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Authors

Berman, Ofer
Levy, Natalie
Parnas, Haim
[et al.](#)

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Technical Note

Exploring New Frontiers in Coral Nurseries: Leveraging 3D Printing Technology to Benefit Coral Growth and Survival

Ofer Berman ^{1,*}, Natalie Levy ^{2,3,*}, Haim Parnas ¹, Oren Levy ² and Ezri Tarazi ¹

¹ Design-Tech Lab, Industrial Design Department, Faculty of Architecture and Town Planning Technion, Israel Institute of Technology, Haifa 3200003, Israel

² Mina and Everard Goodman Faculty of Life Sciences, Bar-Ilan University, Ramat Gan 52900, Israel

³ Department of NanoEngineering, University of California, La Jolla, San Diego, CA 92093, USA

* Correspondence: ofer.berman@campus.technion.ac.il (O.B.); nrlevy@ucsd.edu (N.L.)

† These authors contributed equally to this work.

Abstract: Coral nurseries and associated techniques are the most common and widespread reef restoration methods worldwide. Due to the rapid decline of coral reefs, coral nurseries need to be eco-friendlier and adapted for effective upscaling to support large restoration projects. We suggest new design and fabrication processes associated with coral gardening and transplantation with 3D printing technology to offer a beneficial solution for growing coral fragments in on-land and underwater nurseries. We describe multiple combinations of building nurseries through the integration of biomimetic substrates and novel solutions for attaching coral fragments. Our methods are supported with supplemental testing of two hybrid substrate designs and coral mounting structures, building upon previous studies in the Gulf of Eilat/Aqaba (GoE/A), Red Sea. We identified and quantified marine invertebrates colonizing the surfaces of our substrates with environmental DNA (eDNA) by targeting the mitochondrial COI gene. We evaluated our coral fragments with and without our mounting structures to obtain an indication of total protein as a proxy for tissue health. We demonstrate the ability to design hybrid nurseries with custom mounting structures using biomimetic substrates, such as large ceramic artificial reefs, or with an interlocking mesh for holding numerous fragments to maximize out-planting efforts. We propose several methods for both land and underwater nurseries catered to various restoration initiatives for cost-effective up-scaling to meet the demands of global reef restoration.

Keywords: coral gardening; additive manufacturing; coral fragmentation; biomimetic design; hybrid substrates; coral transplanting; coral reef restoration



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1. Introduction

The troubling global decline of coral cover by 50% in the last 60 years, has made it increasingly hard to maintain biodiversity and ecosystem functions [1]. New techniques and approaches for coral reef restoration in the last decade [2,3] have made progress towards developing innovative solutions [4–8]. However, calculated steps are still needed to sustainably reach the ecological scales necessary for large-scale restoration.

Coral gardening and nurseries are currently the most widespread techniques for implementing coral reef restoration [2]. There are several methods for regrowing corals, depending on the available resources and materials [3]. Some coral nurseries are made by suspending coral fragments with fishing lines or on floating platforms in the water column [9]. Other nurseries are created from coral fragments attached to calcium-carbonate-based ‘plugs’ [10] or clips [4], which are secured to a substrate or directly transplanted onto a reef. Today, most techniques to attach coral fragments involve epoxy gluing (30%), cable ties (18%), cement (10%), cyanoacrylate glue (4%), wire (3%), and ropes (3%) [3], all of which are disposable, prone to deteriorate, and could impact coral health [2]. Issues associated with coral nurseries are often concerned with population management (e.g.,

genotypes and diversity), scalability, the feasibility of methods (e.g., time underwater), and the survivability of corals [3,11], which is why it is important to understand the ways in which restoration projects can maximize propagation efforts and increase coral transplants to natural reefs.

Artificial reefs (ARs) or artificial substrates/structures are commonly used in almost 20% of coral gardening initiatives [3,8] as stabilizing platforms or nurseries to grow corals [12]. With the increasing rise in ecosystem degradation in the last 50 years, ARs are progressively being used to reform the structural complexity of reefs [6,7], as coastal protection structures (e.g., seawalls) [13], substratum for the recruitment and settlement of benthic and pelagic organisms [14–17], and as an educational tool for tourists [18,19].

Studies that combined fragmented corals with ARs observed an average survival rate of 66% [3]. The reason behind this could be related to several aspects, but one of the most apparent could be the AR providing a structurally complex substrate for the accumulation of beneficial benthic communities to supply essential nutrients and microbes to enhance coral health and growth [6,20]. Furthermore, several studies found that the cohabitation of benthic invertebrates with coral fragments is a useful functional tool to promote growth and improve the survival of corals [21,22]. Additionally, ARs can capture planulae that may be released from synchronized spawning events or from polyp bailout to help further the restoration process [23,24].

Based on this concept, the next logical step towards rethinking methods for restoration initiatives should work to combine gardening corals or transplants with ARs in a way that considers both entities as one method. This could also help to solve the scalability and survivability issues typically associated with stand-alone coral gardening and nursery practices. Three-dimensional printing (hereafter, 3DP) is a viable technology that already has been employed in coral reef restoration and ecological studies [5–8,25–29].

Digital fabrication and parametric design used for creating ARs, besides materiality, offer a range of new capabilities and customization tools. Three-dimensional printing can produce unique artifacts without the use of a mold to generate the complexities, textures, and features of natural coral reefs [6]. Three-dimensional printing is one of the best-known modalities to form structures with intricate and highly diverse complexities to help get as close to a design that mimics or replicates nature, known as biomimicry or biomimetic design. In terms of scale, there are various 3DP technologies that can offer increased shelter space to host a variety of reef species. Parametric design differs from conventional design in that it has a specific design space, which is controlled by algorithms. Within this space, one can export a specific outcome based on selected parameters from a variety of inputs. The result does not have to be the volumetric representation of the AR, but could be the final code that directs the 3D printer on how to move and build the requested object [7].

In this research, we describe new methodologies for rapid and accessible approaches for outplanting corals. This was performed by rethinking methods for coral reef restoration—artificial substrates (i.e., settlement tiles) and attaching coral fragments—as one multi-functional, customized tool to provide improvements for the survivability and sustainability of restoration initiatives. Here, we show a novel process for designing and fabricating two different biomimetic settlement tiles from ceramic terracotta that include 3D-printed mounting structures to affix coral fragments with an easy-release system. Our research showcases the successful deployment and performance of our innovative settlement tiles, featuring coral fragments adhered with mounting structures, at two coral reefs in the Gulf of Eilat/Aqaba (GoE/A), Red Sea. These new techniques help with the transition away from the use of potentially harmful materials and provide an accessible solution to globally scale-up coral restoration efforts in a custom and tailored way. With the urgent need for innovation and scalability to restore and rebuild coral ecosystems, our approach addresses a critical need for creating efficient and effective tools to support large-scale restoration projects.

2. Materials and Methods

2.1. Research through Design

The methodological paradigm leading to this study utilizes the Research through Design (RtD) approach, which uses design as a means of inquiry and investigation [30]. Rather than viewing design solely as a means of creating products, services, or experiences, RtD uses the design process as a way of generating new knowledge and insights. As can be seen in this technical study utilizing the RtD methodology, iterative cycles of design and testing were used to explore and conceptualize advanced coral nurseries, and the many prototypes created serve as artifacts that can be analyzed and interpreted to generate new knowledge.

2.2. Material Selection and Preparation

To produce the pressed ceramic tiles (Figure 1), two half-open plaster molds were made using gypsum powder (Supraduro Saint-Gobain); the gypsum powder was added to water and gently homogenized to avoid air bubbles. After finalizing the mold, the atomized clay mixture with 25% grog (Goerg & Schneider Body 0354) and without any modifications was hand-pressed into the mold and lay there for more than an hour, until the clay was firm enough to be pulled out.

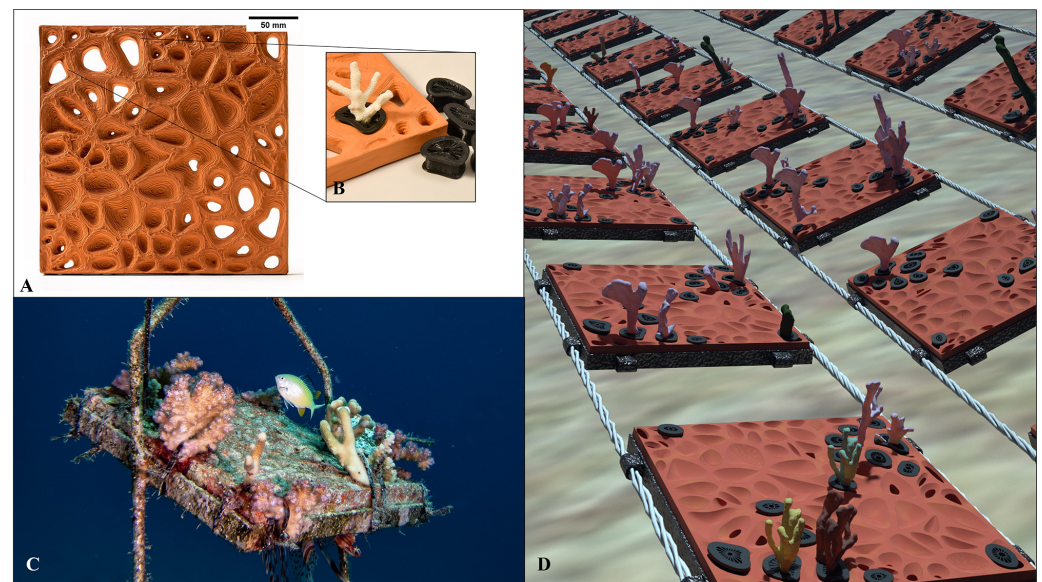


Figure 1. Biomimetic settlement tile adapted with mounting structure. (A) The square ceramic tile after bisque firing, and the initial design of the mounting structure custom-made to fit specific holes in the tiles. (B) Custom mounting structure with coral skeleton; the flexible mounting structures are printed with bottom and top overhangs which help to secure it to the tile. (C) The square tile submerged with the planted fragments (not all attached with the mounting structure). (D) Conceptualization of a horizontal floating coral nursery composed of square ceramic tiles and custom mounting structures.

To produce 3DP ceramic tiles, atomized clay mixtures (Goerg & Schneider Body 0311) were used, containing a high level of iron oxide (6.5%) and 20% humidity (Figure 2). To increase plasticity, 25 mL of water per kilogram of clay was added to the clay body. Additionally, sodium silicate was added in a ratio of 1.25 mL per kg of clay to improve bindery, as suggested by [31]. A pugmill machine (Petter Puggger VPM-60) was used to mix the mixture for an hour before printing.

The main design feature of the mounting structures is the ability to create multiple bristles that hold the inserted coral fragment gently but firmly in place. To allow that functionality, the different mounting structures presented in this study were made using an ‘off-the-shelf’ 3DP thermoplastic polyurethane (TPU) with approximately 90 shores, as

this material is flexible and durable, and frequently used in marine [saline] environments, with minimal weathering and the capability to maintain the tensile properties [32].

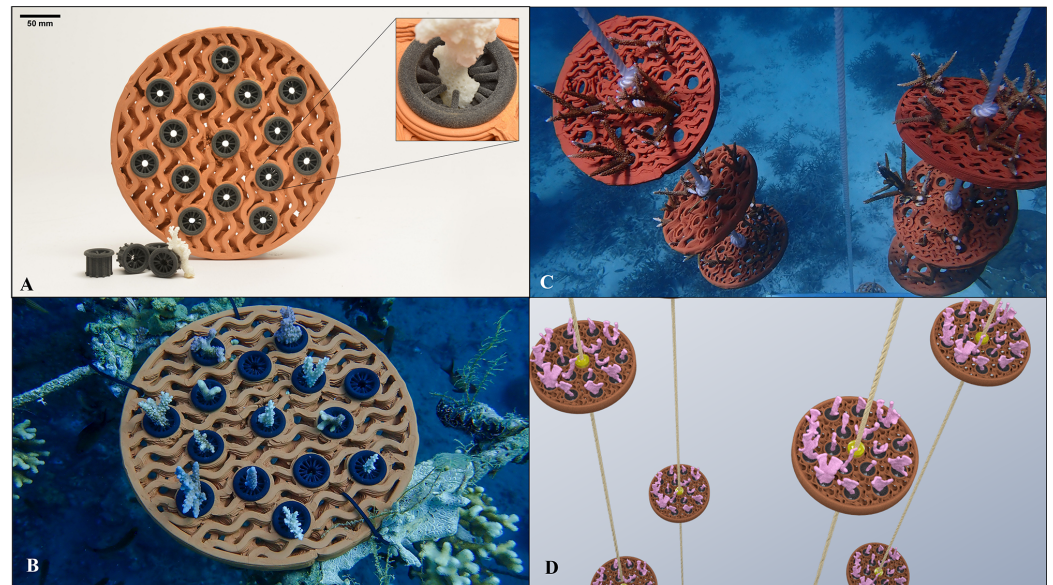


Figure 2. Complex 3DP settlement tile configurations with mounting structure. (A) The round 3DP ceramic tile after bisque firing, with second-version universal mounting structures, exhibiting a more advanced design. Mounting structures are anchored to the holes in the tiles. (B) Underwater testing of the design of the mounting structure to attach coral fragments (in the Northern Red Sea, 2023). (C) Testing different infill patterns of the round tiles with fragments (without inserts) underwater (in the Maldives, 2022). (D) Conceptualization of a vertical floating coral nursery made up of round ceramic tiles and universal mounting structures.

2.3. Preliminary Hybrid Design of Biomimetic Settlement Tiles and Mounting Structures

The preliminary square settlement tile was modeled in the Computer-Aided-Design (CAD) software, Fusion 360 (V.2.0.7029). The tile design featured different topographic textures, such as ridges and cavities similar to the topology of a coral reef (Figure 1A). An archetype gypsum mold was produced using a PLA 2.85 mm filament and a large fused deposition modeling (FDM) printer (3DP workbench). The gypsum mold itself was used to form the tiles by pressing the clay on the open mold. Later, both sides of the tile were fitted together, and after dehydration, were bisque-fired at 900 °C.

After the tiles were used successfully in different underwater experiments, it became necessary to affix several different species of branching corals to the top of the tiles. This led to the creation of three types of customized 3DP solutions by fitting a specific mounting structure to three of the largest already-existing holes of the tiles (Figure 1B). The three mounting structures (types A, B, and C) were designed to hold the coral fragments with multiple soft 3D-printed bristles, while the outer shape was fitted to the segmented opening in the ceramic tile. These units were manufactured using a desktop 3DP machine (Prusa i3 MK3S+, Prusa Research, Prague, Czech Republic) with a 0.4 mm nozzle and a build volume of 25 cm wide, 21 cm deep, and a height of 21 cm. The mounting structure's bristles were divided into two sub-groups. The first group of bristles started at the bottom of the model and contained fifteen strands with a width of 0.4 mm, equally allocated around the center mass of the shape contour. The second group was offset 0.3 mm higher and the equally allocated fifteen strands were shifted twelve degrees around the center. The two sub-groups were patterned vertically sixteen times in steps of 0.9 mm. The printer deposited material between the outer and inner walls of the mounting structure. Eventually, the inner tube wall was removed with a die-cut to leave a hole for inserting the fragments and releasing the bristles. A 0.3 mm layer height and 60 mm/s printer X-Y movement were coordinated with the bristle design.

2.4. Meshed Coral Nursery

The simplicity of printing and using the customized mounting structures led to the conceptualization of the interlocking mounting mesh, which could be used as a new way to establish coral nurseries. A simplified universal mounting structure design (v1) was developed to create a coral nursery mesh. Here, instead of patterning the bristle group upward, the bristles were arrayed over a helix curve in a pitch of 0.3 mm and 16 revolutions total. The thickness of the universal structure was 5 mm, resulting in 300, 0.8 width strands that turned to become 600 separated bristles after drying and cutting the centered tube wall. The universal mounting structure was packaged in 3DP housing with built-in hinges (Figure 3), enabling it to be patterned in two directions, creating a mesh of mounting structures. The 3DP housing was designed in CAD software (Solidworks 2021–2022) and manufactured on a powder-bed, MJF technology printer (HP 580) with nylon12 as the primary material. The hinge axes were made from a metal spring alloy with a diameter of 1 mm.

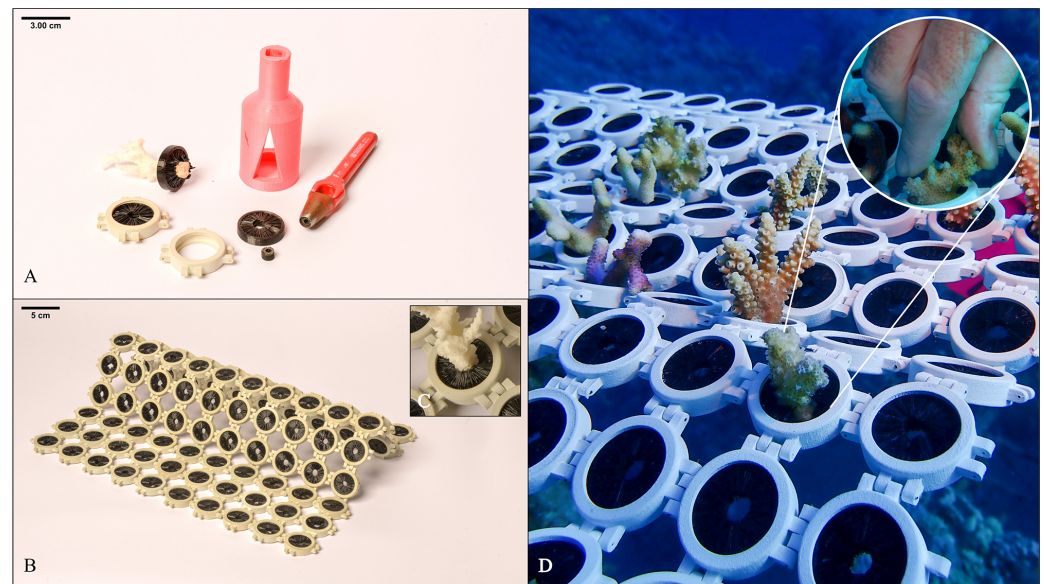


Figure 3. Proof of concept of coral nursery mesh design consisting of universal mounting structures. (A) First-generation design of the universal mounting structure, 3DP modular nylon housing, and a 3DP jig to die-cut the inner wall to release the bristles. (B) Overview of the flexible nursery prototype, which in a 100 m² floating nursery that could contain up to 90,000 fragments. (C) Close-up of a coral skeleton secured to the meshed mounting structure. (D) Testing the prototype underwater with live coral fragments. The design of the mesh mounting structures can wrap around existing artificial structures such as concrete columns or piers, turning them into a nursery.

2.5. Optimized Hybrid 3DP Tile Design and Mounting Structures

Optimized 3DP round tiles, conjointly with universal mounting structures, were designed in CAD software (Solidworks 2021–2022) as simple 30 cm diameter flat circular plates containing an array of fifteen holes for the mounting structures (Figure 2A). The model was exported as a stereolithography model (.stl) and imported into Cura slicer (V 4.11.0). By setting the printer dimensions and choosing the Marlyn Gcode flavor the Cura slicer was fitted to a Wasp 3MT printer (Wasp S.r.l, Massa Lombarda, Italy). Next, the slicer was configured to have zero layers in the top and bottom layers, one perimeter outline, a 1.5 mm layer height, and a Gyroid-type infill of 25%. This ensured finer results coupled with printing with a 4 mm nozzle. The Wasp 3MT printer was used to print the ceramic elements with a building volume of $\approx 1000 \text{ mm} \times 1000 \text{ mm} \times 1000 \text{ mm}$ (h). In this system, clay was extruded through a 4 mm nozzle using a commercially available pneumatic linear extruder (WASP 5L Clay Tank (Wasp S.r.l, Massa Lombarda, Italy)).

The evolved design of the universal mounting structures (v2) fitted to the round ceramic substrate was designed to be printed on a selective laser sintering (SLS) powder bed printer (Fuse 1 + 30 W, Formlabs Inc. Somerville, MA, USA) using a designated TPU material (90A v1, Formlabs Inc. Somerville, MA, USA) to build the model. The building volume of the printer was 165 mm (w), 165 mm (d), and 300 mm (h) and the printer layer thickness setting was 0.11mm, which implicated fine printing results at different orientations of the model. The effective height of the mounting structure was 20 mm height containing 68 2 mm thick and 8 mm length coarse bristles. The universal v2 mounting structure was designed for easy installment on the tiles by pushing it into the 30 mm holes. The outer surface contained fixing anchors to keep it from dislodging, and elevated stripes that helped to stabilize itself by creating constant pressure against the ceramic.

2.6. Field Experiments Using Settlement Substrates and Mounting Structures with Corals

The field experiments in our study build upon previous research [17] by comparing our first terracotta tile design, with and without mounting structures, to eventually provide one restoration device [17]. The square biomimetic settlement tiles (Figure 1A) were deployed underwater in March 2019 at two sites in the northern and southern sections of the GoE/A. It was used as part of a study on the recruitment of reef biodiversity in restoration and evaluation using environmental DNA (eDNA) metabarcoding of organismal biomass samples [17]. Tiles were monitored every two months using photogrammetry to understand the successional growth and settlement of benthic reef organisms for a little over a year. This study confirmed the importance of the microhabitat structure and surface orientation for provisioning habitats for benthic reef organisms and recruiting key scleractinian corals [17].

eDNA was collected from organisms scraped and homogenized from ceramic tiles. DNA was extracted and cleaned using a modified version of the DNeasy PowerMax Soil Kit protocol and the DNeasy PowerClean Cleanup Kit, following the manufacturer's protocol (Qiagen, Hilden, Germany) [17]. The Mitochondrial Cytochrome c. Oxidase subunit I (COI) DNA region was amplified at the 313bp region using primers created to target metazoans (mlCOLintF and jgHCO2198), and index primers were attached with a second PCR for IlluminaMiSeq sequencing. The PCR product was cleaned between PCR cycles using Agencourt AMPure XP paramagnetic beads (Beckman Coulter™, Brea, CA, USA) [17]. Raw reads were processed using DADA2 package version 3.11 [33] and R software v4.1.0 [34]. Reads were converted to amplicon sequence variants (ASVs) after being quality-filtered, trimmed, merged, and chimeras were removed. OTUs were achieved with clustering ASVs from DADA2 with VSEARCH [35] with a 97% identity threshold and transferred to the LULU algorithm [36]. The remaining OTUs were assigned to taxonomic groups using BASTA [37], the Last Common Ancestor (LCA) based on NCBI taxonomies [37], and the Bayesian Least-Common Ancestor (BCLA) Taxonomic Classification software [38] against Midori-Uniquiev20180221 [39,40]. Statistical analysis was conducted in R software v4.1.0 [34] with the vegan package [41]. OTU tables were derived from "core_diversity_analyses.py" in QIIME v1.9 [42]. Data visualizations were created with ggplot2 [17,43].

In a follow-up experiment, the biomimetic square tiles were redeployed in February 2021 at two sites in the northern and southern sections of the GoE/A for both 6 and 12 months (Figure 1C). The goal of this study was to understand the differences between the reef ecobiome of healthy and degraded reefs and the impacts on coral physiology. This study led to the conceptualization of creating mounting structures for attaching corals to artificial substrates. The first preliminary, custom mounting structures were designed after the creation of the biomimetic square tiles.

Several physiological tests were conducted on the coral fragments as part of the study, including the total protein content of coral tissues, used as a proxy for tissue health. Coral tissue was recovered using an airbrush and ice-cold filtered seawater (0.22 μM) at the Interuniversity Institute of Marine Sciences in Eilat (IUI). Tissues were then homogenized for 30 s (Kinematica Polytron™ PT2100 Benchtop Homogenizer, Littau-Lucerne, Switzerland)

and a 100 µL sample of supernatant was collected for total protein concentration. Samples were sonicated in an ice-cold water bath and vortexed. Total protein was prepared using a bovine serum albumin as a standard (Quick Start Bradford Protein Assay Kit 1, Bio-Rad, Rishon Le Zion, Israel) in a 96-well plate and the standard curve was created [44]. Total protein content was read using a Synergy HT microplate reader (BioTek, Agilent, Santa Clara, CA, USA). Protein content was normalized to the volume (mL) of tissue and to the skeletal surface area, determined with a wax dip technique [45,46]. Differences in the total protein between coral species from baseline to the tiles (substrate) were tested using a pairwise Wilcoxon Signed-Rank test. Data were visualized in R software v4.2.1 [34] using the ggplot2 package [43] to create the boxplot.

3. Results

3.1. Production Time, Estimated Cost, and Feasibility

Following these studies, we began working towards creating new settlement substrates, which combined the design, fabrication, and production process synergistically with mounting structures, to make one restoration product. The square tiles were made by a professional ceramicist and involved the construction of the plaster press mold (16 h). Pressing two sides of the tiles in clay and bonding them together into one tile took approximately 4.5 h. The whole process cost about USD 140 per tile and resulted in 18 complete and burned ceramic tiles. The round 3DP tile was fabricated in under 36 min, weighted right after printing at 1.9 kg, and cost in material about USD 1.6 in material. Both the square tile hand-pressing and the round 3DP tile drying and firing processes were similar.

The time taken to 3D-print the customized mounting structures with the FDM printer ranges from 27 min for type A and up to 41 min for type B (Table 1). The time taken to print a single universal mounting structure with the FDM printer is about 18 min; when printing a full build plate containing 42 units, the time drops to 17 min, alternately, per unit. The complete weight of a single universal mounting structure, before cutting the center of it, is 2.1 g, so in a 1 kg standard TPU spool, about 476 units could be made. The typical cost of a standard TPU spool on Amazon [47] is about USD 22, so the estimated direct cost for the universal unit without including the printer purchase is USD 0.04. For the universal v2 mounting structure, 144 units can be made during 24 h of printing, which results in 10 min per unit on average. The estimated cost of each unit is about USD 0.62 and weighs 6.4 g. As for the 3DP housing, 243 units could be printed on a single full batch on Formlabs Fuse1 using PA12 material over a 20 h printing time, Resulting in an average of about 5 min per unit and with an estimated cost of USD 0.71.

While further analysis is required to accurately measure the time required to mount the coral fragments, it is already evident that the anchoring of the fragments using the various mounting structures is a remarkably straightforward and efficient process. The fragment attachment procedure occurs rapidly, similar to inserting a pushpin into a corkboard, taking only a matter of seconds (see Supplementary Materials). Although the reliability of the mounting structures requires additional quantification, numerous fragments have been successfully installed using different mounting structure designs and have remained securely attached for a duration exceeding six months.

Table 1. A comparison of fabrication time, weight, and material costs.

	Square Tile (Hand-Pressed)	Round Tile (3DP)	3DP Housing	Customize Mounting Structure (Type A, Type B, Type C)	Universal Mounting Structure (v1)	Universal Mounting Structure (v2)
Fabrication time (per unit in min)	270	36	4.93	27, 41, 34	17–18	10
Weight (per unit in g)	1570	1366	2.8	2.3, 4.1, 3.5	2.1	6.4
Material cost (per unit in USD)	1.7	1.6	0.71	0.05, 0.09, 0.07	0.04	0.62

3.2. Feasibility of Hybrid Tiles and Mounting Structures

eDNA sequencing of the COI gene, using metabarcoding, was successfully used to identify and quantify the benthic organisms present on the biomimetic ceramic tiles ($n = 40$; includes controls) (Figure 4). There were a total of 7,638,955 sequences across Site A and B of 29 different phyla present on the biomimetic tiles from organismal biomass samples. The most abundant phyla discovered on the tiles, in order, were Chordata (1,533,554), Mollusca (1,308,824), Bryozoa (1,134,064), Arthropoda (1,020,474), Annelida (963,009), and Cnidaria (925,550) (Figure 4).

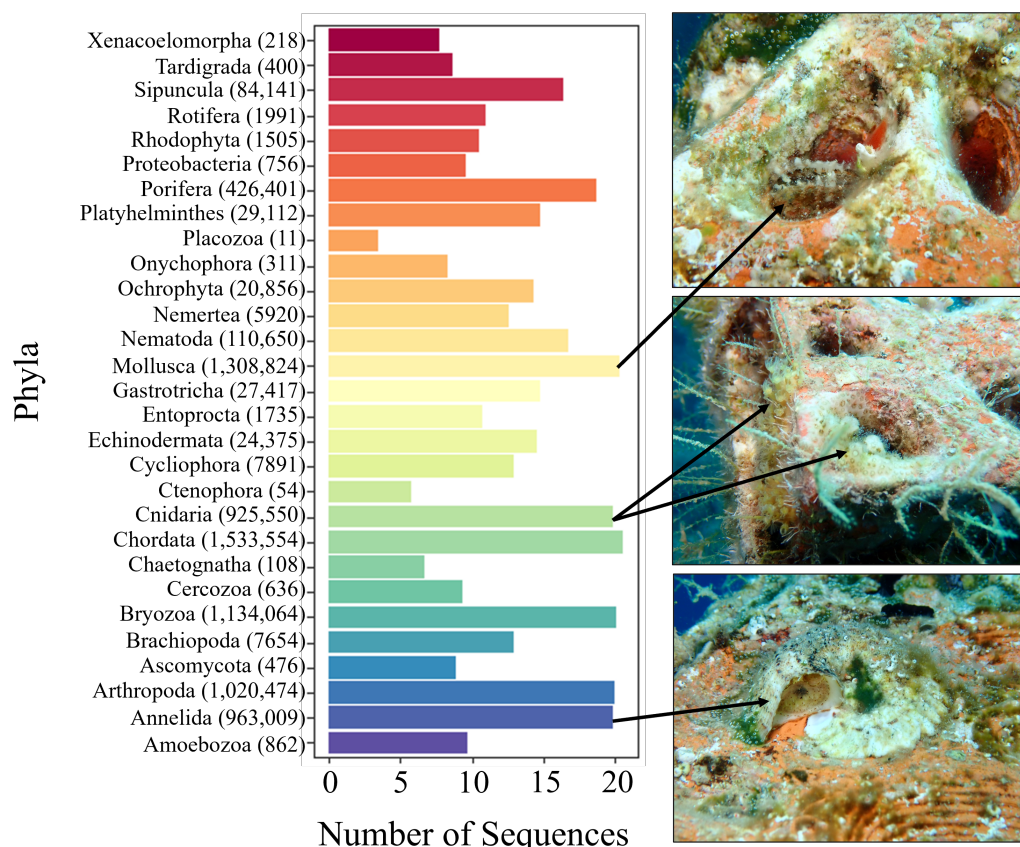


Figure 4. Total number of sequences of phyla found on parametric square tiles. Phyla were detected from eDNA present only on tiles at two study sites in the GoE/A after quality control. Pictures represent the organisms from each phyla to show habitat partitioning on tiles (adapted with permission from Ref. [17]; Science of The Total Environment; Elsevier; 2023).

The total protein content from the tissues of fragmented corals, *Acropora eurystoma*, *Stylophora pistillata*, and *Pocillopora damicornis* ($n = 45$), in mounting structures, was compared with baseline corals that were fragmented from the same colonies after a period of 6 months (Figure 5). The protein content was used as a proxy for tissue health to indicate any changes throughout the duration corals were secured in mounting structures and to tiles. Not all the corals were held with the mounting structures; however, fragments were selected randomly to understand the potential differences between corals originally from the reef site and the corals that were mounted to tiles for 6 months, with a mounting structure or without. There were statistically significant differences between the total protein content in baseline corals collected from each respective site and the fragments that were mounted onto the tiles (substrate) for both *P. damicornis* ($p < 0.01$) and *S. pistillata* ($p < 0.001$) (Figure 5). Total protein content was significantly higher in both *P. damicornis* and *S. pistillata* from tiles compared with baseline corals, whereas no significant difference was observed between baseline and tiles fragments for *A. eurystoma* (Figure 5).

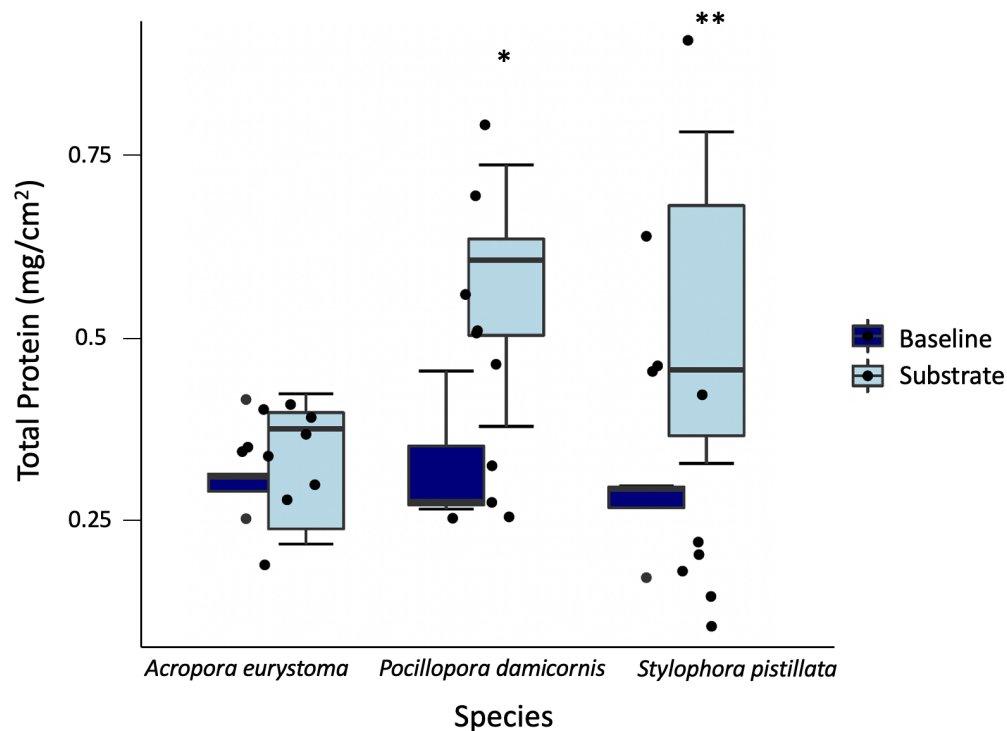


Figure 5. Total protein content from the tissues of three branching corals. Baseline corals represent the fragments before they were attached to substrates (tiles). The substrate treatment is the 6 months after they were mounted to tiles. Asterisks represent the level of p -value significance: $p < 0.01$ (*) and $p < 0.001$ (**). Dots represent replicates.

4. Discussion

Expanding the Coral Restoration Toolbox

The current development of hybrid substrates composed of 3DP ceramic tiles and integrated mounting structures can expand the toolbox of customized and scalable reef restoration solutions. We present two tile designs with high topographical complexity that include a novel way to mount coral fragments securely, providing the ease, speed, and flexibility needed in such tasks, especially underwater. In both tile designs, we suggest how to multiply and expand their use and to create various large-scale nurseries in the form of flat floating surfaces or towers (Figures 1 and 2). Furthermore, we offer an innovative prototype of a coral nursery constructed using 3DP nylon housing with mounting structures that interlock and create a grid for gardening fragments (Figure 3). Additionally, mounting structures can be applied to the topography of large-scale biomimetic ceramic artificial reefs (Figure 6) [6]. The results of our methodology indicate the potential for benefiting fragment survival while maximizing the number of fragments planted per square meter.

Our results build upon previous studies involving the square biomimetic settlement tiles, which demonstrated the colonization of organisms analogous to the benthic biota of each coral reef site [17]. We observed that the phylum Cnidaria was the most diverse in terms of species identified on the tiles. Scleractinian corals were more predominant on the top of the tiles, settling in the textured grooves, whereas soft corals were generally found on the underside or sides of the tiles, which could be due to various factors such as light, heterotrophy vs. autotrophy, spatial competition, and life history strategies [17]. These tiles played a key role in successfully replicating the natural reef topology and complexity by provisioning habitats within its cryptic and sheltered areas. Hybrid tiles and mounting structures can provide newly fragmented corals the platform of a rich and diverse reef community to support their growth, which could help to both ensure their future resilience and provide methods by which to upscale. In the second study, the universal v1 mounting structures were used in the same square biomimetic tiles to hold fragments from three

common branching species from the GoE/A. The mounting structures held coral fragments underwater successfully for 6 months (second half of the study). Although there was some observed tissue loss surrounding the small area of the coral that was inside the mounting structure (like at most attachment points), when the fragments were collected for physiology tests, the corals were visually healthy with no noticeable tissue loss or necrosis in any areas of the coral. The higher protein content discovered in the *P. damicornis* and *S. pistillata* fragments that were secured to tiles could be related to the established layers of the biofilm and organisms colonizing the tiles. This could have provided essential nutrients and microbes to boost the health of mounted corals over the course of the experiment, while *A. eurystoma* did not experience any changes in protein content, which could imply that the mounting structures did not detrimentally impact the coral tissue.

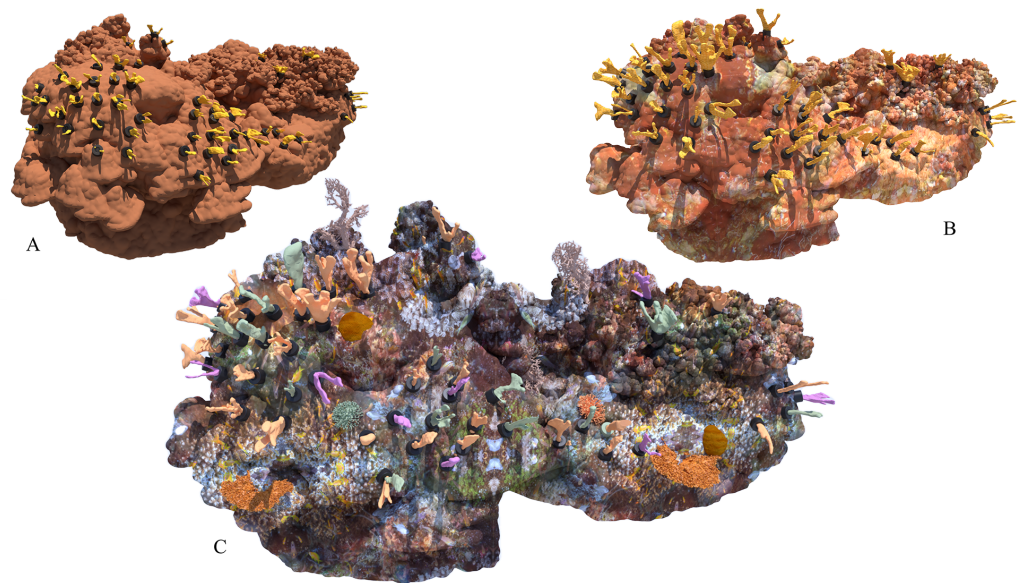


Figure 6. Visualization of a benthic organism succession on a ceramic reef with planted fragments (adapted with permission from Ref. [6]; Science of The Total Environment; Elsevier; 2022). (A) Newly deployed biomimetic AR with fragments. (B) Projection of a six-month- to one-year-old AR with growing fragments, with a gradual accumulation of marine invertebrates, microorganisms, and algae. (C) Fully covered coral reef ecobiome growing on the AR with larger and diverse coral fragments.

Current coral gardening solutions require underwater operational skills to attach fragments, combined with a finite air supply during SCUBA that can limit the amount of planted or treated corals [9,10]. Additionally, most methods involve toxic adhesives or epoxies that may negatively influence the reef environment and the coral itself [2]. Often, they only offer a one-time (disposable or degradable) solution, which may decrease the overall sustainability of the operation [3]. Furthermore, our solution could help to overcome these current challenges, including the flexibility to arrange, move, or transplant multiple fragments or nursery structures at once, from one site to another. Future applications of this technology in marine ecosystem restoration, specifically for coral reefs, can lead to the expansion of floating or benthic nurseries using biomimetic settlement tiles and either customizable or universal mounting structures for fragments (Figure 3). Additionally, this could make it easier to rear corals in aquaria or shallow-water nurseries before transporting them to reefs (Figure 1 and 2). For maximizing out-planting initiatives, an adaptable mesh configuration made of mounting structures in housings could offer many possibilities for scaling-up operations both in and out of the field (Figure 3).

Coral propagation and transplantation remain the most common reef restoration method at present, but operational costs can be expensive when compared with the survival rate of fragments [48], leading to issues with effective upscaling [22]. The practice of coral nurseries requires constant maintenance to grow fragments. Our new design suggests

simple, cost-effective, and feasible solutions to securely mount fragments with an easy-release system for use in different environments. Additionally, our mounting structures were tested with scleractinian branching corals, *A. eurystoma*, *S. pistillata*, and *P. damicornis*, as well as soft corals, such as *Heteroxenia fuscescens*. However, our mounting devices are not yet optimized for other types of corals (e.g., encrusting), but the potential is there.

Combining data-driven, biomimetically engineered ARs with strategic spaces for placing mounting structures could better accommodate large-scale operations by providing habitat complexity, settlement substratum [6], and a refuge to many important reef creatures, such as herbivorous fish [7]. It could also be tailored to already existing commercial and experimental ARs, seawalls, jetties, and coastal structures, providing structural stability, protection and an ecotourist attraction. Our solution could enable more efficient transplantation of fragments without potential stress on the living tissue or damaging the coral, as indicated by our preliminary findings. Furthermore, our approach could be incorporated as a nursery toolkit for coral practitioners, governments, NGOs, or for commercial ecotourism markets (e.g., eco-resorts or SCUBA adventures) and aquaria-enthusiasts [49,50].

Constructing a floating tower nursery, utilizing round tiles and universal v2 mounting structures, with the capacity to accommodate 10,000 coral fragments necessitates approximately 715 tiles. The material cost for these tiles would be approximately USD 1144, and the printing process would require approximately 429 h (equivalent to 17 days). The universal v2 mounting structures required for this nursery would cost around USD 6200 in materials and would take approximately 70 days to print.

To establish a floating mesh nursery capable of accommodating 10,000 fragments within a 16 square meter area, the production of universal v1 mounting structures would require an estimated material cost of approximately USD 400 and a time frame of 125 days. However, with the utilization of four dedicated printers, this production period could be condensed to one month. The housing for the universal mounting structures would be approximately USD 0.71 per unit and could also be manufactured within one month of printing, resulting in a total cost of USD 7100.

5. Conclusions

For extensive projects, like the 10 k fragment nursery benchmark, 3D printing thousands of small identical pieces currently falls short in comparison to mass-production methods and readily available materials like nets, ropes, or PVC pipes. To illustrate these distinctions, we offer a potential strategy for streamlining the production of next-generation nurseries on an industrial scale. Ultimately, this could lead to enhanced accessibility and affordability for coral practitioners and operations worldwide. The primary bottleneck in the financial feasibility of a 10,000-fragment nursery based on 3DP methods is the high cost of the mounting structures and housing. Mass-production techniques such as injection molding could be employed for a quick and economical solution to the fabrication of universal mounting structures. Heading in this direction will require certain geometric modifications to facilitate the molding process, and will lead, potentially, to tight product development (e.g., merging the mounting structure with its housing). Using an inexpensive industrial-grade TPU granule such as Elastollan by BASF [51], or other similar brands, (prices are ranged in several USD per kg) could result in a significantly low-budget nursery cost of approximately USD 200.

We believe more extensive work should be conducted to fully characterize the impact various coral gardening methods may have on coral growth and survival. In the future, our 3D-printed solutions, such as mounting structures, could be 3D-printed with biotechnology from different living materials [52–54] that could be infused with beneficial nutrients [55] to protect corals at fragment sites [56] and boost their resilience [57]. Mounting structures could be 3D-printed from gels or biodegradable materials that could dissolve over time, leaving the natural attachment of corals to either biomimetic substrates (Figure 6) or the reef itself. As some processes to 3DP settlement substrates and ARs may still produce carbon emissions, 3DP substrates for attaching coral fragments from materials such as cellulose

could potentially uptake excess carbon at reef sites [58], leading to the hybridization of advanced biological substrates and coral mounting structures that could serve multi-functional purposes in coral reef restoration.

Supplementary Materials: The following supporting information can be downloaded at: <https://www.mdpi.com/article/10.3390/jmse11091695/s1>. Testing the easy release mounting structure (v2) in a natural coral reef in the Red Sea, Israel.

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References

1. Eddy, T.D.; Lam, V.W.; Reygondeau, G.; Cisneros-Montemayor, A.M.; Greer, K.; Palomares, M.L.D.; Bruno, J.F.; Ota, Y.; Cheung, W.W. Global decline in capacity of coral reefs to provide ecosystem services. *One Earth* **2021**, *4*, 1278–1285. [CrossRef]
2. Bostrom-Einarsson, L.; Ceccarelli, D.; Babcock, R.C.; Bayraktarov, E.; Cook, N.; Harrison, P.; Hein, M.; Shaver, E.; Smith, A.; Stewart-Sinclair, P.J.; et al. *Coral Restoration in a Changing World: A Global Synthesis of Methods and Techniques*; Reef and Reinform Research Center: Cairns City, QLD, Australia, 2018.
3. Bostrom-Einarsson, L.; Babcock, R.C.; Bayraktarov, E.; Ceccarelli, D.; Cook, N.; Ferse, S.C.A.; Hancock, B.; Harrison, P.; Hein, M.; Shaver, E.; et al. Coral restoration—A systematic review of current methods, successes, failures and future directions. *PLoS ONE* **2020**, *15*, e0226631. [CrossRef] [PubMed]
4. Suggett, D.J.; Edmondson, J.; Howlett, L.; Camp, E.F. Coralclip[®]: A low-cost solution for rapid and targeted out-planting of coral at scale. *Restor. Ecol.* **2020**, *28*, 289–296. [CrossRef]
5. Albalawi, H.I.; Khan, Z.N.; Valle-Pérez, A.U.; Kahin, K.M.; Hountondji, M.; Alwazani, H.; Schmidt-Roach, S.; Bilalis, P.; Aranda, M.; Duarte, C.M.; et al. Sustainable and Eco-Friendly Coral Restoration through 3D Printing and Fabrication. *Acs Sustain. Chem. Eng.* **2021**, *9*, 12634–12645. [CrossRef]

6. Levy, N.; Berman, O.; Yuval, M.; Loya, Y.; Treibitz, T.; Tarazi, E.; Levy, O. Emerging 3D technologies for future reformation of coral reefs: Enhancing biodiversity using biomimetic structures based on designs by nature. *Sci. Total Environ.* **2022**, *830*, 154749. [[CrossRef](#)] [[PubMed](#)]
7. Berman, O.; Weizman, M.; Oren, A.; Neri, R.; Parnas, H.; Shashar, N.; Tarazi, E. Design and application of a novel 3D printing method for bio-inspired artificial reefs. *Ecol. Eng.* **2023**, *188*, 106892. [[CrossRef](#)]
8. Schmidt-Roach, S.; Klaus, R.; Al-Suwailem, A.M.; Prieto, A.R.; Charrière, J.; Hauser, C.A.E.; Duarte, C.M.; Aranda, M. Novel infrastructure for coral gardening and reefscaping. *Front. Mar. Sci.* **2023**, *10*, 202. [[CrossRef](#)]
9. Rinkevich, B. Conservation of Coral Reefs through Active Restoration Measures: Recent Approaches and Last Decade Progress. *Environ. Sci. Technol.* **2005**, *39*, 4333–4342. [[CrossRef](#)]
10. Abelson, A. Artificial reefs vs coral transplantation as restoration tools for mitigating coral reef deterioration: Benefits, concerns, and proposed guidelines. *Bull. Mar. Sci.* **2006**, *78*, 151–159.
11. Putchim, L.; Thongtham, N.; Hewett, A.; Chansang, H. Survival and growth of *Acropora* spp. in mid-water nursery and after transplantation at Phi Phi Islands, Andaman Sea, Thailand. In Proceedings of the 11 th International Coral Reef Symposium, Fort Lauderdale, FL, USA, 7–11 July 2008; p. 24.
12. Higgins, E.; Metaxas, A.; Scheibling, R.E. A systematic review of artificial reefs as platforms for coral reef research and conservation. *PLoS ONE* **2022**, *17*, e0261964. [[CrossRef](#)]
13. Sedano, F.; Navarro-Barranco, C.; Guerra-García, J.; Espinosa, F. Understanding the effects of coastal defence structures on marine biota: The role of substrate composition and roughness in structuring sessile, macro-and meiofaunal communities. *Mar. Pollut. Bull.* **2020**, *157*, 111334. [[CrossRef](#)]
14. Harriott, V.J.; Fisk, D.A. A comparison of settlement plate types for experiments on the recruitment of scleractinian corals. *Mar. Ecol. Prog. Ser.* **1987**, *37*, 201–208. [[CrossRef](#)]
15. Leray, M.; Knowlton, N. DNA barcoding and metabarcoding of standardized samples reveal patterns of marine benthic diversity. *Proc. Natl. Acad. Sci. USA* **2015**, *112*, 2076–2081. [[CrossRef](#)] [[PubMed](#)]
16. Higgins, E.; Scheibling, R.E.; Desilets, K.M.; Metaxas, A. Benthic community succession on artificial and natural coral reefs in the northern Gulf of Aqaba, Red Sea. *PLoS ONE* **2019**, *14*, e0212842. [[CrossRef](#)]
17. Levy, N.; Simon-Blecher, N.; Ben-Ezra, S.; Yuval, M.; Doniger, T.; Leray, M.; Karako-Lampert, S.; Tarazi, E.; Levy, O. Evaluating biodiversity for coral reef reformation and monitoring on complex 3D structures using environmental DNA (eDNA) metabarcoding. *Sci. Total. Environ.* **2023**, *856*, 159051. [[CrossRef](#)]
18. Monchanin, C.; Mehrotra, R.; Haskin, E.; Scott, C.M.; Plaza, P.U.; Allchurch, A.; Arnold, S.; Magson, K.; Hoeksema, B.W. Contrasting coral community structures between natural and artificial substrates at Koh Tao, Gulf of Thailand. *Mar. Environ. Res.* **2021**, *172*, 105505. [[CrossRef](#)]
19. Howlett, L.; Camp, E.F.; Edmondson, J.; Edmondson, J.; Agius, T.; Hosp, R.; Coulthard, P.; Edmondson, S.; Suggett, D.J. Adoption of coral propagation and out-planting via the tourism industry to advance site stewardship on the northern Great Barrier Reef. *Ocean. Coast. Manag.* **2022**, *225*, 106199. [[CrossRef](#)]
20. Marangon, E.; Laffy, P.W.; Bourne, D.G.; Webster, N.S. Microbiome-mediated mechanisms contributing to the environmental tolerance of reef invertebrate species. *Mar. Biol.* **2021**, *168*, 89. [[CrossRef](#)]
21. Omori, M. Success of mass culture of *Acropora* corals from egg to colony in open water. *Coral Reefs* **2005**, *24*, 563. [[CrossRef](#)]
22. Neil, R.C.; Humphrey, C.; Bourne, D.G.; Heyward, A. Co-culture with grazers can improve survival and growth of multiple coral species. *Aquaculture* **2021**, *544*, 737095. [[CrossRef](#)]
23. Omori, M. Coral restoration research and technical developments: What we have learned so far. *Mar. Biol. Res.* **2019**, *15*, 377–409. [[CrossRef](#)]
24. Leonard, C.; Hédouin, L.; Lacorne, M.C.; Dalle, J.; Lapinski, M.; Blanc, P.; Nugues, M.M. Performance of innovative materials as recruitment substrates for coral restoration. *Restor. Ecol.* **2022**, *30*, e13625. [[CrossRef](#)]
25. Mohammed, J.S. Applications of 3D printing technologies in oceanography. *Methods Oceanogr.* **2016**, *17*, 97–117. [[CrossRef](#)]
26. Gutierrez-Heredia, L.; Keogh, C.; Reynaud, E.G. Assessing the capabilities of additive manufacturing technologies for coral studies, education, and monitoring. *Front. Mar. Sci.* **2018**, *5*, 278. [[CrossRef](#)]
27. Tarazi, E.; Parnas, H.; Lotan, O.; Zoabi, M.; Oren, A.; Josef, N.; Shashar, N. Nature-centered design: How design can support science to explore ways to restore coral reefs. *Des. J.* **2019**, *22*, 1619–1628. [[CrossRef](#)]
28. de León, E.H.P.; Valle-Pérez, A.U.; Khan, Z.N.; Hauser, C.A. Intelligent and smart biomaterials for sustainable 3D printing applications. *Curr. Opin. Biomed. Eng.* **2023**, *26*, 100450. [[CrossRef](#)]
29. Oren, A.; Berman, O.; Neri, R.; Tarazi, E.; Parnas, H.; Lotan, O.; Zoabi, M.; Josef, N.; Shashar, N. Three-Dimensional-Printed Coral-like Structures as a Habitat for Reef Fish. *J. Mar. Sci. Eng.* **2023**, *11*, 882. [[CrossRef](#)]
30. Frayling, C. Research in art and design. *R. Coll. Art Res. Pap.* **1993**, *1*, 1–5.
31. Shanefield, D.J. *Organic Additives and Ceramic Processing*; Springer: New York, NY, USA, 1995. [[CrossRef](#)]
32. Davies, P.; Evrard, G. Accelerated ageing of polyurethanes for marine applications. *Polym. Degrad. Stab.* **2007**, *92*, 1455–1464. [[CrossRef](#)]
33. Callahan, B.J.; McMurdie, P.J.; Rosen, M.J.; Han, A.W.; Johnson, A.J.A.; Holmes, S.P. DADA2: High-resolution sample inference from Illumina amplicon data. *Nat. Methods* **2016**, *13*, 581–583. [[CrossRef](#)] [[PubMed](#)]
34. Team, R.C. *R: A Language and Environment for Statistical Computing*; R Core Team: Vienna, Austria, 2020.

35. Rognes, T.; Flouri, T.; Nichols, B.; Quince, C.; Mahé, F. VSEARCH: A versatile open source tool for metagenomics. *PeerJ* **2016**, *4*, e2584. [[CrossRef](#)] [[PubMed](#)]
36. Frøslev, T.G.; Kjølner, R.; Bruun, H.H.; Ejrnæs, R.; Brunbjerg, A.K.; Pietroni, C.; Hansen, A.J. Algorithm for post-clustering curation of DNA amplicon data yields reliable biodiversity estimates. *Nat. Commun.* **2017**, *8*, 1188. [[CrossRef](#)]
37. Kahlke, T.; Ralph, P.J. BASTA—Taxonomic classification of sequences and sequence bins using last common ancestor estimations. *Methods Ecol. Evol.* **2018**, *10*, 100–103. [[CrossRef](#)]
38. Gao, X.; Lin, H.; Revanna, K.; Dong, Q. A Bayesian taxonomic classification method for 16S rRNA gene sequences with improved species-level accuracy. *BMC Bioinform.* **2017**, *18*, 247. [[CrossRef](#)]
39. Machida, R.J.; Leray, M.; Ho, S.L.; Knowlton, N. Metazoan mitochondrial gene sequence reference datasets for taxonomic assignment of environmental samples. *Sci. Data* **2017**, *4*, 170027. [[CrossRef](#)]
40. Ratnasingham, S.; Hebert, P.D.N. BARCODING: Bold: The Barcode of Life Data System (<http://www.barcodinglife.org>). *Mol. Ecol. Notes* **2007**, *7*, 355–364. [[CrossRef](#)]
41. Oksanen, J. Vegan: Community Ecology Package. 2010. Available online: <http://vegan.r-forge.r-project.org/> (accessed on 26 August 2023).
42. Caporaso, J.G.; Kuczynski, J.; Stombaugh, J.; Bittinger, K.; Bushman, F.D.; Costello, E.K.; Fierer, N.; Peña, A.G.; Goodrich, J.K.; Gordon, J.I.; et al. QIIME allows analysis of high-throughput community sequencing data. *Nat. Methods* **2010**, *7*, 335–336. [[CrossRef](#)]
43. Wickham, H. *ggplot2: Elegant Graphics for Data Analysis*; Taylor & Francis: Boca Raton, FL, USA, 2016.
44. Bradford, M.M. A Rapid and Sensitive Method for the Quantitation of Microgram Quantities of Protein Utilizing the Principle of Protein-Dye Binding. *Anal. Biochem.* **1976**, *72*, 248–254. [[CrossRef](#)] [[PubMed](#)]
45. Lavy, A.; Eyal, G.; Neal, B.; Keren, R.; Loya, Y.; Ilan, M. A quick, easy and non-intrusive method for underwater volume and surface area evaluation of benthic organisms by 3D computer modelling. *Methods Ecol. Evol.* **2015**, *6*, 521–531. [[CrossRef](#)]
46. Veal, C.J.; Carmi, M.; Fine, M.; Hoegh-Guldberg, O. Increasing the accuracy of surface area estimation using single wax dipping of coral fragments. *Coral Reefs* **2010**, *29*, 893–897. [[CrossRef](#)]
47. Amazon. Amazon.com: Amazon Basics TPU 3D Printer Filament, 1.75 mm, White, 1 kg Spool (2.2 lbs): Industrial & Scientific, 2023. Available online: <https://www.amazon.com/AmazonBasics-Printer-Filament-1-75mm-Spool/dp/B08895Y2CP?th=1> (accessed on 16 July 2023)
48. Randall, C.; Negri, A.; Quigley, K.; Foster, T.; Ricardo, G.; Webster, N.; Bay, L.; Harrison, P.; Babcock, R.; Heyward, A. Sexual production of corals for reef restoration in the Anthropocene. *Mar. Ecol. Prog. Ser.* **2020**, *635*, 203–232. [[CrossRef](#)]
49. Delbeek, J.C. Coral Farming: Past, Present and Future Trends. *Aquar. Sci. Conserv.* **2001**, *3*, 171–181. [[CrossRef](#)]
50. Barton, J.A.; Willis, B.L.; Hutson, K.S. Coral propagation: A review of techniques for ornamental trade and reef restoration. *Rev. Aquac.* **2015**, *9*, 238–256. [[CrossRef](#)]
51. BASF. Elastollan® (TPU)—The Thermoplastic Polyurethane of BASF. 2023. Available online: https://plastics-rubber.basf.com/global/en/performance_polymers/products/elastollan.html (accessed on 16 July 2023).
52. Wangpraseurt, D.; Sun, Y.; You, S.; Chua, S.T.; Noel, S.K.; Willard, H.F.; Berry, D.B.; Clifford, A.M.; Plummer, S.; Xiang, Y.; et al. Bioprinted Living Coral Microenvironments Mimicking Coral-Algal Symbiosis. *Adv. Funct. Mater.* **2022**, *32*, 2202273. [[CrossRef](#)]
53. Wangpraseurt, D.; You, S.; Sun, Y.; Chen, S. Biomimetic 3D living materials powered by microorganisms. *Trends Biotechnol.* **2022**, *40*, 843–857. [[CrossRef](#)]
54. Roger, L.M.; Lewinski, N.A.; Putnam, H.M.; Roxbury, D.; Tresguerres, M.; Wangpraseurt, D. Nanobiotech engineering for future coral reefs. In *One Earth*; Elsevier: Amsterdam, The Netherlands, 2023. [[CrossRef](#)]
55. Peixoto, R.S.; Sweet, M.; Villela, H.D.M.; Cardoso, P.; Thomas, T.; Voolstra, C.R.; Høj, L.; Bourne, D.G. Coral Probiotics: Premise, Promise, Prospects. *Annu. Rev. Anim. Biosci.* **2021**, *9*, 265–288. [[CrossRef](#)]
56. Contardi, M.; Montano, S.; Liguori, G.; Heredia-Guerrero, J.A.; Galli, P.; Athanassiou, A.; Bayer, I.S. Treatment of Coral Wounds by Combining an Antiseptic Bilayer Film and an Injectable Antioxidant Biopolymer. *Sci. Rep.* **2020**, *10*, 988. [[CrossRef](#)] [[PubMed](#)]
57. Damjanovic, K.; Blackall, L.L.; Webster, N.S.; van Oppen, M.J. The contribution of microbial biotechnology to mitigating coral reef degradation. *Microb. Biotechnol.* **2017**, *10*, 1236–1243. [[CrossRef](#)]
58. Reyes, G.; Vega-Coloma, M.; Antonova, A.; Ajdary, R.; Jonveaux, S.; Flanigan, C.; Lautenbacher, N.; Rojas, O.J. Direct CO₂ Capture by Alkali-Dissolved Cellulose and Sequestration in Building Materials and Artificial Reef Structures. *Adv. Mater.* **2023**, *35*, 2209327. [[CrossRef](#)] [[PubMed](#)]

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