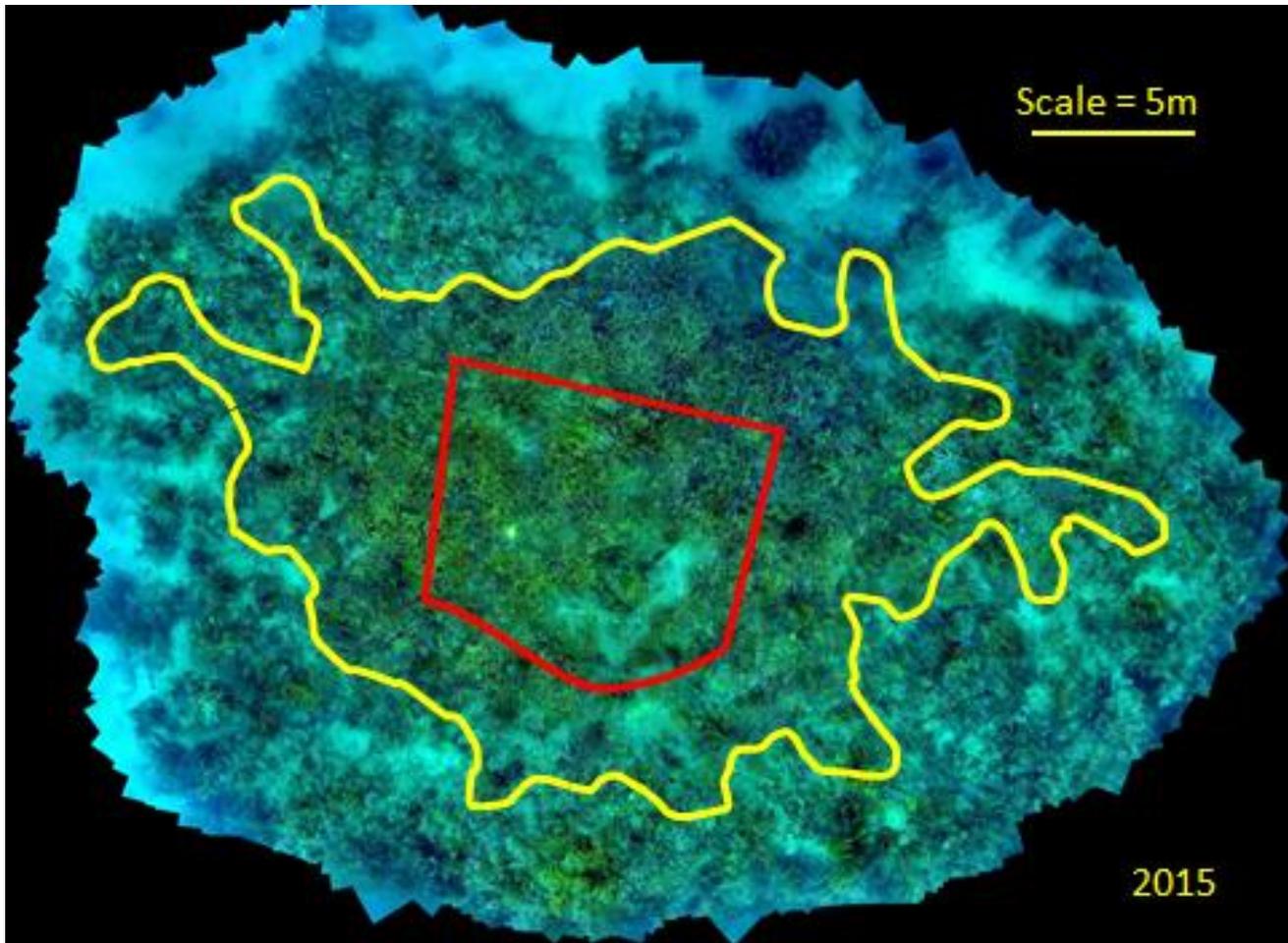


**Fig 2** Changes in benthic cover at Site 120 in Margara from 2013 – 2015. Error bars represent standard error of the mean

rubble from 47% in 2013, 38% in 2014, and 12% in 2015. There was a slight increase in octocoral percent cover over that same time period, from  $7\% \pm 1.3$  SE and  $7\% \pm 0.4$  SE in 2013 and 2014, respectively, to  $10\% \pm 0.7$  SE in 2015. Immediately following restoration and outplanting, *A. cervicornis* was only found within the original 70m<sup>2</sup> of impact (Fig. 3, red polygon). As of 2015, *A. cervicornis* colonies were located in approximately 380m<sup>2</sup> of reef (Fig. 3, blue polygon).

### Matthews Clusters

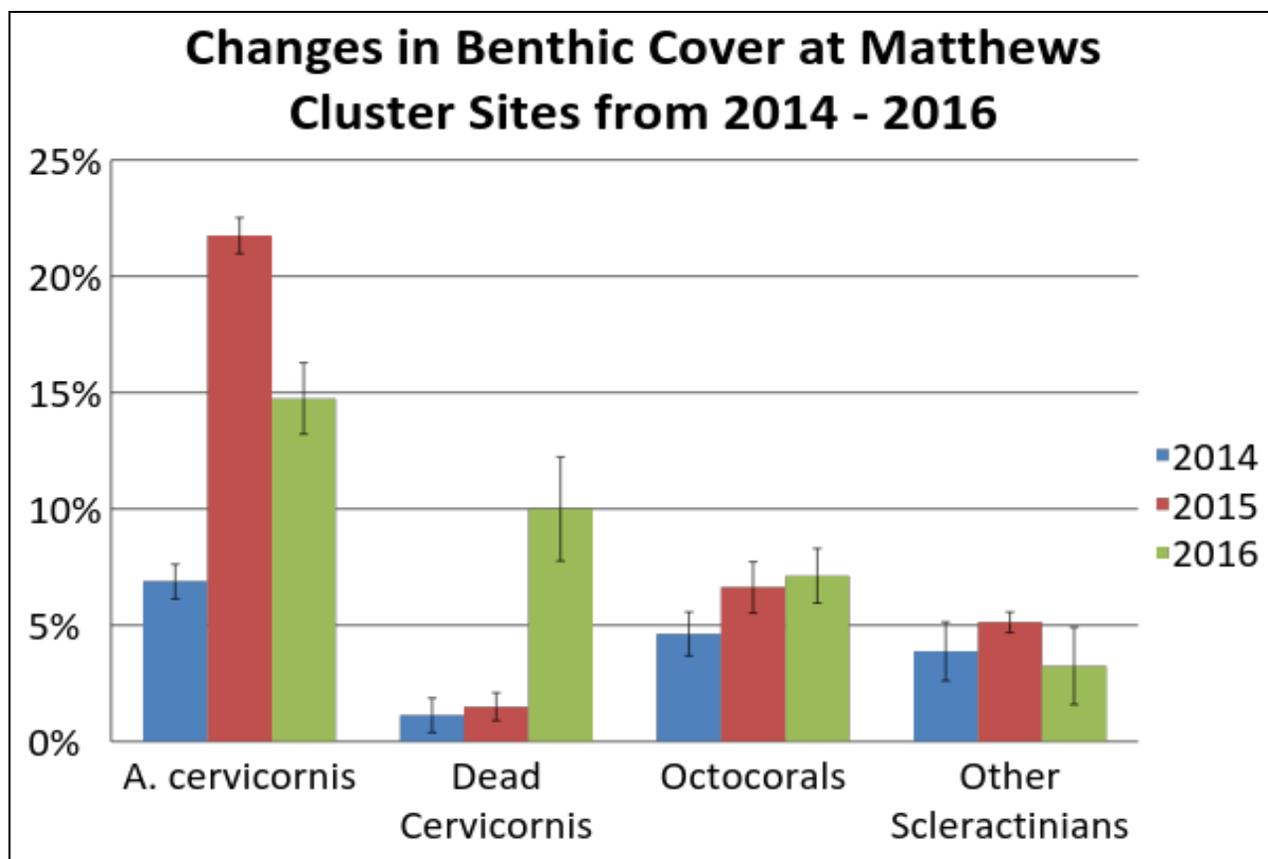
Percent cover of live *A. cervicornis* at the Matthews cluster sites increased from  $7\% \pm 0.7$  SE in 2014 to  $22\% \pm 0.8$  SE in 2015 and then decreased to  $15\% \pm 1.5$  SE in 2016 (Figs. 4 and 5). At the same time, there was an increase in percent cover of dead *A. cervicornis* from  $1\% \pm 0.7$  SE in 2014, to  $2\% \pm 0.6$  SE in 2015 and  $10\% \pm 0.2$  SE in 2016. There was a slight increase in the percent cover of octocorals over that



**Fig. 3** Spatial coverage of *A. cervicornis* in 2015 at Site 120 at Margara. Red polygon ( $\approx 70\text{m}^2$ ) represents the initial impact in 2006. Yellow polygon ( $\approx 380\text{m}^2$ ) shows the borders of where *A. cervicornis* was located in 2015. Colonies at this site were outplanted between 2006 and 2011

same time period from  $5\% \pm 0.9$  SE in 2014 to 7% in 2015 and 2016 while the cover of other Scleractinian corals apart from *A. cervicornis* fluctuated slightly during that time ( $4\% \pm 1.2$  SE in 2014,  $5\% \pm 0.4$  SE in 2015, and  $3\% \pm 1.6$  SE in 2016).

In 2014, the mean size of the clusters was  $128\text{ cm} \pm 5.0$  SE x  $86\text{ cm} \pm 4.1$  SE. In 2015, the clusters grew to a mean size of  $187\text{ cm} \pm 4.8$  SE x  $136\text{ cm} \pm 3.9$  SE. As of May 2016, the mean size of the clusters decreased to  $166\text{ cm} \pm 5.7$  SE x  $114\text{ cm} \pm 4.3$  SE. There was no tissue mortality on the clusters in 2014 when they were first outplanted. In 2015, the clusters had an average of  $17\% \pm 1.9$  SE tissue

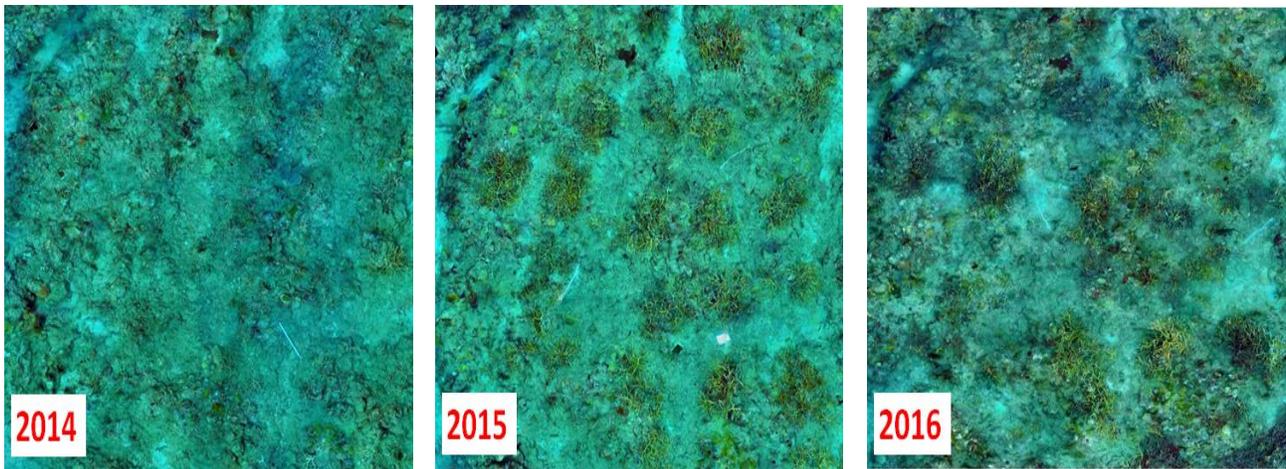


**Fig 4** Changes in benthic cover at Matthews clusters sites from 2014 – 2016. Error bars represent standard error of the mean

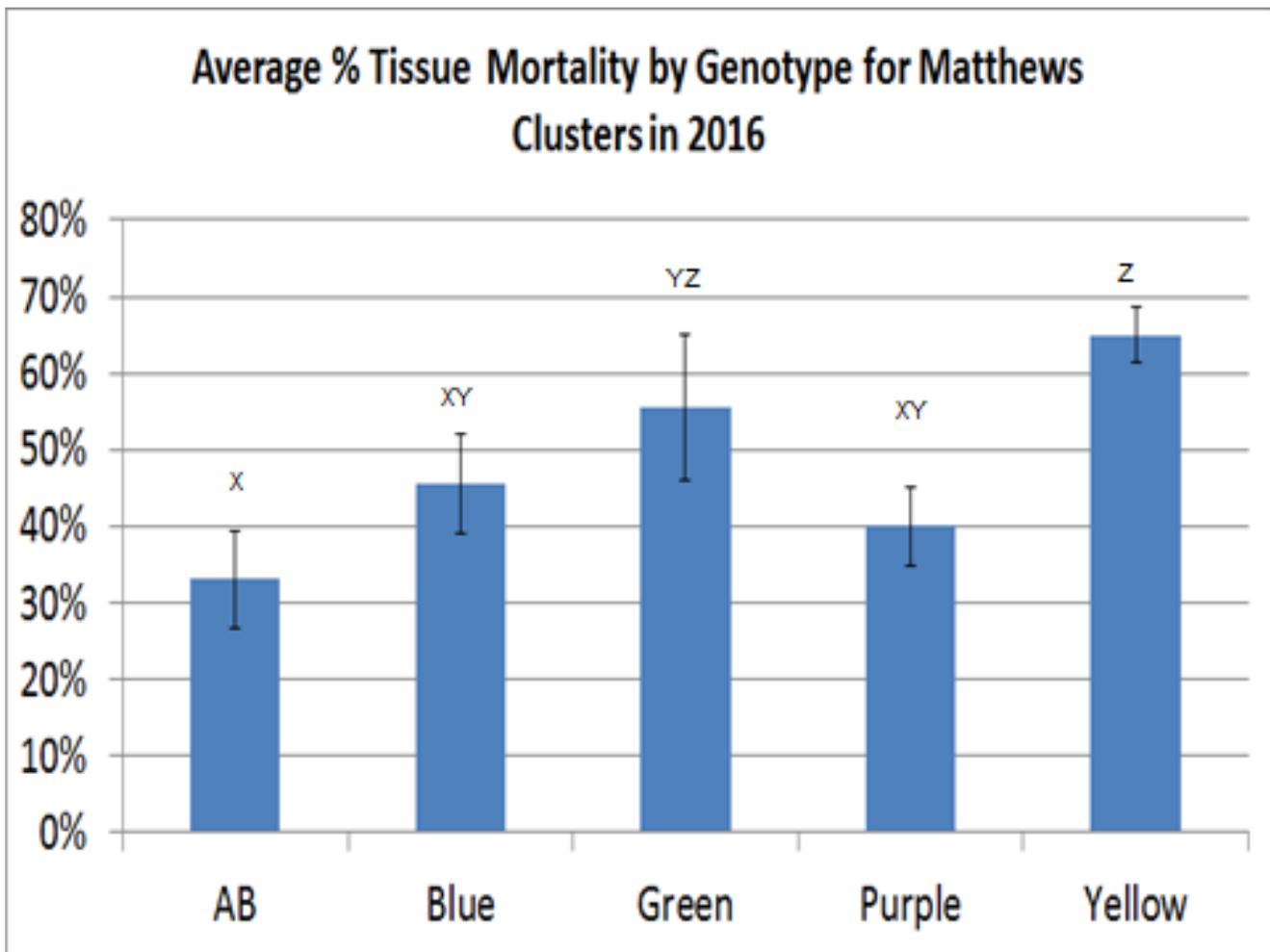
mortality. All of the clusters created in 2014 still had live tissue in 2016, with an average of  $48\% \pm 3.3$  SE tissue mortality. The average percent tissue mortality and LAI in 2016 varied between genotypes (Figs 6 and 7), with genotype “AB” having the lowest tissue mortality ( $33\% \pm 6.0$  SE) and the highest LAI ( $14,244 \text{ cm}^2 \pm 1,668$  SE), while the yellow genotype had the highest tissue mortality ( $65\% \pm 3.6$  SE) and the lowest LAI ( $6,811 \text{ cm}^2 \pm 1,400$  SE).

## Discussion

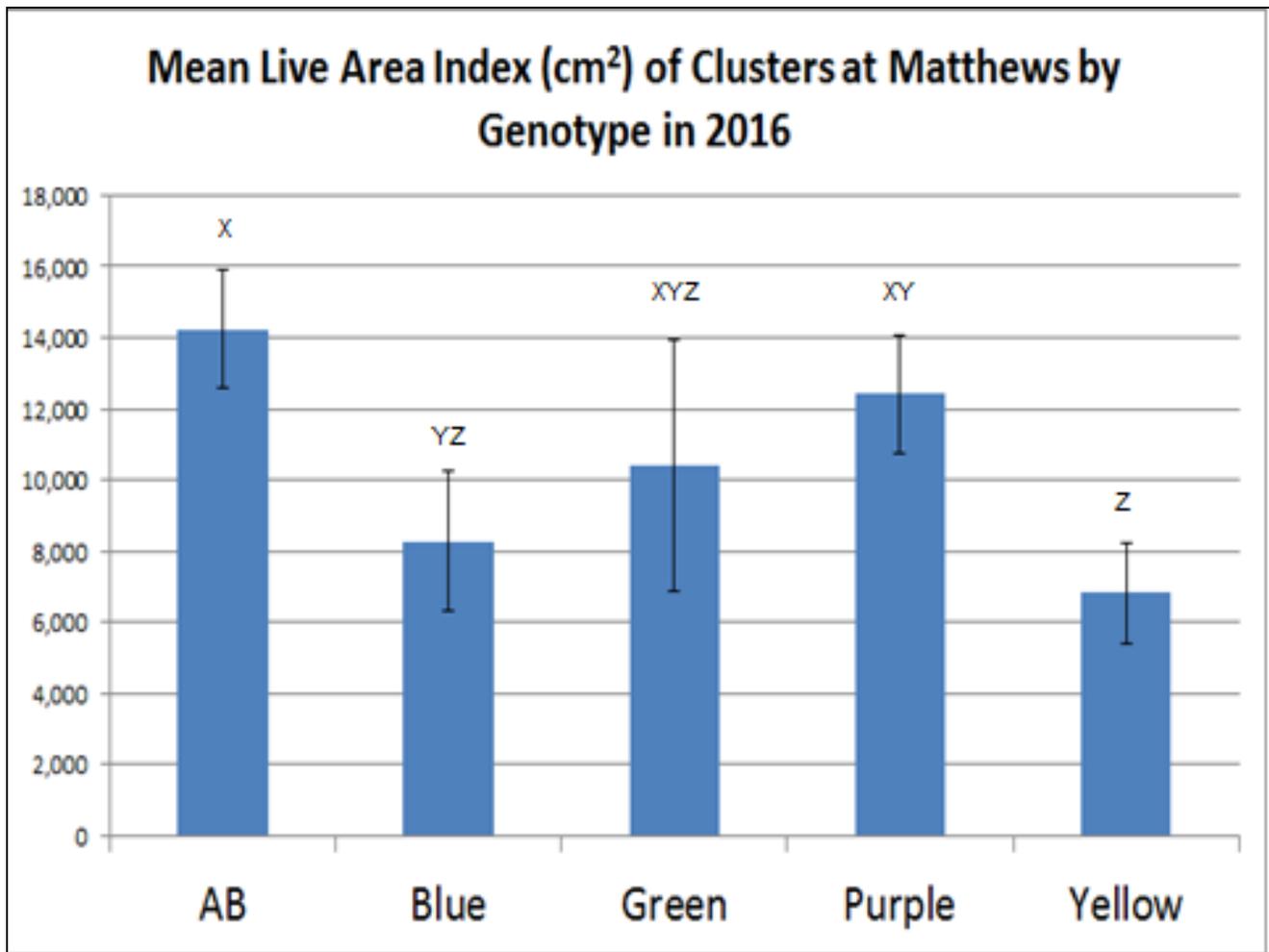
Over the last 10 years, the *A. cervicornis* outplants at Margara site 120 have formed a dense thicket that has not only filled in the area that was impacted (76% cover in 2015), but has expanded from the original  $70\text{m}^2$  where they were outplanted and can now be found in  $380\text{m}^2$  on that patch reef. The



**Fig 5** Photomosaics of the Matthews clusters (Site 44) from 2014 (left photo), 2015 (center photo) and 2016 (right photo)



**Fig. 6** Average percent tissue mortality by genotype for Matthews clusters in 2016. Error bars represent standard error of the mean. Bars with the same uppercase letter (XYZ) above them indicate no statistical difference between genotypes ( $P > 0.05$ )



**Fig. 7** Mean Live Area Index (cm<sup>2</sup>) of clusters at Matthews by genotype in 2016. Error bars represent standard error of the mean. Bars with the same uppercase letter (XYZ) above them indicate no statistical difference between genotypes ( $P > 0.05$ )

percent cover of *A. cervicornis* at the Matthews site is much lower than at Margara, but the clusters at the Matthews site were newer than the thickets at the Margara site. The Matthews clusters were outplanted in 2014 while the corals at the Margara site were reattached starting back in 2006. Future monitoring will determine if the Matthews clusters develop into thickets like at Margara, remain isolated patches or die off. The outplanted corals at the Margara site have developed into self-sustaining thickets that are expanding through asexual reproduction and have withstood impacts from multiple hurricanes and swells > 6 meters in height (Griffin et al., 2015; NOAA, 2015).

Five different genotypes were used in the Matthews cluster study. In terms of robustness, genotypes “AB” and “Yellow” seem to always be at opposite ends of the spectrum and were always significantly different than each other. The “AB” genotype has been shown to have the lowest rates of mortality in

the nursery (Griffin et al., 2012). It is important to outplant as much genotypic diversity as possible to help increase the chances of sexual reproduction and increase the populations' chance of resisting unpredictable disease and bleaching outbreaks (Lirman et al., 2010a). But if the weaker genotypes continue to fail, future efforts may need to focus on the more robust and resilient genotypes. It is still too early to decide whether or not to give up on the "Yellow" genotype because it has not completely died off yet. A few more years of monitoring may be able to answer this question.

In 2016, there was a significant amount of mortality within the clusters at Matthews (48% tissue mortality). As thickets develop, there are years of healthy growth or die off. The percent cover of *A. cervicornis* surged from just 6% in 2014 to 22% in 2015 and then decreased to 15% in 2016 (Fig. 4). Die backs such as this could be due to disease, predation, storms, or other environmental parameters. Continued monitoring at the site will allow us to determine if this is just part of the normal ebb and flow of a developing thicket or if these corals are actually dying. Future monitoring will also track how the different genotypes fair, and genetic analysis will be performed on the different genotypes to try and identify any resilient genes.

Traditional monitoring methods used to assess the efficacy of coral restoration efforts have focused mainly on the growth and survival of individual coral outplants (Johnson et al., 2011). This provides useful information for understanding the success of the individual outplants and comparing the performance of different genotypes. But over time (3-5+ years), it becomes difficult to keep track of these individuals as they fuse with other outplants of the same genotype, move about after storms, and/or fragment and create additional colonies. Longer term monitoring needs to focus on a larger scale looking at the entire reef community. This study expanded on prior, colony-based monitoring efforts by using landscape photomosaics to get an understanding of how thicket formation is occurring and how the thickets themselves are expanding or contracting. This study is an example of how photomosaics provide a useful tool for measuring percent cover, expansion, and colony survival at a plot-patch reef scale and assessing changes in community structure.

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