



Assisted Coral Reproduction in the Dominican Republic: A Successful Story to Replicate in the Caribbean

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Sellares-Blasco RI, Villalpando MF, Guendulain-García SD and Croquer A (2021) Assisted Coral Reproduction in the Dominican Republic: A Successful Story to Replicate in the Caribbean. Front. Mar. Sci. 8:669505. doi: 10.3389/fmars.2021.669505 Coral assisted fertilization, larval rearing and recruit propagation success in significant ecological scales, largely depend on scaling up and replicating these efforts in as many regions as possible. The Dominican Foundation for Marine Studies (FUNDEMAR) has become a pioneer of these efforts in the Dominican Republic, being the first institution to successfully implement coral sexual reproduction techniques in the country and establishing the first mobile larvae culturing facility. Here we share our perspective on three main components behind the success of FUNDEMAR's program: (1) a selfsustainable program in alliance with local and international organizations, (2) the design and construction of the first Coral Assisted Reproduction Laboratory in the country, and a (3) clearly defined scalable structure for outcome performance. Two years after program implementation, FUNDEMAR has successfully produced an annual regional coral spawning prediction calendar, cultured seven coral species, and seeded over 4,500 substrates with more than 268,200 sexual coral recruits in approximately 1,880 m² reef areas. Here, we provide a detailed description of a fully functional assisted coral reproduction program, including the lessons learned during its implementation as well as a series of specific solutions. We hope this work will help and inspire other countries and small institutions to replicate FUNDEMAR's coral assisted reproduction program components and contribute to the expansion of sexual coral restoration efforts in the Caribbean.

Keywords: coral restoration, coral reefs, sexual propagation, coral rearing, science-based solutions, Dominican Republic

INTRODUCTION

During the past few years, coral restoration efforts have increased globally (Boström-Einarsson et al., 2020) and across many Caribbean and Latin-American countries (Bayraktarov et al., 2020). Coral restoration has been proposed as a solution to decrease and/or ameliorate the impacts of local and global stressors as a useful tool to preserve reefs, recover populations of reef building corals or both (Calle-Triviño et al., 2018; Bayraktarov et al., 2020; Boström-Einarsson et al., 2020; Goergen et al., 2020; Shaver et al., 2020).

Coral restoration can be done through asexual and sexual propagation and other types of interventions to enhance substrate suitability for outplanted or gardened corals (Boström-Einarsson et al., 2020). While asexual fragmentation can rapidly increase coral tissue coverage, it does not directly increase genetic diversity in coral populations, making threatened populations potentially vulnerable to diseases and environmental stress (Barton et al., 2015). On the other hand, sexual propagation may increase genetic diversity and therefore resilience, however, low recruit survival after seeding to a reef is still a challenge for restoration practitioners (Baums et al., 2019). Regardless of the method or the approach used, restoration programs still face challenges often associated with the problem of scaling up efforts in space and time while preserving genetic diversity of wild coral populations (Boström-Einarsson et al., 2020).

Recent advances on larval enhancement techniques wouldn't have been possible without the cumulative general knowledge of coral biology and ecology (Guest, 2010; Guest et al., 2014). Reproduction in corals consists of a sequence of events which include gametogenesis, spawning (for spawning species), fertilization, embryogenesis, planulation, dispersal, settlement, and recruitment (e.g., Harrison and Wallace, 1990; Baird et al., 2009; Harrison, 2011). These events have been described from different perspectives, including histological (e.g., Duerden, 1902; Fadlallah, 1983; Szmant, 1986; Richmond and Hunter, 1990; Soong, 1991; Steiner, 1998; Morales, 2006; Ritson-Williams et al., 2009; Weil and Vargas, 2010; Harrison, 2011; Soto and Weil, 2016), observational (e.g., Vermeij et al., 2003; Levitan et al., 2004; Van Woesik et al., 2006; Vize, 2006; Bastidas et al., 2012; Chamberland et al., 2016; Keith et al., 2016; Fogarty and Marhaver, 2019), and experimental (e.g., Morse et al., 1988; Webster et al., 2004; Kuffner et al., 2006, 2007; Nugues and Szmant, 2006; Vermeij et al., 2009; Ritson-Williams et al., 2010; Marhaver et al., 2015; Sharp et al., 2015) studies.

Early attempts to incorporate the concept of larval propagation for restoration purposes started two decades ago (Rinkevich, 1995; Petersen and Tollrian, 2001). In the Caribbean, first successful attempts to rear larvae in the laboratory were conducted by Szmant and Miller (2006) in the Florida Keys and by Randall and Szmant (2009) in Puerto Rico. Currently, Coralium lab at the National Autonomous University of Mexico, SECORE International and the Caribbean Research and Management of Biodiversity (CARMABI) are three leading institutions in the Caribbean on this subject. These institutions have played an important role in expanding larval propagation across geographies, improving capacity building, and developing cutting edge technology and protocols applied for coral restoration (e.g., Marhaver et al., 2015; Chamberland et al., 2016, 2017; Banaszak pers. comm.).

The Dominican Foundation for Marine Studies (FUNDEMAR) has been able to locally adapt these techniques integrating an assisted sexual coral reproduction program, the first one in the Dominican Republic (Calle-Triviño et al., 2018). Here, we provide our perspective explaining the success of FUNDEMAR's program: (1) a self-sustainable program in alliance with local and international organizations, (2) the design and construction of the first Coral Assisted Reproduction

Laboratory in the country, and (3) a clearly defined scalable structure for outcome performance.

KEY COMPONENTS BEHIND THE SUCCESS OF FUNDEMAR'S PROGRAM

FUNDEMAR's Self-Sustainable Coral Restoration Program

FUNDEMAR was founded in 1991 with the mission to preserve marine ecosystems in the Dominican Republic. It is based in Bayahibe, a small town of fishermen which is also a hotspot for tourism activities in the country (**Supplementary Figure 1**). The early strategic alliance between FUNDEMAR and the tourism private sector was a key step for achieving a sustainable program. The first couple of alliances in our coral conservation program motivated and prompted other local stakeholders (hotels, resorts, dive centers, and the community) to get involved. In time, the stakeholders themselves became emotionally engaged with FUNDEMAR to preserve coral reefs resilience for its intrinsic value and for its services to local people.

This structure was consolidated not only with the support of the private sector (economically and in kind) but also by FUNDEMAR's initiative to create permanent income mechanisms (such as hosting educational programs with international students amongst other fundraising strategies). As a result, since 2012 to date, FUNDEMAR has gradually scaled up the program as it was becoming more sustainable. The coral program relies on four pillars: (1) research and monitoring, (2) asexual (Calle-Triviño et al., 2020) and sexual restoration (Calle-Triviño et al., 2018; **Supplementary Figure 2**), (3) management, and (4) community integration and awareness. FUNDEMAR's coral restoration program (**Supplementary Figures 1,2**) is only one of a series of interconnected marine conservation programs that complement each other for preserving coastal ecosystems within the Southeastern Reefs Marine Sanctuary.

In 2017, FUNDEMAR joined SECORE's capacity building program as an implementation partner. Since, SECORE has provided restoration technology and training on larval prorogation to FUNDEMAR staff. This alliance and the partnership with The Nature Conservancy's coral strategy, placed the Dominican Republic as a key location to scale up assisted fertilization and larval propagation efforts in the Caribbean, integrating SECORE's unique Coral Rearing *In Situ* Basins (CRIBs, SECORE International, 2020; **Supplementary Figures 2G,H**) and substrate designs (Chamberland et al., 2017; **Supplementary Figure 3**), amongst other technologies.

Design and Construction of the First Coral Assisted Reproduction Laboratory in the Dominican Republic

In 2019, FUNDEMAR established a mobile *ex situ* coral assisted reproduction laboratory by adapting a storage container ($12 \text{ m} \times 2.44 \text{ m} \times 2.6 \text{ m}$ L, W, H) into a fully functioning laboratory for rearing corals (**Supplementary Figures 2E,F**). Inspired by the experience and knowledge gathered from

trainings in Curaçao by SECORE and in Mexico by CORALIUM and SECORE, various FUNDEMAR partners helped design and build this facility for its intended goal. The main adaptations involved lining the walls with a thermal insulator, running electrical lines, plumbing for fresh and sea water flow and installing fans and air conditioning for temperature regulation. The wet lab itself consists of three major components: (1) a water catchment system, (2) a filtration system, and (3) an aquarium system. An in-depth description of the laboratory including design and system functioning is provided in the **Supplementary Material (Supplementary Figure 4)**.

Furthermore, the CRIB technology allowed us to escalate the production of coral settlers. CRIBs consist of three main parts: (1) a floatable hydrodynamic ring that maintains the structure and is anchored to the bottom, (2) a canopy located above the floatable ring that protects the embryos from ultraviolet radiation as well as fresh water in case of rain, and (3) an underwater vertical enclosure with removable mesh windows (100 and 200 μ m) to allow continuous water exchange (**Supplementary Figures 2G,H**).

Clearly Defined Scalable Structure for Outcome Performance

FUNDEMAR's program structure has been capable of scaling up gradually based on clear annual goals defined in the institution's restoration plan, which is annually evaluated. During the first 2 years of program implementation (2019–2020), efforts were mainly focused on the standardization of coral sexual culturing techniques as well as building capacity for FUNDEMAR staff and other coral restoration practitioners. Yet, we were able to scale up and enhance outcomes from 1 year to the next (presented below). We also focused on gathering enough spawning documentations to create our own coral spawning prediction calendar. The first calendar was available in 2019 and it is updated every year to increase reliability in gamete collection locally.

In Southeastern Dominican Republic, most massive coral spawning events (>25% of colonies spawning) occurred in May and June for *Diploria labyrinthiformis*, August for *Acropora palmata*, *A. cervicornis* and *Dendrogyra cylindrus*, and September for *Colpophyllia natans*, *Orbicella annularis* and *O. faveolata*. *D. labyrinthiformis* had the earliest spawning events while the acroporids and *Orbicella* spp. spawned the latest. For most species, a few colonies spawned a little the day before their massive spawning event. Spawning patterns were less stable through time and had longer windows for acroporids compared to other species (**Supplementary Table 1**).

From 2019 to 2020, gametes were collected, eggs fertilized, embryos cultivated, larvae settled, and recruits seeded from five different coral species: *D. labyrinthiformis*, *D. cylindrus* (Villalpando et al., 2021), *A. palmata*, *A. cervicornis*, and *C. natans* (Figure 1, Table 1, and Supplementary Table 2). In 2020, gametes from *O. faveolata* and *O. annularis* were also collected and successfully fertilized, however, the larvae culture died during the swimming stage in both *in situ* and *ex situ* cultures for unknown reasons.

The implementation of both *in situ* and *ex situ* culturing systems allowed for a high production of substrates with coral sexual recruits or seeding units (SUs: *sensu* Guendulain-Garcia

et al., 2016; Chamberland et al., 2017). In 2019, 1,927 SUs with 28,900 coral settlers were seeded in 850 m² of reef. In comparison, in 2020, 2,615 SUs with 239,300 settlers were seeded in 1,030 m² of reef. In total for both years, over 4,500 SUs with approximately 268,200 sexual coral recruits were seeded in reef areas totaling around 1,880 m² (**Supplementary Table 2**). These results were achieved with systems below their maximum capacity (i.e., *ex situ* system 1,000 substrates and CRIB ~2,000).

In the middle to long term, we expect to produce a database to test specific hypotheses regarding which species and substrate type show higher recruit survival and which are more costeffective to reproduce.

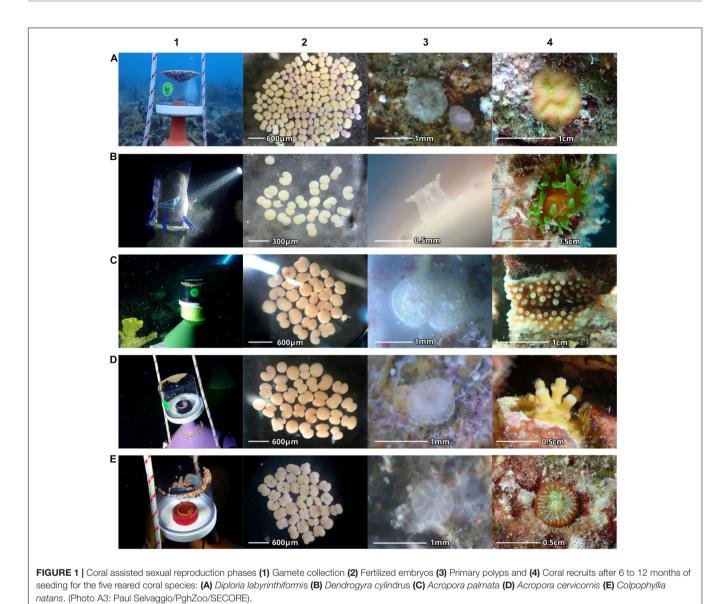
DISCUSSION

In this paper we provide our perspective about a series of guidelines that are needed to implement a sexual propagation restoration program through assisted coral reproduction. We find these 3 points to be essential to create a robust and scalable program: (1) alliances with private and local stakeholders, (2) financial stability, and (3) adoption of novel technology supported by pertinent training to implement them. In our view, the balance between these components allowed FUNDEMAR to build the laboratory and produce results comparable to others in the Caribbean (Chamberland et al., 2016, 2017). Local engagement and key alliances between different stakeholders and the scientific community has been shown to be a strong pillar that supports conservation actions aimed to preserve coastal marine ecosystems (White and Vogt, 2000; Lundquist and Granek, 2005; Reyes-García et al., 2019).

In our perspective, the most valuable lessons learned during the implementation of our sexual propagation program are logistical, technical, and structural. These lessons are often interconnected and must be taken into account holistically as the restoration program is implemented.

From the logistic point of view, getting permits on time is a key step. FUNDEMAR has been able to obtain environmental permits to implement these actions, based on our solid reputation of contributing and supporting the national strategy for coral conservation in the Dominican Republic. Environmental hazards and catastrophes such as hurricanes may impose logistical problems, and in some cases, delay restoration efforts. The best way to cope with the problem is to create an action plan that can be carried out in various seasons and includes more than one species for collection and fertilization as well as diverse cultivation methods to reduce vulnerability to climatic, environmental or unforeseen events, always having a backup plan.

The combination of differing culturing facilities and settlement substrates designs was crucial to increase the production and upscaling the restoration effort. This allowed us to seed a substantial amount of SUs in larger reef areas. The use of the CRIBs represents a feasible strategy for mass production of SUs, nevertheless it is important to consider the local environmental conditions with this method. This type of system works well in relatively calm waters and having an emergency plan for adverse weather conditions is fundamental; in 2019 and 2020, we removed the CRIBs as a hurricane prevention



measure, and sheltered the substrates in the laboratory as well as It is critical to

in underwater structures used for preconditioning substrates. As for the technical lessons, initial time investment in monitoring local spawning events is essential to accurately forecast them, reducing costs in the long term by making time in the field more efficient. This in turn leads to better working plans and therefore a reduction of potential human errors that may compromise the success of gamete collection, fertilization, and culture in the laboratory. Also, the adoption of an experimental design framework and the standardization of protocols to assist coral reproduction is highly recommended. In terms of experimental design, having a clear formulation of hypothesis for the restoration experiment based on the comparison of variables measured in experimental, reference and control plots effectively estimates restoration success (Chapman, 1998; Croquer et al., 2019). Furthermore, sampling efforts should be established a priori based on power analysis.

It is critical to acknowledge that coral restoration by itself is not the solution for preventing local coral reef decline. Instead, coral propagation must be aligned with specific management strategies aimed at reducing local threats such as overfishing, water pollution, unsustainable coastal development and direct physical damage to the reef such as anchoring (Abelson et al., 2020). One of the advantages of FUNDEMAR's coral restoration approach is the holistic strategy implemented to protect Southeastern Dominican reefs. Though local actions are essential, a coordinated international approach is necessary to reduce global threats to coral reefs such as mass coral bleaching and ocean acidification caused by climate change (Abelson et al., 2020).

To conclude, FUNDEMAR's coral assisted sexual propagation program has become a robust program due to key alliances entailing the private sector, NGOs and governmental agencies, the engagement of the local community, the creation of

Species	Spawning year	Spawning date	DAFM	TAS	No. colonies collected	Min. to 80–90% Fert.	MFR (%)	Approx. No. Embryos (Thousand)	Culture method	Days in culture
Dlab	2019	May 28	10	1:40**	5	_	_	_	Ex situ	14
		May 29	11	1:10**	18	80	100	1,500 —	In situ Ex situ	13
	2020	May 17	10	1:05	4	80	96.2	498	Ex situ	18
		May 18	11	1:00	10	80	94.6	301	Ex situ	17
		Jun 16	11	1:15	11	80	96.5	457	Ex situ	13
Apal	2019	Aug 17	2	2:15	10	220	90	-	ln situ Ex situ	24
	2020	Aug 7	4	2:05	10	160	84.8	328	In situ	21
		Aug 8	5	2:20	9	180*	74.5	118	Ex situ	21
								477	In situ	20
Dcyl	2019	Aug 18	3	1:35	4	60	83.3	_	Ex situ	43*
	2020	Aug 6	3	1:45	3	80	85.9	_	Ex situ	25
Acer	2019	Aug 20	5	2:35	9	180*	67.8	_	In situ	28
		-							Ex situ	
	2020	Aug 7	4	2:35	9	160	88.5	57	Ex situ	21
Cnat	2019	Sep 20	7	1:00	8	140	90	_	Ex situ	25
	2020	Sep 8	6	0:35	5	80	97	410	In situ	23
Ofav	2020	Sep 7	5	2:20	8	180	87.1	516	In situ	_
								_	Ex situ	
Oann	2020	Sep 7	5	3:25	8	120	92.2	_	Ex situ	_

TABLE 1 Assisted reproduction output from 2019 and 2020.

DAFM, Days After Full Moon; TAS, Time After Sunset (hour:minutes); and MFR, Maximum Fertilization Rate. In 2020, Orbicella annularis and O. faveolata developed into larvae but, inexplicably, all cultures died off before settlement. Species name abbreviated to first letter to denote genus and first three letters of species. *In 2019 Dendrogyra cylindrus was ready to be seeded earlier but was kept in culture for a longer time to monitor progress. **Time Before Sunset.

job opportunities and training to become coral restoration technicians, and the creation of sources of income that make the program self-sustainable. The program relies on clear missions and objectives and integrates local stakeholders. Finally, the key of a successful and replicable program is to adapt the existing technologies and customize it to the reality of each country, the conditions of the site where it will be implemented and the technical and economic capacity of the institution.

FUNDEMAR is expected to continue growing in the near future and will continue sharing experiences with other NGOs in the Dominican Republic and the Caribbean region. We hope our story will be useful for others to design their programs and to replicate efforts across the region to scale up coral reef restoration efforts.

DATA AVAILABILITY STATEMENT

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

AUTHOR CONTRIBUTIONS

RS-B conceived, designed and coordinated FUNDEMAR's coral reproduction program, designed and led the Coral Assisted Reproduction Laboratory, acquired funds and resources and performed field work, supervised the data collection and execution of all activities, and contributed equally to the manuscript. MV led data collection, processing and analysis, elaborated the spawning prediction calendars, performed the field work, created figures and tables, and contributed equally to the manuscript. SG-G designed, led, adapted, and improved the construction of the Coral Assisted Reproduction Laboratory, performed field work, trained the technicians for the data collection, supported the creation of figures and tables, and contributed equally to the manuscript. AC contributed to the manuscript conception and structure, performed field work, provided support for the data analysis process, and contributed equally to the manuscript. All authors contributed to the article and approved the submitted version.

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SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fmars. 2021.669505/full#supplementary-material

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