

Strengthening coral reef resilience to climate change impacts: A case study of Reef Restoration at Laughing Bird Caye National Park, Southern Belize



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2011**



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Cover photo: A Goliath grouper. *Epinephelus itajara* (an endangered species) takes refuge under the coral nursery table at Laughing Bird Caye National Park, in Southern Belize, a No-Take Zone since 1994.

Background and Significance of the problem

Coral reefs worldwide are declining in abundance and health due to multiple impacts. Pollution, coastal development, sewage, overfishing and irresponsible tourism practices negatively affect coral reefs but climate change is the most significant threat today. Elevated sea temperatures have caused massive bleaching events for over two decades which often leads to coral death, slows down the growth rates for the corals that do survive, and are often coupled with coral disease outbreaks as well. It is estimated that 75% of the world's coral population is at risk from these combined threats¹. In the Caribbean, live coral cover has been reduced by at least 60% in just a few decades².



The reefs near Placencia, Belize early 1970's³.



Sadly many reefs have not recovered from recent impacts. Dead Elkhorn reef (*Acropora palmata*), Gladden Spit & the Silk Cayes Marine Reserve, 2006.



Dead Staghorn reef (*A. cervicornis*), Laughing Bird Caye National Park, 2010.

¹ February 2011, World Resources Institute (www.wri.org).

² Pala, Chris. *The Ecologist*, 2011.

³ From Straughan, Robert ,P.L., *Adventures in Belize* (1975).

In 2006 over 140 reef sites in Belize were surveyed using the Atlantic and Gulf Rapid Reef Assessment (AGRRA)

methodology (www.agrra.org). Belize's average live coral cover was a lowly 11% (compared with 40-60% as a 'healthy reef' average).

However, these surveys also confirmed what many tour guides and fishermen had been observing in the waters near Placencia: the near-shore, shallow reefs were actually healthier than the outer coral reefs. The question was why? As these areas are hotter and stiller and closer to mainland human impacts, the opposite would be expected.

There are many theories that include pollution coming from Guatemala and Honduras up to our reefs in the outer ocean currents, but one of the premises of this project lies in the theory of thermal adaptation for corals. Meaning if corals thriving in Placencia's inner cayes are already used to hotter sea temperatures, and have survived several impacts already, these might be the best ones to study for resilience to bleaching events and to propagate for long-term climate change mitigation strategies.

Read more in the Basic Coral Biology and Bleaching explained section.

Hurricanes are increasing in both frequency and intensity and Laughing Bird Caye National Park in Southern Belize took an indirect hit from Hurricane Mitch (Category 5, 1998) and a direct hit from Hurricane Iris (Category 4) in 2001. Even tropical storms (Mathew, 2010) can cause significant structural damage, overturning old large coral heads.

After Hurricane Iris, there were virtually no Elkhorn nor Staghorn coral colonies left alive at Laughing Bird Caye National Park. These two species (*Acropora palmata* and *A. cervicornis*, respectively) were formally the most common corals in Belize and the Caribbean but their abundance has been reduced by over 98% Caribbean-wide, directly due to climate change impacts, in just decades. They are fast growing, branching species that provide habitat for hundreds of other marine creatures, including the commercially important Caribbean Spiny Lobster. They are also the main reef building coral species in the Caribbean, providing important shoreline protection from surge, waves and storms and help to prevent coastal erosion. They have aesthetic value for tourism, which in Belize, employs one in four people, which translates to economic value. In 2006 they were placed on the IUCN's Red List (www.iucnredlist.org) as Critically Endangered species (along with their hybrid, *Acropora prolifera*), which is only one step away from Extinct in the Wild.

In May 2009, Southern Belize was damaged by a 7.1 earthquake that struck 80 miles NE of La Ceiba, Honduras. While people were worried about damage topside, fishermen were reporting open crevasses on patch reefs and underwater 'landslides'. In an emergency survey to check the nurseries⁴ the fishermen's reports were confirmed. The western and southern sides of every patch reef and caye surveyed had underwater landslides that

⁴ Funded by Healthy Reefs Initiative.

destroyed live reef but also dramatically revealed the historical coverage of *A. cervicornis* by exposing so much skeletal remains.

The nurseries were happily unaffected.



The new edge at Tarpon Caye, revealing the historical coverage of *A. cervicornis* by the exposed remains.



More exposed *A. cervicornis* skeletal remains at French Louie Caye.



Lobsters love *Acropora palmata* not only for hiding but because they like to eat a snail that eats the coral, so they get food and shelter from these corals. This coral was transplanted to Laughing Bird Caye National Park in 2006, and the lobster is so big because the park is completely off limits to fishing and has been for over ten years.

Acroporid corals, although animals that can sexually reproduce (they are hermaphrodites that spawn), can also reproduce asexually by fragmentation-similar to how some plants can be propagated from cuttings. This is a natural adaptation strategy because they live in high wave action environments: if storms cause breakage and the pieces don't get too knocked around, they will adhere to the substrate and grow in their new location. This work takes advantage of these corals' natural ability to reproduce asexually (see more in basic coral biology section).

Fragments broken from storms can continue to live if they remain stable. Gladden Spit & the Silk Cayes Marine Reserve, 2006.



Restoration work at Laughing Bird Caye National Park first started in 2006 with funding from PACT (Protected Areas Conservation Fund) based in Belize, and Project AWARE (2007). In a trial project, 'fragments of opportunity' (pieces

of Elkhorn coral broken naturally from storms, but still living), were moved to Laughing Bird Caye National Park from healthy reefs in Gladden Spit and the Silk Cayes Marine Reserve. Harold Hudson, formally with NOAA in the United States, and known as the "reef doctor" for his extensive experience with coral restoration, was an inspiration, mentor and advisor for this work.

A literature search for the project proposal revealed one of the first publications on this type of work from Dr. Austin Bowden-Kerby. We met in person in Mexico in 2006 and formed the liaison that led to the first nine nursery sites established in Belize (2009), funded by the World Bank and the Caribbean Community Climate Change Centre. World Wildlife Fund Central America has kept the project alive with additional support funding since 2009. This document outlines the methodologies used, results to date, lessons learned, and the suggested next steps.

The goals of this project include saving the Acroporids from extinction in the wild, restoring lost ecosystem services at Laughing Bird Caye National Park like shoreline protection, habitat for marine creatures, and aesthetic value for tourism. Besides the restoration efforts, it is hoped that the science behind this project will reveal new information on how and if corals can adapt to rising sea temperatures, and give us new insight into the biology behind resilient corals.

An overview of the Belize project, including two short videos, can be found on the Facebook page "Fragments of

Hope". Austin Bowden-Kerby's award winning work in Fiji is explained on his website www.coralsforconservation.com.

Basic coral biology and bleaching explained

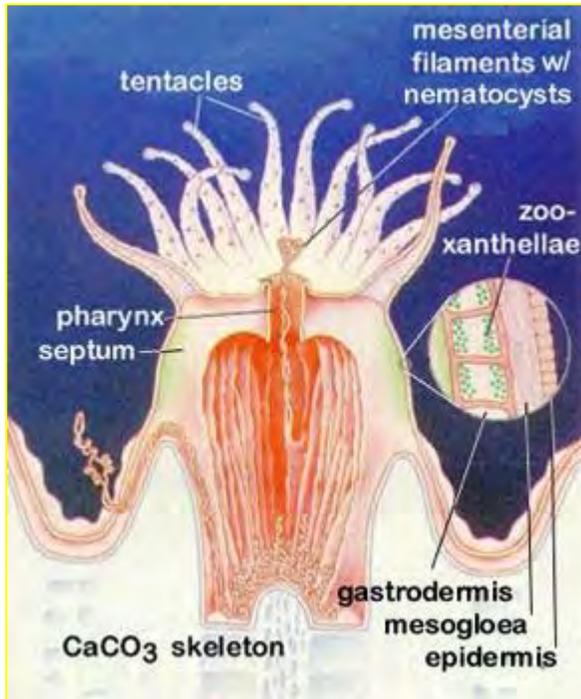
Yes, all corals are animals! They are marine invertebrates in the Phylum Cnidaria, which means they are related to jellyfish. Most corals are also colonial organisms, which means many individual animals make up a coral colony. An important aspect about corals is their symbiotic relationship with single-celled algae called zooxanthellae.

A symbiotic relationship is one where both organisms benefit from their association with each other. In this case the zooxanthellae have a permanent home (host), the coral polyp—they reside in the tentacles. The zooxanthellae are what give corals their color, and being algae, they can photosynthesize (make energy from sunlight) like plants. Although the corals do eat with their tentacles, they benefit greatly by using the by-products of photosynthesis for energy and growth; in fact corals get about 90% of their energy from the zooxanthellae.

Until recently it was believed that there was only one type of algae that forms a relationship with corals, however genetics have revealed a wide array of diversity within zooxanthellae. They are being characterized as "clades"

(which is somewhat broader than a species level) labeled A-I, and sub-clades (more like a species level distinction), as in A3, D1. It seems there are some specific associations: certain coral species are associated with certain algae clades, but this may also be influenced by environment. For example, corals with Clade D are most often found in warmer waters. To make matters more complicated, corals can host more than one type of clade; these are referred to as dominant and background clades.

Understanding the biology of these relationships is crucial to understanding the mechanisms of bleaching, and more importantly, to understanding the mechanisms of 'resilient' corals: the ones that can survive bleaching.



A basic diagram showing the location of the zooxanthellae within the coral's polyp (tentacles). From NOAA's Coralwatch.



Bleached reefs showing varying degrees of paleness, at Middle Silk Caye, Belize (October, 2008).

The most common description of a bleaching event is that the zooxanthellae are expelled from the coral host, usually associated with elevated sea temperatures⁵. This causes the coral to turn white (varying shades of pale depending on the level of bleaching occurring). Bleaching can actually be caused by any stress: cold water, fresh water, sedimentation, etc. However, over the last few decades, we have gotten better at predicting bleaching events by the weather: flat, still hot days for a long period can be expected to cause bleaching events. NOAA has a website that uses sea surface temperature data (from satellites) and weather conditions to predict bleaching events.

⁵ The zooxanthellae can actually be absorbed, or the entire polyp cell containing the zooxanthellae can die. Weis, 2008.

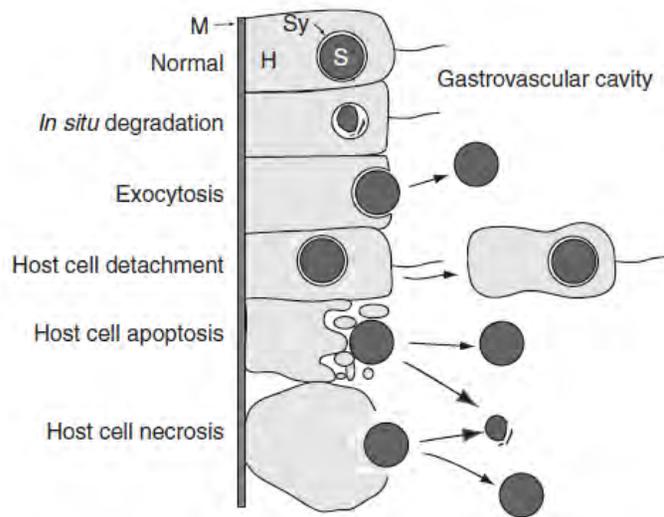


Illustration of the different mechanisms of bleaching. Weis, 2008.

However even knowing the bleaching events will occur is not enough. At this point we still cannot predict which corals will survive bleaching events and which will not, and there are no real preventative actions to take...in small scale experiments bleaching has been prevented by 'shading' the corals from the sun, but this is obviously not feasible for an entire reef/ecosystem.

Instead, we are striving to learn more about this special relationship corals have with their algae clades by studying both the coral host and their symbiotic algal genetics, and observing bleaching patterns with these specific corals over time.

As this is relatively new research there are still many debates and unanswered questions among the experts.

Many feel the answer to a coral's resilience to bleaching lies within the type or types of algae clades it hosts, while a smaller few believe the genetic make-up of the coral host may be more significant to its survival of bleaching events.

Some of the most important questions in coral biology today is if any corals have the ability to adapt to rising sea temperatures, understanding this adaptation process, and if we can facilitate this adaptation process so that they may adapt as quickly as the sea temperatures rise (and acidification increases). It is hoped the results of this project will continue to shed new light on these topics.

Acroporid reproduction

Much of this work capitalizes on the Acroporids' natural ability to reproduce asexually. It seems this reproductive strategy evolved because of these corals' preferred habitat: high wave action is where they thrive. When storms (or sometimes even the weight of the coral branch) cause breakage, these fragments can continue to survive once they remain stable (don't get rolled around too much). We are now calling these 'fragments of opportunity' because nature (or a ship grounding, or anchor damage) creates loose, living coral pieces. These pieces can either be

reattached to the reef, or moved to a nursery site to make more corals.

Acroporids can also sexually reproduce. They are hermaphrodites (both male and female) and breed by mass spawning events (usually around July, August or September full moons but the dates can be variable both regionally and between reefs).

Crucial to understanding our efforts at effective reef restoration is understanding the difference between genets and ramets. Genets, or genotypes, are unique, distinct corals of the same species. They have a genetic make-up unique to other genotypes, and this is created by sexual reproduction.

Ramets are 'clones' of the same genotype, for example a fragment of opportunity is the exact same genetic make-up as its 'mother' colony, and the two cannot sexually reproduce. See Fig X.

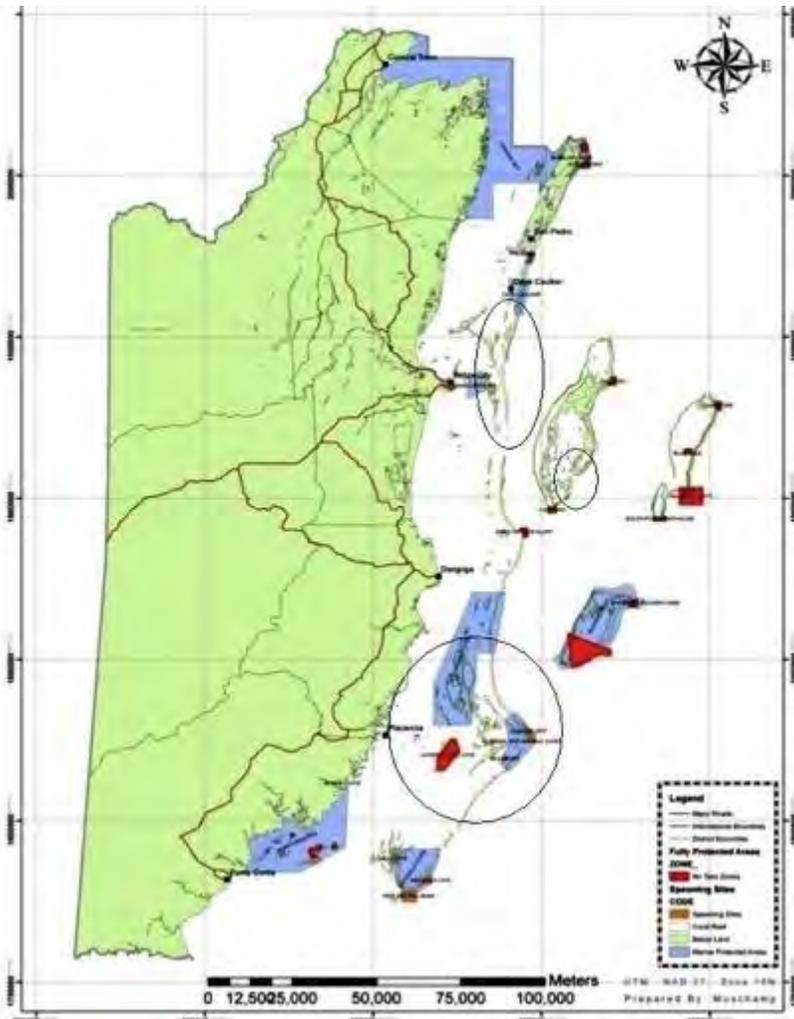
Re-seeding a reef with genetically diverse genotypes of the same species is mandatory if natural spawning is to occur, and sexual reproduction is one of the long-term goals and success indicators for this project.

Methods

"Scoping" mother corals near Placencia to propagate in the nurseries was fairly straightforward as there are so few remaining Acroporid patches left here. Mapping these remaining corals began in 2006 and with the assistance of local fishermen and tour guides many thriving stands have been sampled. In most cases there are loose fragments of opportunity to use, but in some cases the mother colony would be trimmed, never taking more than 10% of the mother coral.

One of the key questions is whether there is a difference between the corals that grow on the outer reef versus the ones in the inner cays. The waters near Placencia are very unique due to the shape of the reef at Gladden Spit-'point of reef' or the 'elbow' as its called locally. The reef juts out in a 90 degree angle and is located ~26 miles from Placencia. The many inner cays behind the reef have calmer, stiller (and thus hotter) water than on the main barrier reef, and so corals were chosen from representative locations similar to Laughing Bird Caye National Park, the out-planting site.

Map of Belize showing marine protected areas and scoping areas.



Southern Belize Sites: Stars represent coral nurseries, yellow dots collecting areas (For scale, Gladden Spit is 60km from Placencia).

Deciding on the nursery locations in Southern Belize was easy too. Only cayes with permanent residents were used (with one exception, the nursery at Gladden). This is to prevent any curious tampering with the nurseries. Nurseries are set up on the leeward side of cayes, and when possible, nestled between or behind large coral heads for protection.

In-situ coral propagation⁶ simply means we are growing the corals in their natural environment (as opposed to a laboratory or aquarium). “Frames” are made from a metal construction fabric and secured with cement blocks as anchors. The corals themselves are attached with plastic cable ties, which they quickly overgrow. This method is excellent for comparing the growth, survival and bleaching patterns between different genotypes.

We have six identical frames with 17 different Acroporid genotypes (see Results, Genetics) in six different locations in Southern Belize. This has allowed us to also compare growth, survival and bleaching patterns between nursery locations.



Gladden nursery Day 0.

⁶ All three *in-situ* coral propagation methods came from Bowden-Kerby, A.

“Tables” support trays with ‘cookies’, and ‘ropes’. The tables are made with steel rebar and the same metal construction fabric as a platform. The trays are made from a durable plastic coated mesh and the ‘cookies’ are made from cement and sand and affixed to the trays with fishing line⁷. These cement cookies allow slower growing coral species to be propagated in the nurseries. Real restoration cannot be effective with only a few species and so eight other stony coral species were included in the nurseries.



Whipray Caye table Day 0.

⁷ The fishing line works fine with Acroporids, but epoxy would be better for the slower growing species.

“Ropes” are strung between the table bars and support prolific three-dimensional growth of the Acroporids. There are two tables with ropes: one at Whipray and one at Laughing Bird Caye.



Whipray Caye ropes Day 0.

Growth data were collected by measuring the coral fragments at Day 0, seven, and 12 months respectively. Three measurements are taken: maximum diameter, the length perpendicular to the maximum diameter, and height or thickness⁸. Absolute growth data is difficult to calculate

⁸ From the AGRRA (Atlantic & Gulf Rapid Reef Assessment) methodology.

for the fast growing Acroporids; you can measure one branch to get linear extension rates but meanwhile it can be growing onto the substrate and in three dimensions. So initial and final volumes were calculated to say how many times the coral expanded from its original size.



Independence high school student measuring 7-month growth.

Bleaching data is collected by observations and photographs. There are also temperature loggers at each nursery site.



Example of photographic , time series monitoring. See more in the Results section.

Out-planting methods

Transport

The most ideal conditions are flat calm days not only so the corals don't get too knocked around on the boat ride, but also because of the underwater cement work. The corals need to be kept in seawater and shaded if possible. In our experience we have transported corals for up to 45 minutes by monitoring and changing the seawater so they remain as little stressed as possible.

When the work started in 2006 we only moved a few pieces at a time, but as the nurseries expanded we needed larger and larger containers to transport the corals to the out-plant site.



Corals stay as least stressed as possible during transport by keeping them in fresh seawater and out of the sun.

Corals need to be planted above the bottom to avoid sedimentation and predators. Old dead coral heads are perfect substrate for planting, but they must be scrubbed clean of algae first. It's easy to identify old dead Elkhorn and Staghorn stands so you know where these species once thrived. Cement is mixed with fresh water on the surface and then used to adhere either the newly trimmed corals, or the cement cookies.



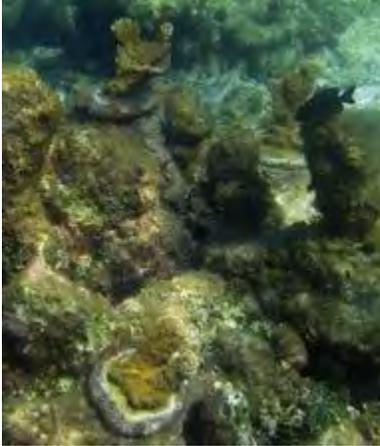
Scrubbing the old dead coral heads free of algae is necessary to allow the cement to adhere to the substrate. We always avoid areas where other benthic organisms live as they might compete with the corals' growth for space (like sponges and zooanthids).



Applying the cement to the substrate. We work in teams, with some people on the surface mixing the cement and passing it to divers in a ziplock bag.



Freshly planted corals in cement.



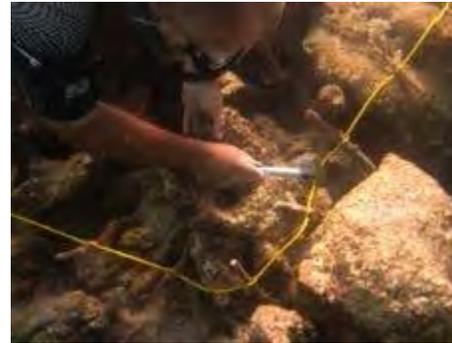
Freshly planted "cookies" with cement.

In some cases fragments can be wedged into place without the use of cement, and if remain secure, will grow onto the substrate in time.



"Plugging" in fragments snugly, without cement.

Freshly trimmed or even newly made ropes can be laid out onto the substrate and nailed into place. As long as there is no slack or play in the rope, the corals will attach themselves to the substrate.



Dr. Austin Bowden-Kerby planting a 'rope' strong with a single genotype of Staghorn coral. See the Results section for how this rope looks one year later.

If the substrate is appropriate and the fragment the right size, plastic cable ties can be used to secure the corals-in the case of the Acroporids only they grow so fast they grow right over the cable tie.



Example of the Acroporids' rapid growth rate over the plastic cable tie. Normally the free end is trimmed as it tends to accumulate algae.

Results to date

Nurseries established

In 2009 six nurseries were established in Southern Belize: six identical frames (the same 17 corals on each frame) and two identical tables with ropes and 80 cookies on trays.

The 17 Acroporids on the six replicate frames have had both the host and symbionts analyzed genetically (see below).

Eight additional stony coral species were included on the tables to verify this propagation method for slower growing (and non-asexually reproducing) corals. These include star corals (*Montastrea annularis*, *M. favelota*, *M. cavernosa*) brain corals (*Diploria strigosa*, *D. clivosa*, and *Colpophyllia natans*), finger coral (*Porities furcata*) and the increasing rare pillar coral (*Dendrogyra cylindrus*)

Three identical (but different from Southern Belize corals) were established on Turneffe atoll using Acroporids from Goffs Caye and Calabash Caye. Although one year growth and survival data were collected, sadly all three frames were moved and turned over in Hurricane Richard (October 2010), yet the corals remain intact and thriving on the top side of the frames.

Two replicate frames were set up in Hol Chan Marine reserve with corals from there and as far south as Caye Caulker.

The results discussed here refer only to the six Southern Belize nursery sites.

Genetics of coral hosts

In 2007 Dr. Iliana Baums' laboratory performed genetic analyses on 24 *A. palmata* samples collected from Gladden Spit (where the 2006 fragments of opportunity came from) using her methodology⁹ which revealed 17 distinct genotypes (of 24), representing relatively high genetic diversity for a reef. Based on these results, we knew we need to house multiple genotypes in the nurseries for effective out-planting that will allow for eventual sexual reproduction.

With funding from the World Wildlife Fund-Central America a different set of 23 Acroporid samples were analyzed by Dr. Iliana Baums' laboratory at Penn State in 2009. Seventeen of the corals are the ones planted on the six frames, and five samples were from some of the Elkhorn fragments of opportunity moved to Laughing Bird Caye

⁹ Baums, et al (2006). Geographic variation in clonal structure in a reef-building Caribbean coral, *Acropora palmate*.

National Park in 2006, and one natural recruit found there. All were found to be distinct genotypes, excellent news!

Genetics of symbionts (zooxanthallae)

In 2009 the same set of 23 corals were sent to Dr. Andrew Baker's laboratory at the University of Miami to have their zooxanthellae Clade identified. Those results showed the inner reef sourced corals had either Clade A or Clade D and the outer-reef sourced corals only had Clade A.

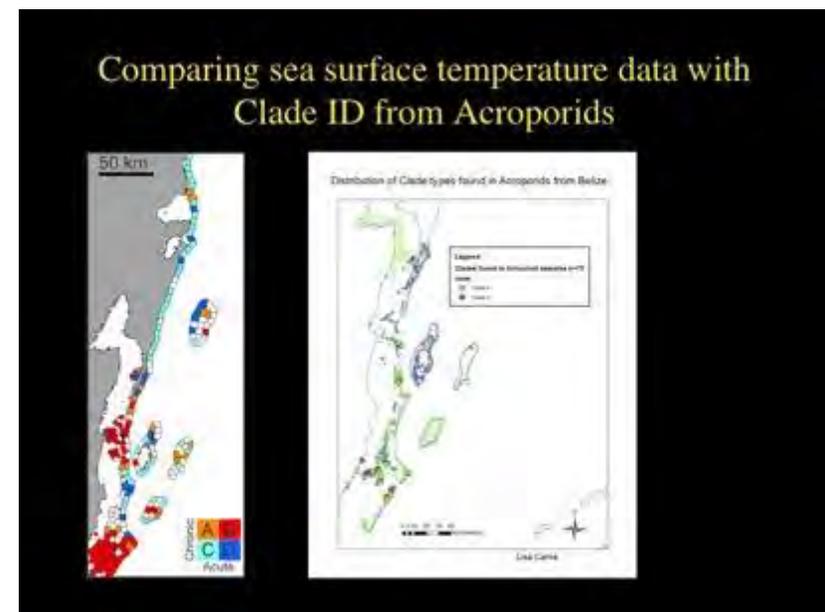
Many researchers feel Clade D lends a coral better resistance to bleaching and temperature changes; did these results support the hypothesis that the inner-reef corals are already somewhat adapted to higher sea temperatures?

We decided analyzing another 50 Acroporid samples from inside and outside the reef might give us a better answer.

In 2010 the World Wildlife Fund-Central America funded the Clade analysis of 50 additional corals, combined with a mapping project to identify the remaining healthy stands of Acroporids on the Main Barrier Reef in Belize. Of these only three had Clade D and two were very close to shore (inner reef) and only one was an outer reef Elkhorn from Pompion Caye in Southern Belize.

In the slide below, the map on the left hand side shows color-coding based on ten years of sea surface temperature (1985-2005, satellite data, from Paris, C. & Mumby, P.). The red indicates the areas with the both the highest recorded

temperatures (acute) and longest sustained high temperatures (chronic). These are the areas then where we would suppose corals are already somewhat thermally adapted and supports both the project's main supposition that Placencia's inner reef corals are more resilient to elevated temperatures, as well the results from the AGRRA surveys in 2006. The map on the right shows the distribution of the two Clade types (A3 and D1) found in 73 different Acroporid samples. With one exception, all of the Clade D's were inside the main barrier reef. These results also support other findings (Baums, 2010) that indicate Clade D is found in areas with the highest temperature fluctuations.



The map on the left is ten years of sea surface temperature data (1995-2005) from Paris and Mumby. The red indicates areas with both the most acute (highest) and chronic (long term) temperatures. The map on the right shows the distribution of Clades identified from 73 Acroporid samples.

Growth rates on the frames

After one year it was clear there are differences in growth rates between nursery locations, between coral genotypes of the same species and between the outer reef and inner reef sourced corals.

Corals grew fastest in the more shallow settings: depths of the top of the six frames ranged from only two feet to nine feet deep. The two pictures of replicate frames at different sites and depths clearly show a difference in growth rates.

The zooxanthellae use sunlight to generate energy which the corals then capture (chemically) for their own use.

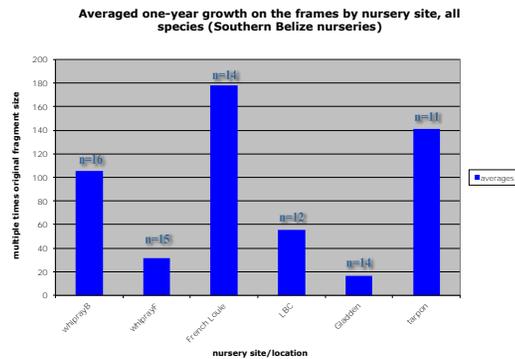


One year's growth on the frame at Gladden Spit (depth is nine feet, the deepest frame with the slowest average growth rate).



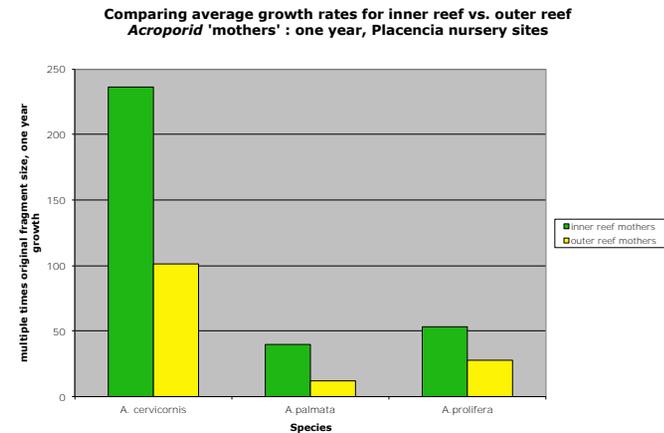
One year's growth rate at the French Louie Caye frame (depth is only two feet with a faster average growth rate).

The graph below illustrates the differences in averaged growth rates on the frames between all six Southern Belize nursery sites. N = the number of corals used to average the growth rate and is slightly different in each site due to breakage; there are three replicates of each coral genotype on the frames but only the top replicate was used for Growth measurements.



When the averaged growth rates were compared from corals sourced from the inner reef versus corals sourced from the outer reef, the inner reef sourced corals grew faster in every location for all three Acroporids.

In the graph below the three Acroporid corals are on the x-axis, with growth rate displayed on the y-axis as the amount of times their three-dimensional volume grew (see Methods). The green represents inner-reef sourced corals and the yellow are out-reef sourced corals.



Bleaching

2006-present, Elkhorn transplants

Time series photography and observations on five *A. palmata* transplants from the outer reef and one natural *A. palmata* colony at LBCNP have revealed different bleaching patterns. 2008 was a fairly serious bleaching event in Southern Belize and all but one of the transplants (12H) bleached fully, and the natural colony did not bleach. All of the transplanted Elkhorn have Clade A3 and only the natural colony has Clade D1. They are placed at similar depths. All the bleached corals recovered with only one colony (12A) exhibiting partial mortality on one branch.

In 2009, only two of the above transplants partially bleached, and four time series samples were collected from the bleached and non-bleaching portion on all six colonies (five transplants, one natural colony) over time (October 2009-June 2010) to have their Clade analyzed¹⁰. The bleached corals recovered fully. The hypothesis was that 'shuffling' of Clades might occur. Many people wonder when the corals bleach and lose their zooxanthellae, when they recover, is it the same Clade type returning? In this case, it was, and there was no evidence of 'shuffling' clades.

In 2010 Southern Belize experienced another fairly serious bleaching event (although not as severe as in 2008). This time the bleaching patterns were different: 12H that had never bleached previously, bleached and recovered in 2010, 1G that had bleached previously (2008) did not bleach, 12A that bleached before (2008 and 2009) bleached again and recovered, but 1B that bleached and recovered (2008 & 2009) sadly bleached in 2010 and did not recover. However it is inconclusive if fragment 1B's mortality (>99%) is due to bleaching, disease, or predation.

Keeping in mind that all of the six colonies are separate genotypes (coral hosts) and all five of the transplanted corals have Clade A3, do these results support the idea that it is the coral host more so than the zooxanthellae clade that lend adaptive resilience to thermal stress? Examples of time series photographs are displayed on the next page. Fragment 1g is shown in the methods section (p. 12).

¹⁰ Rachel Silverstein for Dr. Andrew Baker, University of Miami.



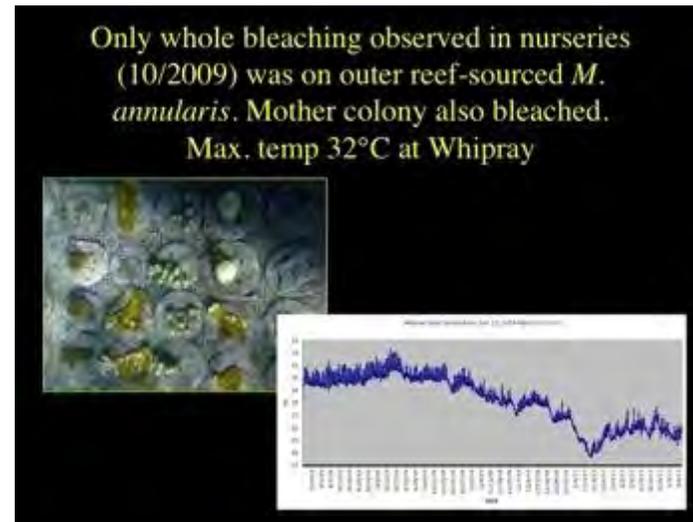
Transplant 12A has Clade A3 and bleached and recovered 2008-2010.



Transplant 12H, also with Clade A3 never did bleach until 2010, and has recovered.

Bleaching in the nurseries-2009

2009 was a relatively minor bleaching year in Southern Belize. Although maximum temperatures reached 32.22°C at the Whipray Caye nursery in September, only the outer-reef sourced *Montastrea annularis* (Boulder Star coral) fully bleached (October 2009), see Figure X. These fragments fully recovered by November and the mother or donor coral was visited at Gladden for bleaching monitoring. Fig X shows the mother coral in the outer reef was still partially bleached in November, ruling out the idea that placing this out-reef sourced coral in a back reef environment caused the bleaching observed in the nursery.



Montastrea annularis fragments sourced from the outer reef fully bleached in the Whipray Caye nursery October 2009 but recovered by November.



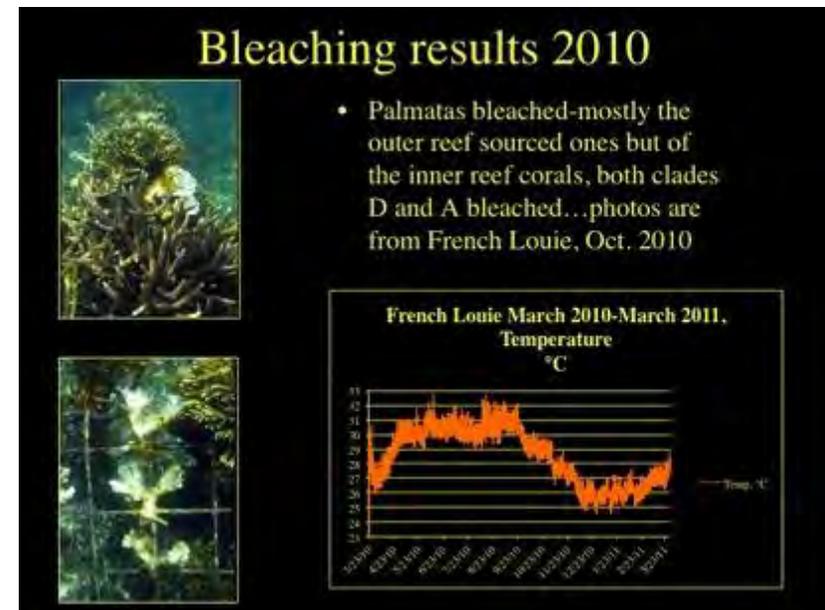
The outer-reef source (Gladden Spit) of the bleached *M. annularis* fragments was visited in November 2009 and was still partially bleached, discounting the notion that relocating the fragments to the inner reef caused the bleaching observed in the nursery.

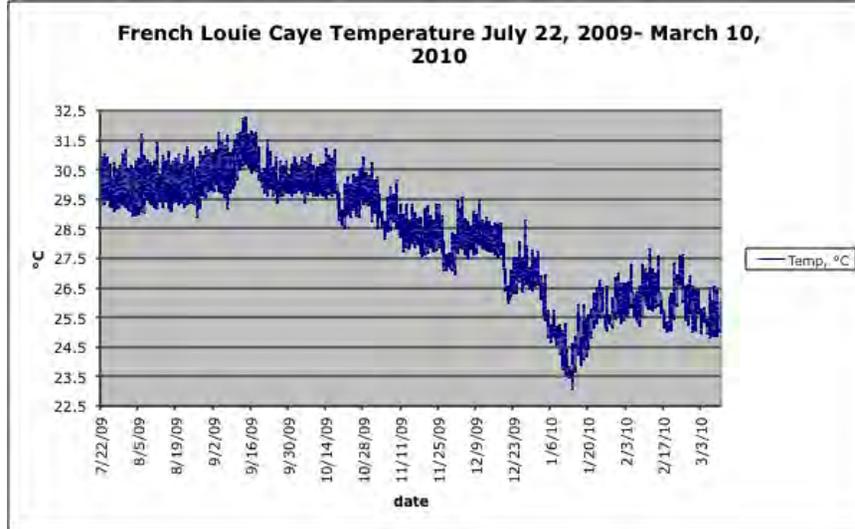
Bleaching in the nurseries-2010

Bleaching was observed on Elkhorn corals in the nurseries in 2010, as well as on the Elkhorn transplants at LBCNP from 2006. While it was primarily the outer-reef sourced Elkhorn corals that bleached, all with Clade A3, some of the inner-reef sourced Elkhorn corals that bleached had Clade A3 and some had Clade D1. In other words, unfortunately there was not a clear pattern of bleaching or not associated with either Clade...

Figure X shows photographs of the frame at French Louie Caye (October 2010). In the top photo the *A. palmata*

bleaching on the right is inner-reef sourced (Larks Caye) with Clade D1. However only the top replicate bleached, the two replicates underneath did not bleach. The bottom photograph is outer-reef sourced (Gladden) *A. palmata* with Clade A3. The temperature graph shows a maximum temperature of 32.71°C in June and again 32.69°C in August. More significantly though, temperatures in 2010 almost consistently remained above 31°C for over three months (June-August), whereas in 2009 temperatures were only above 31°C for a few weeks in September (Fig. X).





Temperature data from the French Louie frame in 2009. Sustained temperatures were much higher (>31°C for three months) in 2010.

Out-plants

Since those first few *A. palmata* transfers in 2006, almost 4000 (~3847) Acroporid fragments have been planted at Laughing Bird Caye National Park in three large scale efforts: February, April and December 2010. Participants included were SEA, Fisheries and the University of Belize staff, Independence High School students, local and international conservation NGO staff, Placencia tour guides and fishermen, and in December, community volunteers (see the Dissemination section for more details).

In November 2009 the Smithsonian, NOAA and Counterpart co-sponsored an Acroporid workshop in Washington DC. Here both experts on asexual and sexual reproduction convened and a document was drafted for best practices. To facilitate sexual reproduction of out-planted, nursery-grown corals it was mutually decided separate genotypes of *A. cervicornis* should be planted a minimum of 10 cm and maximum of 10m apart. Because *A. palmata* grows much larger, recommended distances from each out-plant is 1-10m.

The map below (aerial photograph courtesy Tony Rath) shows the location of out-plants with each out-plant site labeled. Detailed records are kept as to how many of which genotype (and Clade) are planted in each site.



Coral out-plant site map for LBCNP. The yellow X's indicate where *A. palmata* fragments were planted in 2006. The red numbers are

sites planted in February and April 2010. The purple numbers indicate where new out-plants were placed in December 2010 and the purple X's show December's additional out-plants on old sites.



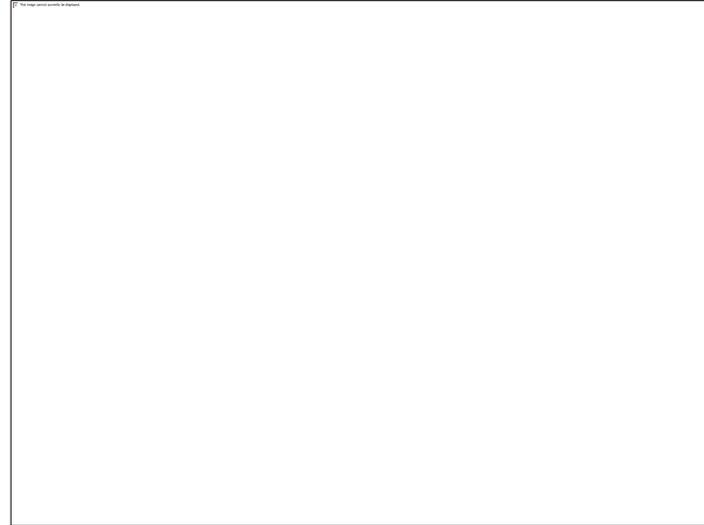
The Laughing Bird Caye table. Divers trimmed ropes, planted those trimmings with cement, planted some ropes, and some of the cookies, and then made new ropes to replace the planted ones.



Trimming the ropes for out-planting.



We had lift bags on hand for the large containers full of trimmed corals.



Trimmed corals from the ropes being planted in cement.



Example of planted corals ~1 year after planting. Notice how they spread onto the substrate, naturally.



Example of planted rope, one year later (see fig X in the Methods Section for how they looked at Day 0).



Gently removing cookies from the table for out-planting. This is ~1.5 years' growth in the nursery.



Elkhorn coral cookies being planted on old dead Elkhorn substrate.

Dissemination, Workshops Conducted and Community Involvement

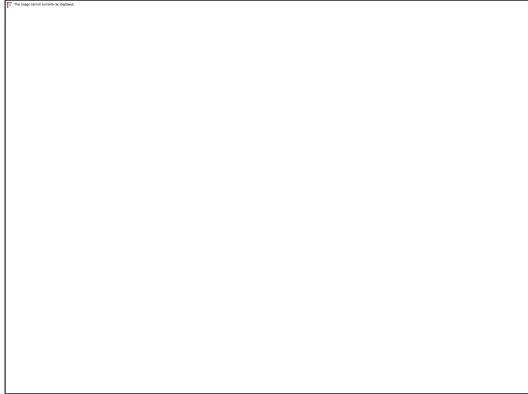
The focus of this work has always been in Placencia and since 2006 there have been six dissemination or out-reach consultations held there, including two hands-on training workshops. The stakeholders in a coastal community that relies on fishing, tourism and ecosystem services from a healthy reef like shoreline protection are every single resident. Participants have included students from the primary school level through university, tour guides, fishermen, marine reserve staff, community members and tourists.

In February 2010 the first hands-on training was conducted over three days with funding from the World Bank and the World Wildlife Fund and included regional participants from the Dominican Republic and Mexico, local marine reserve staff including the Belize Fisheries Department and SEA (Southern Environmental Association) personnel and local tour guides. Participants learned the background on the project and how to make ropes and cookies as well as practicing both nursery maintenance and out-planting methodologies.

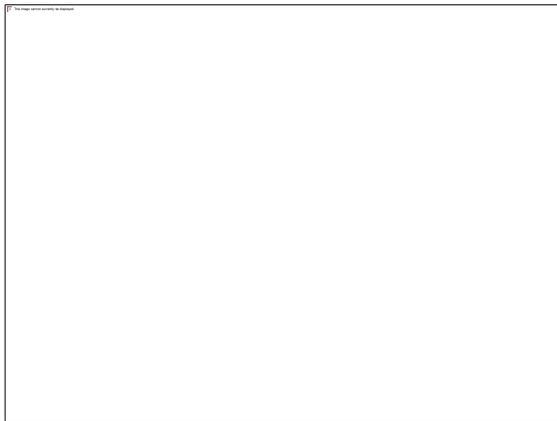


In April 2010 the Independence high school students got involved with funding from COMPACT at SEA's request. Students learned about the entire project and spent two field days making new ropes and out-planting trimmings from the old ropes. Then returned to the nurseries in November 2010 to collect growth data on their ropes, and monitor their out-planted corals for bleaching and survival.

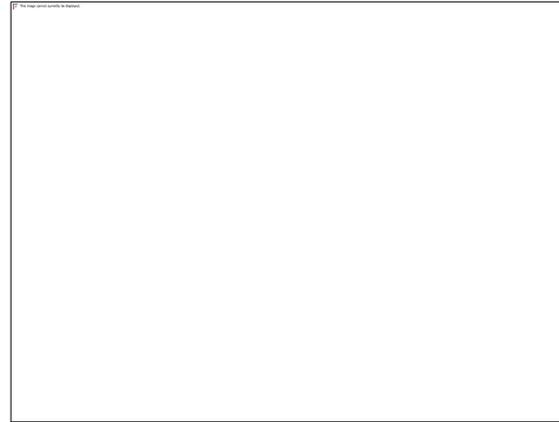




The student group then compiled a report and won first place at SEA's science fair with their life-size diorama of the coral rope nurseries.



In December 2010 another three days of out-planting were conducted using experienced tour guides and fishermen, local conservationists, SEA, UB and Fisheries staff and for the first time volunteers from the dive tourism industry as well as local community members participated. We needed all these hands as there were so many corals to trim and out-plant!



In San Pedro, Belize two demonstration frames were assembled (December 2009) in Hol Chan Marine Reserve with participation from FRAMRACC and CORAL. A local consultation was held with over 40 participants from the tourism and conservation sector.



While these frames were mostly for demonstration of nursery techniques, there was enough coral harvested one year later to demonstrate out-planting techniques to marine reserve staff onto some reef balls¹¹ that were sunk in the 90's and are perfect substrate for planting corals.

¹¹ www.reefball.org. These were sunk on the initiative of the San Pedro Tour Guide Association.



Other regional dissemination has occurred in San Andreas, Columbia through CORALINA with funding from the World Bank. Participants here included conservationists, government biologists and tour guides.



International and local professional meetings where this work has been highlighted include a Reef Restoration Workshop in Puerto Rico sponsored by NOAA (2007), the 11th International Reef Symposium (Florida, 2008), the Smithsonian’s “Success Stories in Marine Conservation Symposium” (Washington DC, 2009), the Pan-Caribbean Reef Restoration Workshop sponsored by USAID (Jamaica, 2010), and four of the five Natural Resources Management Symposiums held by the University of Belize. The project is also highlighted as a success story in Belize’s based Healthy Reefs for Healthy People’s “Report Card for the Mesoamerican Reef” (2010. www.healthyreefs.org).

Lessons Learned/Recommendations-Methods

The frames, while providing a good platform for comparing growth rates and bleaching between genotypes, don’t support the *A. palmata* growth as well as the cookies. Because they grow in a hand or palm shape, they become awkward on the frame and need to be fragmented for out-planting. While Elkhorn can be grown on the ropes, they quickly get heavy, so the cookies are the best method for growing *A. palmata* and are very easy to out-plant.

The ropes are the best method for growing *A. cervicornis* and *A. prolifera* (cookies also supported *A. Prolifera* growth).

Also an important lesson learned was the varying qualities of the construction fabric itself for the frames. While materials purchased in Belize City have lasted over two

years supporting heavy growth, the frames purchased in San Pedro collapsed in less than a year, and not because of the weight of the corals!

For these reasons, future nursery sites should have tables with cookies and ropes, even though these take longer to assemble than the frames. They are stronger and support more coral growth. As noted earlier, only the Acroporids grow fast enough to use fishing line on the cookies; all other species should be fixed to the cookie with epoxy or cement as algae collected at the fishing line.

A variation on the frames used here is a design by “reefscapers”¹² that has a dome shape. Although they plant multiple species on each dome, in Belize we hope instead to try out this shape with a single genotype on each dome. Multiple domes, each with a distinct genotype, could be planted in rubble areas with little remaining substrate, 1-10m apart and left permanently in the hopes that spawning would occur eventually.

Maintenance, Monitoring and Site Selection

Regular (monthly is ideal) monitoring of both nurseries and out-plants is essential. The nurseries need regular algae removal, especially in the hotter months, as even areas with healthy populations of grazing fish can quickly be taken over by algae. Regular monitoring also allows for predator removal, observations of disease or bleaching, collection of

¹² <http://reefscapers.com>

growth data and any repairs necessary (to the structures and/or rescuing broken fragments).

When choosing a nursery location these factors must be kept in mind, as fuel is expensive, the site must be easily accessible as well as protected from storms. In southern Belize the leeward side of inhabited cayes has proven most effective. Nurseries in areas without permanent residents have been tampered with (temperature loggers stolen).

The out-planted corals also need regular monitoring, especially initially, to make sure they don't become dislodged, and begin to grow onto the substrate themselves. Unless restoring a specific damaged site (ship grounding or anchor damage), it is recommended to out-plant corals in No-Take Zones, where no fishing is allowed, for reasons outlined in the next section.

Lessons Learned-Predation and Disease

The snail *Coralliophila abbreviata* is a known coral predator and they seem to prefer Acroporid corals although they also eat the Lettuce Leaf (*Agaricia tenuifolia*) and the Boulder Star Coral (*Montastraea annularis*). Their abundance is thought to be controlled by the Caribbean Spiny lobster, who eats them, and this may explain why lobsters love the Acroporid corals (especially Elkhorn): the corals provide shelter and foraging grounds. In areas where lobsters have been over-harvested, snail populations are higher. With so few Acroporids left predation can be a serious threat to survival.

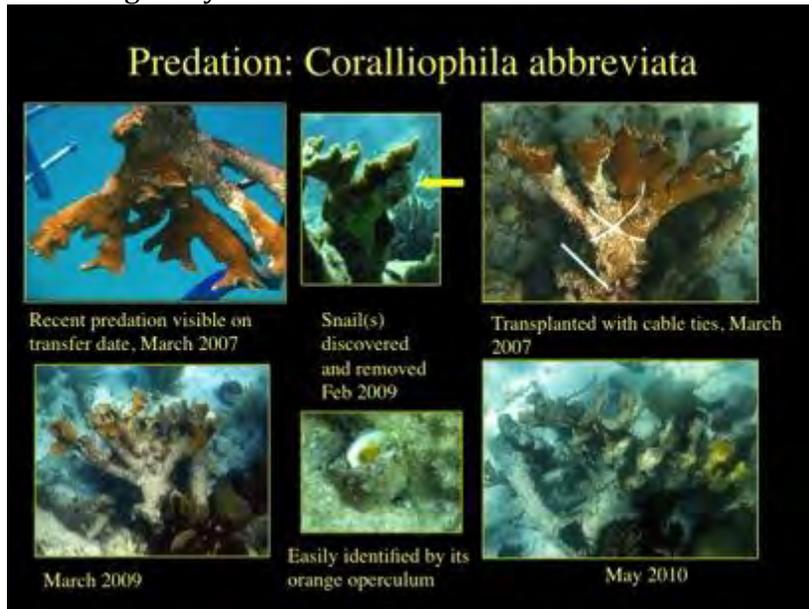
Although forewarned and aware of these predators, they are very cryptic (camouflage well by looking like dead reef) and often hide during the day and feed at night. So they are easiest to find by training your eye to look for fresh predation scars on the coral tissue, and then hunting for the snails to remove them. These snails can be positively identified by their bright orange operculum when you turn them over (see Fig X).

In 2007 there was an offer by the German organization Aldebaran (www.aldebaran.org) to film the first transplant project for free, but we only had one day to do it. So although each initial fragment of opportunity was carefully examined for any sign of disease or predation (2006), the large piece of Elkhorn coral selected for the filming was transferred too hastily and after two years of monitoring mortality (March 2007-February 2009) snails were finally found and removed. However, snails are thought to transfer disease, so either this was the case and/or not every snail was removed because this coral fragment eventually died.

Fig X shows the fragment the day of transfer, with fresh predations scars visible. After monitoring tissue loss for almost two years (!), finally snails were discovered and removed.

In other cases, when the snail(s) were discovered immediately and removed, the lesions healed and the coral survived. The lessons learned here are many: don't let filmmakers rush you or dictate the pace of your work (re-

learned in 2010)! And carefully inspect all your transfer fragments and continue to monitor all your out-planted corals regularly.



Predation by the snail *Coralliophilla abbreviata* documented over two years.

While *C. abbreviata* is a known coral predator, thought to be kept in check by lobsters, two other undocumented coral predators were discovered on the out-plants and in the nurseries.

In 2009 the West Indian topsnail (*Cittarium pica*), and locally known as “wiliks”, was found on an elkhorn transplant at LBCNP. Although the snail was removed the coral later appeared to suffer from disease, became

weakened, and crumbled in TS Mathew (September 2010). A remnant piece remains and monitoring continues.



The West Indian Topsnail (*Cittarium pica*) found on one of the Elkhorn coral transplants.

In April 2010 a strange disease was noted on natural Elkhorn colonies at Larks Caye during a routine bleaching survey. It didn't appear as 'normal' white band and no predators were observed. During the same month out-planting (*A. cervicornis*) was conducted with the Independence High School students and at just one of the sites, abnormally high mortality was observed just two weeks after the planting. Many of these out-plants were planted right on the bottom (supervised by an assistant) when normally we plant on old dead coral heads above the

substrate, so initially predation was assumed to be the cause of mortality. However by June this ‘affliction’ was apparent in the Whipray Caye nursery, affecting both the *A. plamata* and the *A. cervicornis* but not the hybrid.



Unknown affliction-recent mortality-first observed on natural Elkhorn colonies at Larks Caye in April 2010.



Same pattern observed in the Whipray Caye nursery June 2010.



Strange mortality on *A. cervicornis* in the nurseries, note the ragged edges at the tissue loss border.

Through discussion with several colleagues a decision was made to make a night assessment for predators; could the nursery (s) be plagued by nocturnal fireworms (*Hermodice carunculata*), another well-known coral predator?

The night observations revealed zero evidence of any predators, so again upon consultation with colleagues a decision was made to try to ‘cull’ or removed the affected corals, in hopes of preventing total mortality in the nurseries. During these culling sessions careful

observations were made in the nurseries. Austin Bowden-Kerby suggested the Four-eye butterfly (*Chaetodon capistratus*) as a possible culprit based on his observations in other nurseries; these fish are known to predate on some soft corals gametes, could it be the corals in the nurseries were producing gametes and attracting these predators?

Damselfish, another plague for Acroproids because they build an algae nest in these corals which can cause mortality, were observed on both the *A. cervicornis* and the *A. palmata* but when strong surge caused long pauses between culling, the four-eye butterfly fish made appearances.



The damselfish seem equally attracted to both Acropora species.



An intermediate-stage cocoa damselfish (*Stegastes variabilis*) exhibiting territorial behavior near afflicted *A. cervicornis* in the Whipray nursery (June 2010).



The four-eye butterfly fish (*Chaetodon capistratus*) caught in action, picking at the *A. cervicornis*.



Are the butterfly fish the cause of the recent mortality? Are they exposing the corals to disease by their predation?

Although it was clear the butterfly fish were picking at the corals' tissue, it is not clear if they caused the rapid spread of mortality in multiple nursery sites and also on natural corals. Researchers in Florida had observed the ragged, uneven tissue loss in previous years, which they call simply "Rapid Tissue Loss" but no one had previously documented the butterfly fish predating on Acroporids. Samples of afflicted corals have been sent to Dr. Steve Vollmer at Northwestern University, who is an expert on white band disease, and also to Dr. Esther Peters at George Mason University, a leading histologist and expert on coral diseases. Their lab results are still pending.

The culling itself had mixed results; healthy looking fragments re-attached from afflicted colonies in most cases died, but the rapid spread of mortality did seem to be

halted, whether by this intervention or timing, it is not clear.

Lessons Learned-Growth Rates

Differences were observed between corals sourced from the outer reef versus the inner reef, suggesting that the inner reef corals are already somewhat adapted to higher temperatures.

Differences were observed between genotypes of the same coral species, giving credence to the concept of 'eco-types' borrowed from botany, meaning micro-environmental factors can influence corals' growth rates.

Climate Change, Other Impacts and Personal Choices

Despite the science and data available, it seems most Americans do not 'believe' in climate change, as if it is a religion or philosophy. In a recent article in the LA Times (<http://latimesblogs.latimes.com/greenspace/2011/05/climate-change-arctic-ice-melting-faster-sea-level-to-rise-more-report-says.html>) researchers found that glaciers are melting even faster than previously predicted, and estimated that 7700km³ of fresh water have been added to the oceans in recent years.

Extra fresh water into the ocean's salt water changes densities that drive ocean currents, which directly affect weather patterns. Every North American in recent years has experienced colder winters and has thus questioned

“global warming” when in fact, global temperatures in 2005-2010 were the highest ever recorded since 1880 (when records began). Please see the article from NASA at this link for more information on how ‘global warming’ can cause colder winters in North America and Northern Europe: http://science.nasa.gov/science-news/science-at-nasa/2004/05mar_arctic/.

Included here are just a few slides borrowed from NOAA’s ¹³presentation on reef resilience.

What is the root cause?

Carbon dioxide, methane, and nitrous oxide are increasing in our atmosphere.

The CO₂ increase comes from burning fossil fuels; the methane and N₂O increases are mostly from agriculture.

These are known as “greenhouse gases”

Trap some of the heat from the sun so it can’t leave the atmosphere

How the Earth's Greenhouse Effect Works

Incoming Radiation

Outgoing Radiation

Reflected

Absorbed by Greenhouse Gases

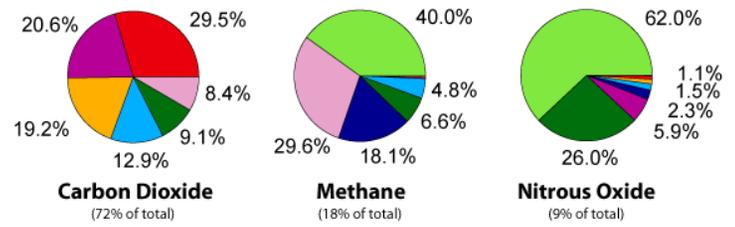
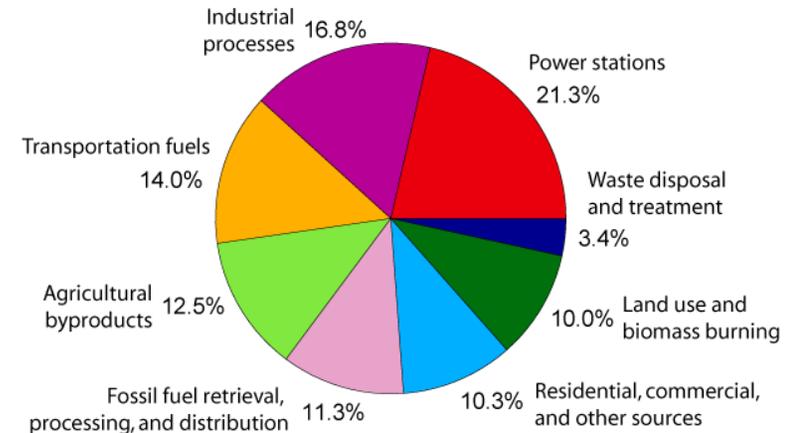
+ 59° F

Average Temperature

CG Page 16

Causes of global warming (climate change) explained. Of course there are multiple other factors accelerating the effect of emissions, like the loss of natural carbon sinks from deforestation.

Annual Greenhouse Gas Emissions by Sector



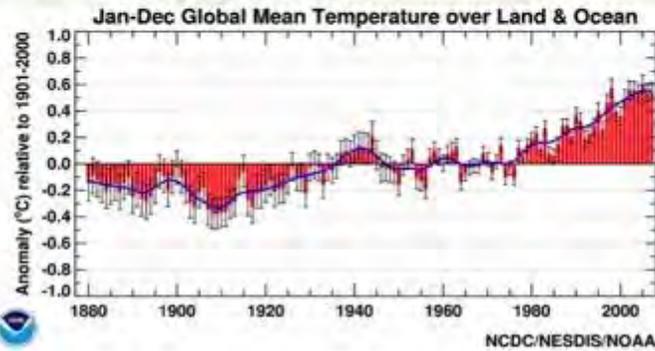
This chart shows the sources of emissions by sector from 2000 (http://en.wikipedia.org/wiki/File:Greenhouse_Gas_by_Sector.png). While it is true that countries like America, China and India have the most emissions, in a 2010 online article in Nature (<http://www.nature.com/news/2010/100505/full/465018a/box/1.html>) researchers report developed countries may not be accurately reporting all of their greenhouse gas emissions.

¹³ National Oceanic and Atmospheric Administration (USA).

Is climate change real?

The greenhouse effect has led to a steady rise in global temperatures.

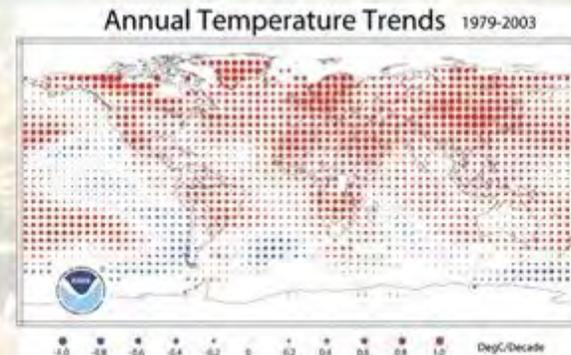
125 years of direct measurements.



In the graph shown above, what is shown is air temperature anomalies which means changes from the normal, or average temperature expected. The averages were based on temperatures recorded 1901-2000.

Is climate change real?

Ocean temperatures have increased almost everywhere, as deep as 3000 m.

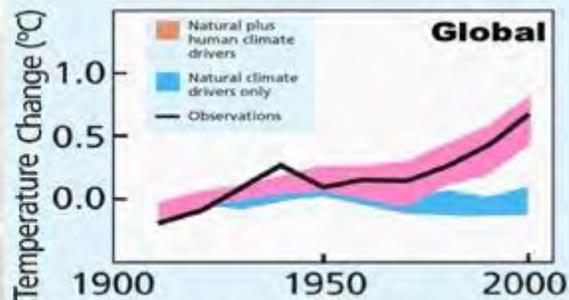


This slide depicts sea surface temperature (from satellite data) anomalies (changes from the normal temperature) from 1979-2003. The larger the red dot, the greater the temperature increased from the average.

Is it caused by humans?

Computer models show that global warming can not be explained by natural cycles alone.

We can say with very high confidence that most of the warming is caused by the greenhouse gases that humans have released.

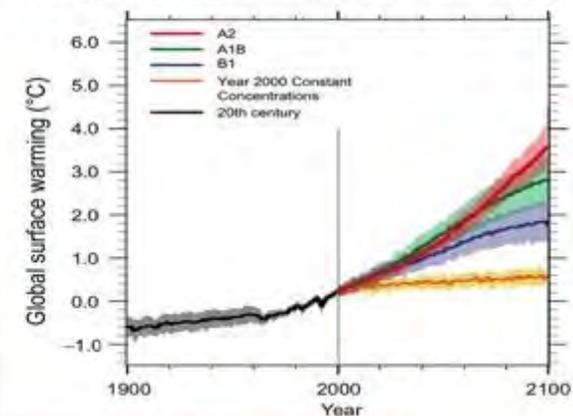


Because some argue that changes we are observing and documenting are part of a 'natural' cycle, NOAA developed this model where they extracted natural causes of greenhouse gases from human related sources. The black line is observed changes in temperature from the normal and the blue line is what would be expected without our emissions (very little change), and the pink line includes our emissions' effects, which matches observed changes.

What will the future bring?

Temperature

Averages and Assessed Ranges for Surface Warming



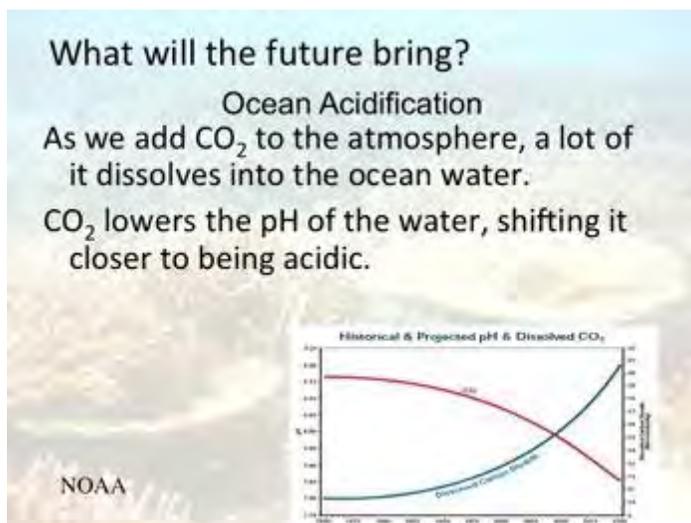
Caribbean has already gone up 1.1°C in the last 100 years., Dr. Leslie, 5C's.

In this slide NOAA offers several predictions for the future. The bottom, yellow line, is temperature predictions if global emissions remained at the same level as 2000 and the top, red line is if we stay with our current habits, increasing emissions every year, and make no changes. The blue and green lines are best-case scenarios if emissions are reduced. Dr. Kenrick Leslie, Executive Director for the Caribbean Community Climate Change Centre based in Belmopan, Belize has stated that temperatures in the Caribbean have already increased by 1.1°C in the last 100 years.

Ocean Acidification

Some call it the "other CO₂ problem" but of course it is the same atmospheric problem causing "climate change". The oceans absorb CO₂ from the atmosphere, which is good for us, but bad for marine organisms. The problem is the excess amount of CO₂: some sources say the ocean has already absorbed as much as four tons! This is changing the

chemistry of the ocean itself. In a nutshell, absorbed CO₂ lowers the pH of the ocean making it more acidic. This directly effects all marine organisms that use calcium carbonate for their structures or shells. It makes it harder to form and can effect what has already been formed. Corals use calcium carbonate to build reefs, conchs to make their shells, and countless microscopic plants and animals need calcium carbonate for their structures. The predictions are not good; many models suggest that by 2050 marine life as we know it will cease to exist. NOAA has a short explanatory video on YouTube (<http://www.youtube.com/watch?v=xuttOKcTPQs>).



NOAA's graph shows the relationship to date with dissolved CO₂ in the oceans and includes a predictive model for the future.

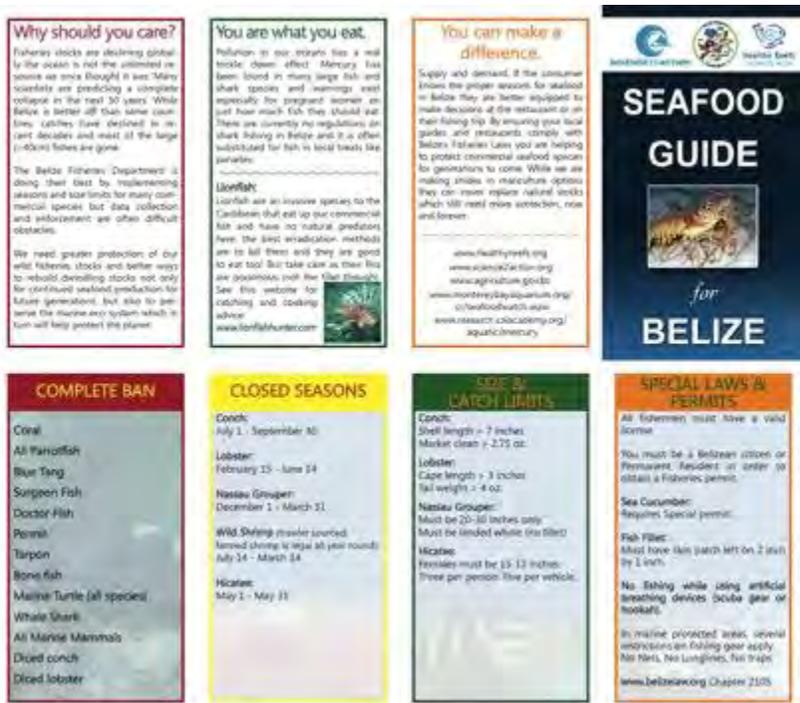
Perhaps it is because the whole concept of climate change is so overwhelming for most people, and our dependency on fossil fuels is married to our lifestyles, that many people

choose to ignore the facts. Here in Belize, most people recognize the reality of climate change, but know it is also true that Belize's (nor any other Caribbean nor Central American nation) emissions are significant. Without America, China and India curbing their emissions (and according to many, even if they did, it may be too late to slow down much less reverse the effects) personal attempts at reducing energy use might seem futile.

However, every aspect of our daily lives and the choices we make are directly related to earth's ecosystems, including the health of our reefs. While the discussions should really focus on alternative energy solutions, (instead of the current battles in Belize over whether or not to allow off-shore drilling), what we choose to eat and the products we buy also have long-reaching effects.

Seafood globally is being over-harvested and besides worrying about mercury contamination, sustainable choices can be difficult. The Monterey Bay Aquarium established pocket seafood watch guides for North Americans that can be downloaded from their website (<http://www.montereybayaquarium.org/cr/seafoodwatch.aspx>) or even as apps from Apple, and they have a Facebook page. We like this idea so much we made a local version for Belize also available for downloading at www.science2action.org/files/s2a/seafoodguideforbelize.pdf.¹⁴

¹⁴ Funded by Conservation International's MMAS Science2Action program.



The Take-Home Message and Next Steps

While it is impossible to restore all of the degraded reefs globally or even regionally, it is possible to accelerate natural coral recovery in select sites. The chosen restoration sites, in this case Laughing Bird Caye National

Park, should have importance to the local economy and ecology.

Laughing Bird Caye National Park receives over 10,000 visitors a year, and marine tourism is vital to the social economics of Southern Belize.

Acroporids play a crucial role in the reef ecosystem, providing habitat for multiple other marine species including the commercially important Caribbean Spiny lobster. Because Laughing Bird Caye National Park is a properly enforced No-Take Zone it is the ideal choice site for reef enhancement.

Acroporids alone cannot complete an impacted coral reef system, and trials with propagating multiple other stony coral species will continue, with an added focus on *Agaricia tenuifolia* (Lettuce leaf coral), observed to be one of the faster recovering species after impacts, and an important reef builder in Southern Belize.

While individuals may not do much to lesson larger impacts like climate change effects on the marine ecosystem as a whole, there are many small steps divers can make to maintain select reef sites. To this end we are developing a Reef First Aid/Coral Care certification course that will train participants on how to remove smothering sand from corals, remove select harmful algae species, and rescue broken and dislodged corals.

With support from the Belize Fisheries Department we hope to begin this process with local marine reserve staff, local tour guides, and one day offer this course to visiting marine tourists as well so that we may maintain as many healthy reef sites as possible in the face of continued threats and impacts to coral reefs.

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Acknowledgements

Funders include PACT (Protected Areas Conservation Trust). Project AWARE, the World Bank, the Caribbean Community Climate Change Centre, the World Wildlife Fund-Central America, with some support from Healthy Reefs Initiative.

In-kind contribution and support has come from The Placencia Tour Guide Association, SEA (Southern Environmental Association), The Inn at Robert's Grove, David Alveraz (owner of Ranguana Caye), Beverly and Julian Cabral (owners of Whipray Caye), Kitty Fox and Rannie Villanueva (owners French Louie Caye), Charles Leslie, Sr. (owner Tarpon Caye), Seahorse Dive Shop, Avadon Dive Shop, Ramon's Dive Shop, Independence High School, and the Hol Chan Marine Reserve.

Harold Hudson (retired NOAA reef restoration biologist) and Austin Bowden-Kerby, PhD, provided methodologies and inspiration.

None of this work would be possible without the continued support and endorsement of the Belize Fisheries Department.