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Article

Population Genetics Assessment of the Model Coral Species *Stylophora pistillata* from Eilat, the Red Sea

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Abstract: The successful management of coral reefs necessitates understanding the genetic characteristics of reefs' populations since levels of genetic diversity play a critical role in their resilience, enabling them to withstand environmental changes with greater efficacy. To assess the genetic diversity and connectivity of the widespread Indo-Pacific coral, *Stylophora pistillata*, eight microsatellite loci were employed on 380 tissue samples collected from eight sites along the northern Gulf of Eilat, Red Sea. We documented deviations from the Hardy–Weinberg equilibrium and observed low heterozygosity and high values of expected heterozygosity (0.59 and 0.82, respectively). The relatively high F_{ST} values and STRUCTURE analysis results showed population fragmentation along the short coastline (<12 km). These results signify isolation by distance, low gene flow between most populations, and possible non-random mating. These results are connected to this species' sexual reproduction traits, a brooding coral species with planulae that settle shortly upon release with limited connectivity that are most probably further exacerbated by anthropogenic impacts imposed on Eilat's reefs. This study provides insights into the connectivity and population genetics of *S. pistillata* residing in an urbanized northern Red Sea reef and reinforces the need for better management of the current MPA, employing future active coral reef restoration in the area.

Keywords: *Stylophora pistillata*; microsatellite markers; population genetics; connectivity; gene flow; Gulf of Eilat; reef restoration; climate change; urbanized reef

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1. Introduction

Genetic diversity plays a pivotal role in the processes of speciation and adaptation, influencing not only a specie's resilience but also its ability to withstand catastrophic events and respond to climate change that may otherwise lead to extinction [1–4]. This principle is also valid for coral reefs, one of the most diverse ecosystems on earth, which host over a quarter of all known marine species [5,6] in less than 0.2% of the earth's surface [7]. Scleractinian corals are the keystone species in the coral reefs and are further considered as the ecosystem engineers of coral reefs [8–11]. Corals are extremely sensitive to environmental changes; thus, the continuous anthropogenic impacts and global climate change pressures they face (such as mass bleaching and coral mortality following the heating oceans, decreased levels of calcification due to seawater acidification, habitat degradation, algae/coral phase shifts, overfishing, tourist activities, diseases, and other interacting stressors [12,13]) are leading to the global decline of coral reefs. The ICRE report for 2020 indicates that over half of the world's coral reefs have already been lost, projecting that the anticipated 2 °C increase in seawater temperatures over the next two decades might lead to the loss of over 99% of coral reefs [14]. As coral reefs decline globally, there is a need for better management tools and improved conservation and restoration protocols [15–17]. An important aspect of steering the future of coral reefs involves managing their genetic diversity: the higher the

genetic diversity at the commencement of conservation or restoration ventures, the greater the potential for sustained success over the long term [18–20]. Hence, to ensure the success of any restoration initiative or conservation plan, it is imperative to first evaluate the genetic diversity prior to any conservation and/or rehabilitation action of the targeted species.

The genetic diversity of a species is primarily shaped by genetic drift, selection, and migration [21,22]. As in other marine ecosystems, scleractinian coral populations are theoretically open to gene exchange with distant reefs. Scleractinian corals are sessile organisms and depend on the planktonic movement of gametes and larvae for gene exchange. Since tracking those gametes and larvae is virtually impossible, the population genetic of scleractinian corals relies on studying adult populations rather than their larvae [22–24].

Population genetics studies commonly use the efficient molecular markers of microsatellite loci [25], and this is also the case with corals [26]. To analyze the population genetics of coral species along the Israeli coast of the Gulf of Eilat, northern Red Sea, we employed eight microsatellite markers on the 'smooth cauliflower' coral *Stylophora pistillata* (Esper 1797) (Figure 1). This branching coral is a common and widespread Indo-Pacific species of the family Pocilloporidae [27–29]. While *S. pistillata's* sexual reproduction traits have been thoroughly studied in the northern Gulf of Eilat [27,28], little is known about its population genetics [30,31]. In this research, we endeavor to explore the population genetics properties of this species by collecting samples from a wide range of sites along the Israeli coast of the Red Sea and, for the first time, also from shallow- and deep-water sites. We hope that this initiative might shed light on the coral population genetics in the gulf, which can then be translated into a better understanding and improved management practices of the urbanized coral reefs in the Gulf of Eilat and used to harness the ecological engineering properties of this species [28,32] for the 'reef of tomorrow'.

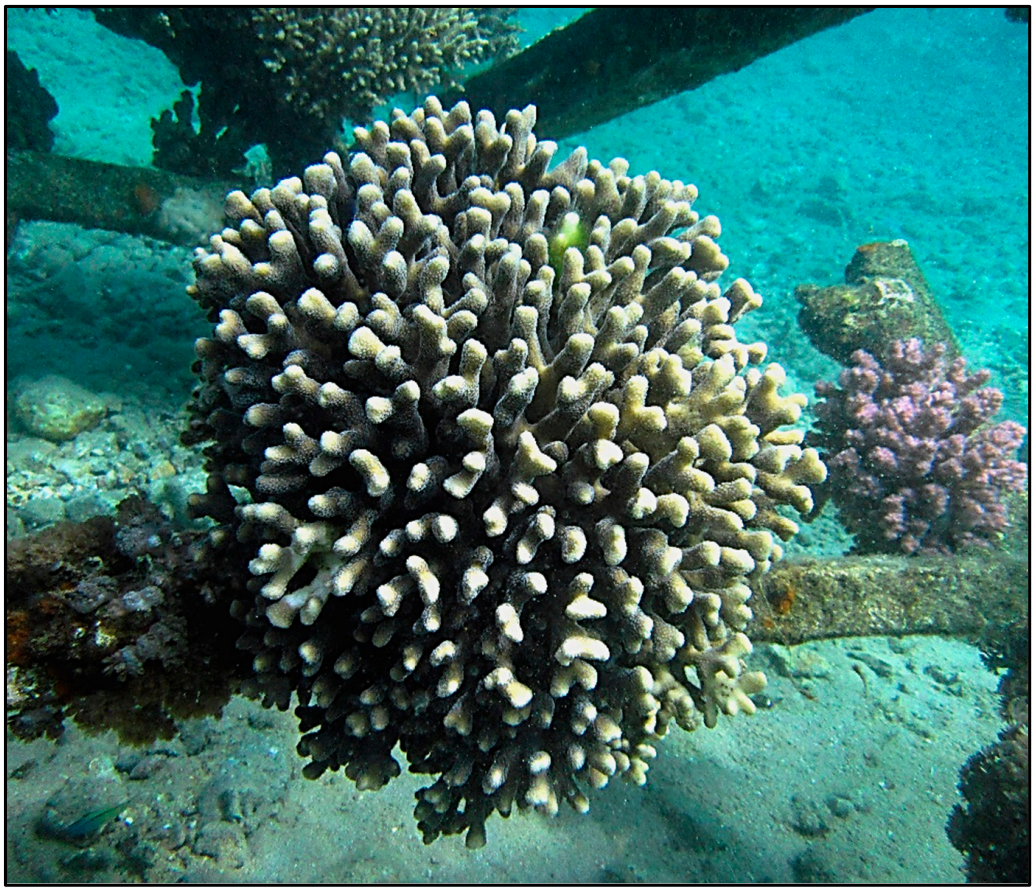

Figure 1. *Stylophora pistillata* (Esper 1797) colonies in the Gulf of Eilat.

2. Materials and Methods

2.1. Stylophora Pistillata Sampling

Tissue samples of 380 *Stylophora pistillata* colonies were collected by SCUBA diving from 8 reef sites along the Israeli coast in the Gulf of Eilat (Figure 2 and Table 1 for site locations, depths, number of samples collected at each site, and collection dates), in accordance with the Israeli Nature and National Parks Protection Authority permits 2016/41540, 2020/42540, and 2021/42878. A single branch tip (about 1 cm^3) was clipped off, using electrician clippers, from each sampled large coral colony (>15 cm in diameter) and placed in a 5 mL plastic vial submerged in seawater. Each colony was sampled only once, and the next sampled colony was haphazardly chosen at least 4 m away. The collected samples were brought to the laboratory at the InterUniversity Institute (IUI) for marine sciences in Eilat, and each sample underwent a brief drying process on dry paper towels for less than a minute and was then individually placed in a new 1.5 mL vial containing a solution of 200 µL of lysis buffer (0.25 M Trisborate pH 8.2, 0.1 M EDTA, 2% SDS and 0.1 M NaCl), 40 µL of 5 M sodium perchlorate, and 240 µL of phenol:chloroform:isoamyl alcohol (25:24:1) [33] and stored at 4 °C.

Figure 2. Sampling sites along the Gulf of Eilat's coastline: Jordan Border beach (JB), North Shore beach (NB), Kisoski beach (KI), Dekel beach shallow (DES), Dekel beach deep (DED), Tour Yam beach (TY), Underwater Observatory beach (UO), Lighthouse beach shallow (LIS), Lighthouse beach deep (LID), and Egypt Border beach (EB). *—Study area.

Table 1. Collection sites in the Gulf of Eilat (site I.D., name of site, geographic location, and depth), collection dates, and the number of coral fragments collected at each site.

Site I.D.	Site Name	Site Location	Depth (Meters)	Collection Dates	Number of Samples Collected
JB	Jordan Border beach	29°32′34.08″ N 34°58′25.02″ E	8–12	27–28 February 2022	30
NB	North Shore beach	29°32′46.89″ N 34°57′59.31″ E	8–12	1–2 March 2022	40
KI	Kisoski beach	29°32′49.85″ N 34°57′13.72″ E	8–12	1–2 March 2022	40
DES	Dekel beach shallow	29°32′18.97″ N 34°56′46.30″ E	8–12	3 March 2022	40
DED	Dekel beach deep	29°32′18.97″ N 34°56′46.30″ E	26–40	3 March 2022	40
TY	Tour Yam beach	29°31′0.03″ N 34°55′36.45″ E	8–12	27–28 February 2022	30
UO	Underwater Observatory beach	29°30′12.57″ N 34°55′7.55″ E	8–12	27–28 February 2022	40
LIS	Lighthouse beach shallow	29°30′2.76″ N 34°54′58.89″ E	8–12	9 November 2022	44
LID	Lighthouse beach deep	29°30′2.76″ N 34°54′58.89″ E	26–40	9 November 2022	46
EB	Egypt Border beach	29°29′38.07″ N 34°54′23.52″ E	8–12	27–28 February 2022	30

2.2. Genomic DNA Extraction

The samples were transported in a cooler box with ice packs from Eilat to the Israel Oceanographic and Limnological Research Institute (IOLR) in Haifa, Israel. DNA extraction from each coral sample followed the protocols outlined by Graham [34] and Douek et al. [33] of phenol/chloroform extraction and ethanol precipitation. The integrity and the quantity of each genomic DNA were assessed using a NanoDrop 2000 (Thermofisher Scientific, Inc., Pittsburgh, PA, USA). Samples were held at 4 °C until further analyses.

2.3. Microsatellite Amplification and Analysis

Microsatellite alleles (developed for *Stylophora* sp. from the Red Sea [35]) for each DNA sample were amplified according to [35] in 11 µL of the total volume containing 2 µL of the DNA sample (diluted 1:50 with DDW, 50–100 ng/µL), 0.1 µL of the two primers, forward and reverse mix (different dyes were used for different reactions, Table 2), 3.9 µL of DDW, and 5 µL of ready-to-use commercial *Taq* polymerase mixture (2× *Taq* PCR Master-Mix (Tiangen Biotech, Beijing, China). The conditions for the PCR cycles (in Mastercycler X50s thermocycler, Eppendorf, Hamburg, Germany) were an initial denaturation step at 95 °C (5 min), followed by 25 cycles at 94 °C (30 s), 55 °C (90 s), and 72 °C (60 s), and a final extension at 60 °C for 30 min [27,35]. The fluorescent-labeled PCR products were examined in an agarose gel (1.5%). Positive PCR products were prepared for analyses by mixing 0.25 µL of each amplification product labeled with VIC, FAM, NED, and PET, and 0.4 µL of LIZ size standard (MapMarker DY632, 50–500 bp, BioVenture Inc. Murfreeboro, TN, USA) was added, as well as 8.6 µL of HiDi Formamide (Thermofisher Scientific, Inc., Pittsburgh, PA, USA). Samples were analyzed off site (ABI PRISM 3100 Genetic Analyzer, Applied Biosystems, Foster City, CA, USA); to the DNA Sequencing Facility, Department of Biochemistry, University of Cambridge, Cambridge UK). The chromatograms of the fluorescent amplification products were scored and binned at the IOLR using the genotyping software Geneious Prime version 2023.2 with the Microsatellites Plugin [36,37].

Table 2. Chosen microsatellite loci from [35], their fluorescence labels used, and size range for the Israeli *Stylophora pistillata* populations.

Locus	Fluorescent Label	Size Range–Eilat
Stylo_17	6-FAM	110–230
Stylo_45	PET	210–340
Stylo_48	PET	210–340
Stylo_55	VIC	220–360
Stylo_72	6-FAM	30–165
Stylo_73	NED	110–280
Stylo_80	VIC	140–270
Stylo_82	NED	170–300

2.4. Data Analysis

We used GenAlEx software version 6.5 [38,39] to calculate the number of alleles, frequencies, observed heterozygosity (H_O), and expected heterozygosity (H_E). The number of private alleles, genetic differentiation among populations (F_{ST}), and the fixation index (F) were established and used to investigate the deviation from the Hardy–Weinberg equilibrium (HWE). A Mantel test for pairwise geographic distance between populations versus pairwise genetic distance (F_{ST}) was employed to evaluate isolation by distance. To assess the significance of all estimates, we employed an analysis of molecular variance (AMOVA) utilizing 999 random permutations. This approach was used to quantify the proportion of genetic variation between locations. The genotypic structure analysis of the population was conducted using STRUCTURE software version 2.3.4 (July 2012) [40] with the following defining parameters: each run consisted of 100,000 iterations with a burn-in of 100,000 for each value of K, from K = 1–20, and with randomize turned off. The results were entered into STRUCTURE Harvester web version 0.6.94 (July 2014) [41], estimating the most likely value of K. BAPS software version 5.3 (October 2009) [42] was used for a Bayesian analysis of population structure to establish genetic distances and relationships between populations. A factorial analysis of correspondence (FAC) for relationships between populations was carried out with GENETIX software version 4.05 (January 2004) [43]. A dendrogram describing the genetic distances and relationships between the coral populations was generated using POPTREE2 software (December 2013) based on distance (D_A) [44] and refined using MEGA11 software version 11.0.13 (June 2022) [45]. CERVUS software version 3.0.7 [46] was used to produce polymorphic information content (PIC) analysis and to calculate the significance of deviation from the Hardy–Weinberg equilibrium.

3. Results

Each of the 380 *Stylophora pistillata* DNA samples was PCR amplified using eight fluorescent microsatellite markers. Analyses of all PCR products (eight per sample, 380 samples) from all collection sites showed high allelic diversity, ranging from 24 alleles (locus Stylo_72) to 61 (locus Stylo_45). Observed heterozygosity (H_O) values for each of the eight loci ranged from 0.43 to 0.73, and the overall value was 0.59 ± 0.02 (mean ± SE; Table 3). Expected heterozygosity (H_E) at the eight loci ranged from 0.78 to 0.87, and the overall value was 0.82 ± 0.01 (Table 3). The fixation index (F) for each locus ranged from 0.08 (Stylo_80) to 0.46 (Stylo_48) with a total average of 0.28 ± 0.03 (Table 3. Results further revealed that the *S. pistillata* populations diverged from the Hardy–Weinberg equilibrium (Table 3). The polymorphic information content (PIC) values ranged from 0.88 to 0.95 for the eight loci. Analyses of the microsatellite data by population (according to their collection sites) revealed that the Dekel beach shallow site had the highest average number of alleles of all loci per site (14.13 ± 1.29), while the Lighthouse beach shallow site had the lowest value of average number of alleles on all loci, 10 ± 0.93 (Table 4). H_O ranged from 0.46 ± 0.08 (North beach site) to 0.72 ± 0.06 (Dekel beach shallow), averaging 0.59 ± 0.02 at all sites (Table 4). H_E ranged from 0.75 ± 0.03 at the Jorden Border beach to 0.87 ± 0.01 at the Dekel beach shallow site (mean = 0.82 ± 0.01) (Table 4).

Table 3. *Stylophora pistillata* population statistics per locus (from [35]) and for all sampling sites. N—number of individuals tagged by the locus, Na—number of different alleles, H_O—observed heterozygosity, H_E—expected heterozygosity, F —fixation index, PIC—polymorphic information content, and significance of the deviation from the Hardy–Weinberg equilibrium: *** = $p < 0.001$.

Locus	N	Na	H_O	H_E	F	PIC	Significance
Stylo_17	379	49	0.54	0.86	0.37	0.94	***
Stylo_55	379	28	0.47	0.79	0.41	0.88	***
Stylo_82	379	28	0.67	0.83	0.2	0.88	***
Stylo_48	379	29	0.43	0.79	0.46	0.85	***
Stylo_72	380	24	0.63	0.78	0.18	0.85	***
Stylo_80	380	34	0.73	0.79	0.08	0.91	***
Stylo_73	363	48	0.66	0.85	0.25	0.95	***
Stylo_45	379	61	0.65	0.86	0.25	0.94	***
Mean ± SE	377.25 ± 2.04	37.63 ± 4.71	0.59 ± 0.02	0.82 ± 0.01	0.28 ± 0.03	0.9 ± 0.01	

Table 4. Population statistics per sampling site (mean ± SE) for all loci per site. N—number of individuals collected per site, Na—average number of different alleles from all loci per site, I—information index, H_O—observed heterozygosity, H_E—expected heterozygosity, F —fixation index. Jordan border beach (JB), North Shore beach (NB), Kisoski beach (KI), Dekel beach shallow (DES), Dekel beach deep (DED), Tour Yam beach (TY), Underwater Observatory beach (UO), Lighthouse beach shallow (LIS), Lighthouse beach deep (LID), and Egypt Border beach (EB).

Pop	N	Na	I	H_O	H_E	F
JB	30	8.5 ± 0.57	1.68 ± 0.09	0.65 ± 0.06	0.75 ± 0.03	0.13 ± 0.06
NB	40	11.75 ± 1.67	1.98 ± 0.18	0.46 ± 0.08	0.81 ± 0.04	0.46 ± 0.09
KI	40	12.25 ± 1.24	2.02 ± 0.12	0.6 ± 0.08	0.81 ± 0.03	0.27 ± 0.1
DES	40	14.13 ± 1.29	2.28 ± 0.09	0.72 ± 0.06	0.87 ± 0.01	0.18 ± 0.06
DED	40	13 ± 1.27	2.23 ± 0.13	0.66 ± 0.05	0.86 ± 0.02	0.23 ± 0.05
TY	30	11.88 ± 1.11	2.13 ± 0.1	0.66 ± 0.04	0.85 ± 0.02	0.22 ± 0.05
UO	40	12.5 ± 1.21	2.14 ± 0.11	0.54 ± 0.08	0.84 ± 0.02	0.36 ± 0.09
LIS	44	10 ± 0.93	1.79 ± 0.11	0.52 ± 0.11	0.78 ± 0.03	0.3 ± 0.17
LID	46	12.38 ± 0.89	2.15 ± 0.09	0.48 ± 0.09	0.85 ± 0.02	0.44 ± 0.1
EB	30	11.63 ± 0.84	2.05 ± 0.11	0.68 ± 0.04	0.82 ± 0.03	0.18 ± 0.04
Total	37.7 ± 0.61	11.8 ± 0.38	2.05 ± 0.04	0.59 ± 0.02	0.82 ± 0.01	0.28 ± 0.03

Pairwise F_{ST} values performed on collection sites ranged from 0.037 between the shallow and deep sites at the Dekel beach to 0.205 between the North Shore beach and the Aqaba Border sites. The average for all pairwise analyses was 0.0998 (Table 5). The variance calculated by AMOVA was 0.38 (10%) among populations and 3.341 (90%) within populations. In total, F_{ST} based on the standard permutation across the full dataset was 0.102 and was significantly different among populations ($p < 0.001$) (Table 6). A Mantel test for pairwise geographic distance between populations versus pairwise genetic distance (F_{ST}) was not significant ($p \leq 0.08$), with an R^2 of 0.0358 (Supplementary Figure S1).

Assessing the likelihood of assigned alleles as originating from the same site population or other populations showed a high chance of 'self-site' assignment (87%; Table 7). North Shore beach had 100% 'self-site' assignment, while the Egypt Border site had the lowest 'self-site' assignment (57%; Figure 3, Table 7).

A 3D factorial analysis of correspondence (FAC) carried out with GENETIX showed three main clusters: one consisting only of the North beach, the second encompassing the two sampling depths at the Dekel beach site, and the third entailing all other sites with some internal differentiation and zonation to each sampling site (Figure 4).

Table 5. Pairwise population F_{ST} values for the 10 Eilat sampling sites. Jordan Border beach (JB), North Shore beach (NB), Kisoski beach (KI), Dekel beach shallow (DES), Dekel beach deep (DED), Tour Yam beach (TY), Underwater Observatory beach (UO), Lighthouse beach shallow (LIS), Lighthouse beach deep (LID), and Egypt Border beach (EB).

	JB	NB	KI	DES	DED	TY	UO	LIS	LID	EB
JB	0.000									
NB	0.205	0.000								
KI	0.093	0.180	0.000							
DES	0.089	0.140	0.082	0.000						
DED	0.108	0.152	0.104	0.037	0.000					
TY	0.118	0.158	0.108	0.047	0.061	0.000				
UO	0.096	0.165	0.039	0.066	0.085	0.068	0.000			
LIS	0.106	0.195	0.074	0.105	0.121	0.117	0.066	0.000		
LID	0.135	0.158	0.100	0.084	0.073	0.061	0.069	0.100	0.000	
EB	0.080	0.170	0.095	0.052	0.069	0.046	0.052	0.076	0.086	0.000

Table 6. AMOVA results performed on all *Stylophora pistillata* populations. SS—sum of squares, Est. Var.—estimated variance, %—percentage of total variance. F_{ST} based on the standard permutation across the full dataset was 0.102, $p < 0.001$.

Source	*df*	SS	MS	Est. Var.	%
Among Pops	9	289.698	32.189	0.38	10
Within Pops	750	2505.621	3.341	3.341	90
Total	759	2795.318		3.721	100

Table 7. Summary of population assignment outcomes to the 'Self-site' or 'Other sites' population employed on the 10 sampling sites: Jordan Border beach (JB), North Shore beach (NB), Kisoski beach (KI), Dekel beach shallow (DES), Dekel beach deep (DED), Tour Yam beach (TY), Underwater Observatory beach (UO), Lighthouse beach shallow (LIS), Lighthouse beach deep (LID), and Egypt Border beach (EB).

Population	'Self'	'Other'
JB	29	1
NB	40	
KI	33	7
DES	35	5
DED	38	2
TY	27	3
UO	27	13
LIS	41	3
LID	44	2
EB	17	13
Total	331	49
Percent	87%	13%

The clustering of individual plots by STRUCTURE suggests the presence of four distinct genetic clusters with a K value of 4, as suggested by Structure Harvester. In this arrangement, JB, KI, UO, and LIS formed one clustered population. Another clustered population encompassed TY, LID, and EB sites, while North Shore beach is considered as a population on its own. Both deep and shallow water samples at the Dekel beach site clustered together as the fourth population (Figure 5, top). BAPS clustering of individual plots showed an optimal K value of seven populations where both sampling depths at the Dekel beach site were clustered together as one population, Kisosky beach and Underwater Observatory beach were clustered together as one population, and Tour Yam beach and Egypt Border beach formed an additional population. JB, NB, LIS, and LID were each considered as a separate population (Figure 5, bottom).

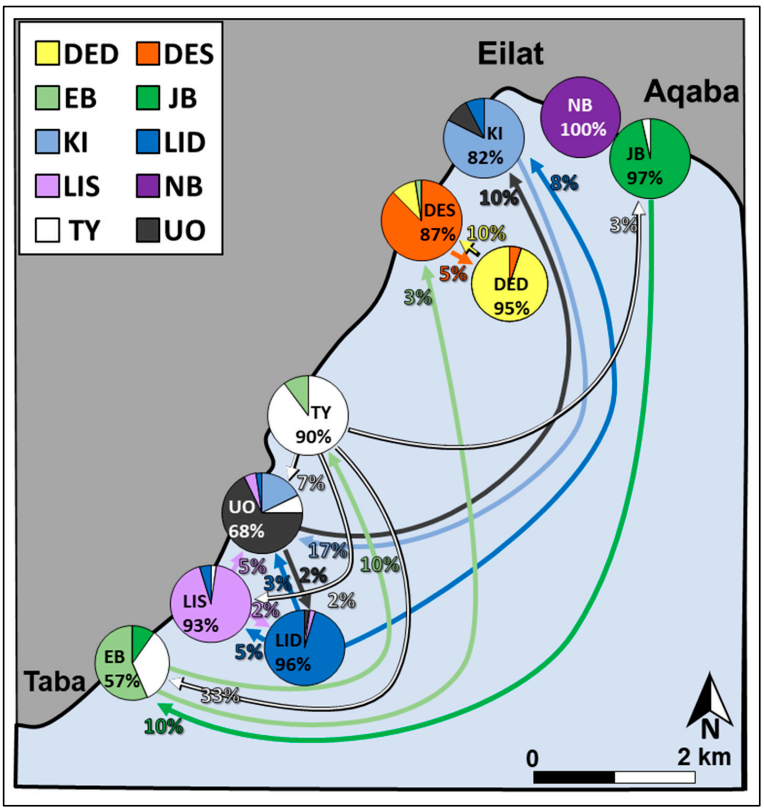

Figure 3. Gene flow trajectories for each sampling site [%], assessing the likelihood of the gene pool (microsatellite alleles) to be the 'self-site' or 'other-site' population. Arrows indicate gene flow from one site to the others. Numbers are percentage [%] of contribution. Values for all 10 sampling sites of *Stylophora pistillata* in the Gulf of Eilat, Israel: Jordan Border beach (JB), North Shore beach (NB), Kisoski beach (KI), Dekel beach shallow (DES), Dekel beach deep (DED), Tour Yam beach (TY), Underwater Observatory beach (UO), Lighthouse beach shallow (LIS), Lighthouse beach deep (LID), and Egypt Border beach (EB).

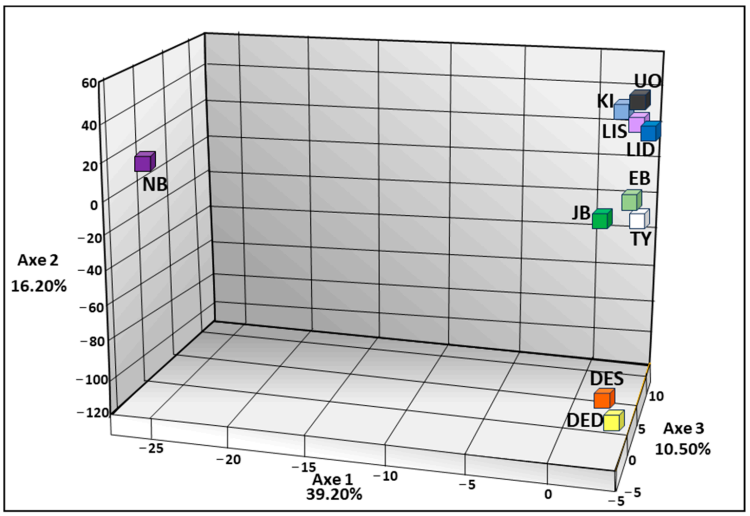

Figure 4. A 3D factorial analysis of correspondence (FAC) of *Stylophora pistillata* populations in the Gulf of Eilat, Israel, generated by GENETIX on the sampled individuals per site. Jordan Border beach (JB), North Shore beach (NB), Kisoski beach (KI), Dekel beach shallow (DES), Dekel beach deep (DED), Tour Yam beach (TY), Underwater Observatory beach (UO), Lighthouse beach shallow (LIS), Lighthouse beach deep (LID), and Egypt Border beach (EB).

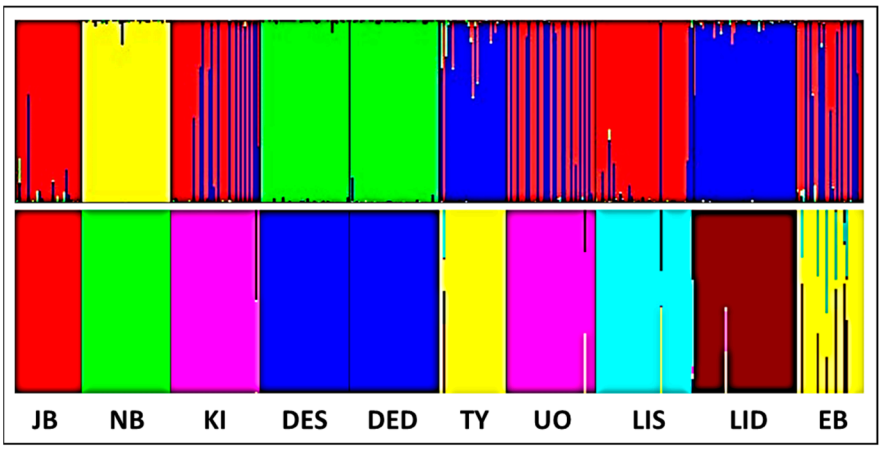

Figure 5. Cluster analysis for all 10 *Stylophora pistillata* sampling sites in Eilat, Israel. (**Top panel**) was created by STRUCTURE (optimal K = 4) and (**bottom panel**) BAPS (optimal K = 7). The y-axis indicates the allocation probability of each sampling site into a distinct cluster, as indicated by the assigned colors. Sampling sites are shown on the x-axis. Jordan Border beach (JB), North Shore beach (NB), Kisoski beach (KI), Dekel beach shallow (DES), Dekel beach deep (DED), Tour Yam beach (TY), Underwater Observatory beach (UO), Lighthouse beach shallow (LIS), Lighthouse beach deep (LID), and Egypt Border beach (EB).

POPTREE2 generated a dendrogram illustrating the genetic distances and relationships among populations through distance (Da) analysis. According to this dendrogram, North Shore beach emerged as a distinct population when compared to all other sampling sites. The shallow- and deep-water sampling depths at Dekel beach represented a separated population from all other sites, which further clustered into two additional groups, the first consisted of Kisoski beach and Underwater Observatory beach together, as compared to the external branch of the Lighthouse beach (both shallow- and deep-water sites) that emerged as an external group. The second group consists of the Egypt Border beach and Tour Yam beach together with Jordan Border beach as a separate population (Supplementary Figure S2).

4. Discussion

In this study, we analyzed 380 tissue samples of adult *Stylophora pistillata* colonies collected from eight locations along the whole Israeli coastal reef in the northern Gulf of Eilat, Red Sea (a 12 km coastline from the Jordanian border to the Egyptian border, a highly urbanized coral reef area). DNA samples were amplified for eight microsatellite loci to create a dataset that characterizes the basic population genetics of *S. pistillata* in the region. While focusing only on the Israeli reefs at the Gulf of Eilat, analyzing the dataset revealed relatively low observed heterozygosity (H_O) against high expected heterozygosity (H_E), implying a non-stable distribution of genetic variation in all studied *S. pistillata* populations over generations, some of which are probably related to non-random mating. Clearly, additional work is needed for the whole Gulf of Eilat evaluations, including sites from Jordan, Egypt, and Saudi Arabia, to increase the dataset and improve and strengthen the conclusions.

Our results revealed that the entire studied *S. pistillata* populations along the 12 km of the coastal contour of the Israeli coast are sub-divided into several isolated populations. This surprising outcome is further supported by the positive F values, indicating low gene flow between most populations, further bolstering isolation by short distances of several hundred meters to a few kilometers, and also by the results supporting high levels of inbreeding within each studied site. The above conclusion is in line with Zvuloni et al.'s [30] outcomes, showing significant genetic differences in the rDNA ITS among two *S. pistillata* populations in the northern gulf, separated by $\leq$10 km. Yet, the above population structure of *S. pistillata* from the most northern part of the Red Sea does not

corroborate with other populations of this widely distributed coral species. For example, Takabayashi et al. [47] have documented panmixia between *S. pistillata* populations in the Western Pacific separated by distances over thousands of kilometers. In addition, Monroe [31] showed no distinctive population structure across the length of the Red Sea (spanning approximately 2000 km of the Saudi Arabian reefs between Maqnah in the north and the Farasan Islands in the south) but showed a greater population structure on a fine scale, suggesting genetic selection based on fine-scale environmental variations.

S. pistillata is a hermaphroditic and brooding coral species, where oocytes are fertilized by sperm originating from neighboring colonies. In many marine invertebrates with such internal fertilization, sperm usually travel limited distances, estimated at a few meters and up to a few hundreds of meters [48–51], increasing the possibility of within site inbreeding. Gravid *S. pistillata* colonies release mature planula larvae almost 8 months/year [27,52,53]. These larvae are short-term swimmers with a limited planktonic phase, where >28% settle within 12 h from release, and the vast majority of settlers (60–80% of released larvae) are recorded within the first 48 h after their release [54,55]. While under laboratory conditions, larvae continue to swim for over a month; only a few larvae continue to settle after the first 2–3 days and within the first week upon release. These biological features lead to a considerable number of planulae that settle near their maternal colonies within the source site and restrict the actual dispersal into a short geographical range, further supporting the significant genetic differences recorded between the sites studied over a short coastline of about 12 km long, and enhanced inbreeding. While modeling larval connectivity and their dispersal in the northern Gulf of Eilat, Berenshtein et al. [56] concluded that the majority of larval supply is within closely situated sites, allowing only a limited flow from north to south trajectory. Berenshtein et al. [56] showed that the movement of planulae was greatly reduced over 20 km of distance, further suggesting the existence of physical barriers that isolate distant sites, a notion further supported by an additional model for coral larvae dispersal [57]. This model further predicts connections between the two sides of the gulf (not studied here) and the possible contributions of reef sites located further to the south [56].

Pairwise F_{ST} analyses showed that the three most northern sites, JB, NB, and KI, which are close to each other, about 0.8–1.2 km apart, significantly differentiate. This result may reflect physical parameters, such as water currents and bathymetry of the area, on top of the continued anthropogenic pressures. The northern part of the Gulf of Eilat depicts a relatively closed water system that is supplied through the straights of Tiran at the south by a chain of eddies along the south-to-north axis [58], creating a relatively mixed sea on an annual scale, and these eddies might trap planula in the direction of a certain areas or cast them out to either into the open sea or toward sandy flats where they cannot settle [59,60]. The bathymetry at the northern tip of the gulf is relatively uniform, composed of relatively sandy flats with very patchy reef modules, which do not offer many settlement sites for planulae [59] when compared to the southern areas that have more pronounced and uniform reef formations [61]. It is evident that the whole northern tip of the Gulf of Eilat is heavily impacted by anthropogenic stressors, such as continuous municipal development, municipal sewage, marine constructions, and other diverse maritime activities (commercial ports, phosphate and nitrogen loading docks, oil platforms, and ship anchoring) and recreational diving pressure [30,62–69]. These anthropogenic impacts cause continuous degradation of the reef [70,71], creating a fragmented urbanized reef [72,73] with high possible impacts on coral population genetics. These proposed anthropogenic and physical conditions at the North Shore beach site might have extreme effects of isolating coral populations in this area from other surrounding populations. At the southern sites (TY, UO, LI, and EB), the *S. pistillata* populations, while still differentiating from each other, exhibit elevated connectivity among them. These results reveal heightened genetic mixing at the more southern sites (TY, UO, LIS, LID, and EB) as compared to the northern sites (JB, NB, KI, DES, and DED; Table 7, Figure 3), which are further mirrored by the lower F_{ST} values between most southern sites (Table 5).

The deep and shallow sites at Dekel beach revealed the lowest pairwise F_{ST} value in this research, which might indicate that these sites are acting as a single population and together are experiencing differentiating forces from the other areas in the region, creating an isolated larger Dekel beach population. On the contrary, when examining the deep and shallow Lighthouse beach sites, the F_{ST} values reveal a significant genetic distinction between these two closely located sampling depths (>100 m apart), indicating restricted gene flow between them. This outcome may be attributed to local currents and reef geomorphology that reduce genetic connectivity between the shallow and the deep sites or to other, undisclosed factors [74], like those linked to distinct symbiont clades harbored in shallow vs. deep water *S. pistillata* corals [75]. Several studies in other reef locations have found that depth, while serving as a divider between deep and shallow reefs, not always cause population genetics differences [24,76,77]. Yet, depth may constitute a barrier to gene flow, and our results may question the assumed roles of mesophotic habitats as refuges [78–80]. While the deep area of the Dekel beach site may contribute larval recruitment to shallower depths, the deep area of the Lighthouse beach site may not, but this requires further research.

Here, we studied one of the most common and widely distributed coral species in the Gulf of Eilat, emphasizing the necessity for comparing its population genetics properties with other common coral species in the region. This should include species with different reproduction strategies (such as broadcasters) in order to clarify spatial scales for conservation and restoration efforts. The results further underscore the importance of gaining a better understanding of the population genetic structures of key species in this urbanized reef. Situated in proximity to the city of Eilat and near Aqaba, Jordan, another major city the coral reef in Eilat has experienced a gradual decline over nearly four decades due to human activities (resulting from heedless development of the city of Eilat, intermittent municipal sewage outflow, industrial and maritime installations, tourism and others [30,62–69]). This degradation of the reef has continued despite the implementation of best practices, such as the creation of an MPA, including a small zone (ca. 350 m of coastline) of a limited access area within it, which has been enforced since 1992 in hopes of creating a refuge area. Some of the assumptions for this conservation strategy are grounded on unrealistic expectations that do not have clear long-term positive impacts in preventing ongoing degradation [62,70,71,81,82]. Now, when considering the broader impacts of global climate change, there is a growing necessity for innovative management approaches, such as active reef restoration [15,83], which can be aided by monitoring the genetic properties of key species [18]. The results of the present study that elucidate a surprisingly fragmented population of one of the most abundant coral species in the gulf offer insights into various ecological engineering approaches to counteract the genetic decline. These approaches may involve initiatives, like the seeding of isolated reef sites with coral planulae using floating nurseries [84], the transplantation of gravid nursery-farmed colonies [85], or the establishment of floating devices to restore biological connectivity through stepping-stone connections [32].

Through our findings, we aspire to contribute to a more profound comprehension of the population genetics dynamics of *S. pistillata* and, consequently, the entire coral reef in the region. Such knowledge may prove instrumental [86] in enhancing management practices for the coral reef in the area, offering potential benefits for innovative strategies toward climate change and anthropogenic adaptation.

Supplementary Materials: The following supporting information can be downloaded at https://www.mdpi.com/article/10.3390/jmse12020315/s1, Supplementary Figure S1. Mantel test for the pairwise geographic distances between populations (*x*-axis) and the pairwise genetic distances (FST) (*y*-axis) for the sampling sites of Stylophora pistillata in the Gulf of Eilat created by GenAlEx software version 6.5. Supplementary Figure S2. The genetic distances and relationships between the Stylophora pistillata populations (sites) in Eilat created by POPTREE2. Jordan Border beach (JB), North Shore beach (NB), Kisoski beach (KI), Dekel beach shallow (DES), Dekel beach deep (DED), Tour Yam beach

(TY), Underwater Observatory beach (UO), Lighthouse beach shallow (LIS), Lighthouse beach deep (LID), and Egypt Border beach (EB). Values at the nodes represent bootstrap values.

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References

1. Barrett, R.D.H.; Schluter, D. Adaptation from Standing Genetic Variation. *Trends Ecol. Evol.* **2008**, *23*, 38–44. [CrossRef]
2. Hoegh-Guldberg, O.; Bruno, J.F. The Impact of Climate Change on the World's Marine Ecosystems. *Science* **2010**, *328*, 1523–1528. [CrossRef] [PubMed]
3. Grünwald, N.J.; Everhart, S.E.; Knaus, B.J.; Kamvar, Z.N. Best Practices for Population Genetic Analyses. *Phytopathology* **2017**, *107*, 1000–1010. [CrossRef] [PubMed]
4. Pearman, P.B.; Broennimann, O.; Aavik, T.; Albayrak, T.; Alves, P.C.; Aravanopoulos, F.A.; Bertola, L.D.; Biedrzycka, A.; Buzan, E.; Cubric-Curik, V.; et al. Monitoring of Species' Genetic Diversity in Europe Varies Greatly and Overlooks Potential Climate Change Impacts. *Nat. Ecol. Evol.* **2024**, *8*, 267–281. [CrossRef]
5. Mulhall, M. Saving the Rainforests of the Sea: An Analysis of International Efforts to Conserve Coral Reefs. *Duke Environ. Law Policy Forum* **2009**, *19*, 321–351.
6. Kinsey, D.W.; Hopley, D. The Significance of Coral Reefs as Global Carbon Sinks—Response to Greenhouse. *Palaeogeogr. Palaeoclimatol. Palaeoecol.* **1991**, *89*, 363–377. [CrossRef]
7. Moberg, F.; Folke, C. Ecological Goods and Services of Coral Reef Ecosystems. *Ecol. Econ.* **1999**, *29*, 215–233. [CrossRef]
8. Tanner, J.E.; Hughes, T.P.; Connell, J.H. Species Coexistence, Keystone Species, and Succession: A Sensitivity Analysis. *Ecology* **1994**, *75*, 2204–2219. [CrossRef]
9. Shafir, S.; Rinkevich, B. The Underwater Silviculture Approach for Reef Restoration: An Emergent Aquaculture Theme. In *Aquaculture Research Trends*; Schwartz, S.H., Ed.; Nova Science Publishers, Inc.: New York, NY, USA, 2008; pp. 279–295, ISBN 9781604562170.
10. Jones, C.G.; Lawton, J.H.; Shachak, M. Organisms as Ecosystem Engineers. *Oikos* **1994**, *69*, 373–386. [CrossRef]
11. Hastings, A.; Byers, J.E.; Crooks, J.A.; Cuddington, K.; Jones, C.G.; Lambrinos, J.G.; Talley, T.S.; Wilson, W.G. Ecosystem Engineering in Space and Time. *Ecol. Lett.* **2007**, *10*, 153–164. [CrossRef]
12. Bindoff, N.L.; Cheung, W.W.; Kairo, J.G.; Arstegui, J.; Guinder, V.A.; Hallberg, R.; Hilmi, N.; Jiao, N.; Karim, M.S.; Levin, L.; et al. Changing Ocean, Marine Ecosystems, and Dependent Communities. Chapter 5. In *IPCC Special Report on the Ocean and Cryosphere in a Changing Climate*; Pörtner, H.-O., Roberts, D.C., Masson-Delmotte, V., Zhai, P., Tignor, M., Poloczanska, E., Mintenbeck, K., Alegría, A., Nicolai, M., Okem, A., et al., Eds.; IPCC: Geneva, Switzerland, 2019.
13. Kleypas, J.; Allemand, D.; Anthony, K.; Baker, A.C.; Beck, M.W.; Hale, L.Z.; Hilmi, N.; Hoegh-Guldberg, O.; Hughes, T.; Kaufman, L.; et al. Designing a Blueprint for Coral Reef Survival. *Biol. Conserv.* **2021**, *257*, 109107. [CrossRef]
14. Souter, D.; Planes, S.; Wicquart, J.; Logan, M.; Obura, D.; Staub, F. (Eds.) *Status of Coral Reefs of the World: 2020*; Global Coral Reef Monitoring Network (GCRMN) and International Coral Reef Initiative (ICRI) Australian Institute of Marine Science: Townsville, Australia , 2021; ISBN 978-1-399-95144-9.
15. Rinkevich, B. Ecological Engineering Approaches in Coral Reef Restoration. *ICES J. Mar. Sci.* **2021**, *78*, 410–420. [CrossRef]
16. Rinkevich, B. Augmenting Coral Adaptation to Climate Change via Coral Gardening (the Nursery Phase). *J. Environ. Manage.* **2021**, *291*, 112727. [CrossRef]

17. Vardi, T.; Hoot, W.C.; Levy, J.; Shaver, E.; Winters, R.S.; Banaszak, A.T.; Baums, I.B.; Chamberland, V.F.; Cook, N.; Gulko, D.; et al. Six Priorities to Advance the Science and Practice of Coral Reef Restoration Worldwide. *Restor. Ecol.* **2021**, *29*, e13498. [CrossRef]
18. Pearman, P.B.; Broennimann, O.; Albayrak, T.; Alves, P.C.; Bertola, L.D.; Biedrzycka, A.; Buzan, E.; Cubric-Curik, V.; Fedorca, A.; Godoy, J.A.; et al. Conserving Genetic Diversity during Climate Change: Niche Marginality and Discrepant Monitoring Capacity in Europe. *bioRxiv* **2023**, bioRxiv:2023.03.24.533448.
19. Jamieson, I.G.; Allendorf, F.W. How Does the 50/500 Rule Apply to MVPs? *Trends Ecol. Evol.* **2012**, *27*, 578–584. [CrossRef]
20. Lacy, R.C. Loss of Genetic Diversity from Managed Populations: Interacting Effects of Drift, Mutation, Immigration, Selection, and Population Subdivision. *Conserv. Biol.* **1987**, *1*, 143–158. [CrossRef]
21. Ayre, D.J.; Hughes, T.P. Climate Change, Genotypic Diversity and Gene Flow in Reef-building Corals. *Ecol. Lett.* **2004**, *7*, 273–278. [CrossRef]
22. Van Oppen, M.J.H.; Gates, R.D. Conservation Genetics and the Resilience of Reef-Building Corals. *Mol. Ecol.* **2006**, *15*, 3863–3883. [CrossRef]
23. Fuchs, Y.; Douek, J.; Rinkevich, B.; Ben-Shlomo, R. Gene Diversity and Mode of Reproduction in the Brooded Larvae of the Coral *Heteroxenia fuscescens*. *J. Hered.* **2006**, *97*, 493–498. [CrossRef]
24. Serrano, X.M.; Baums, I.B.; Smith, T.B.; Jones, R.J.; Shearer, T.L.; Baker, A.C. Long-Distance Dispersal and Vertical Gene Flow in the Caribbean Brooding Coral *Porites astreoides*. *Sci. Rep.* **2016**, *6*, 21619. [CrossRef]
25. Zimmerman, S.J.; Aldridge, C.L.; Oyler-McCance, S.J. An Empirical Comparison of Population Genetic Analyses Using Microsatellite and SNP Data for a Species of Conservation Concern. *BMC Genom.* **2020**, *21*, 382. [CrossRef]
26. Alvarado-Cerón, V.; Muñiz-Castillo, A.I.; León-Pech, M.G.; Prada, C.; Arias-González, J.E. A Decade of Population Genetics Studies of Scleractinian Corals: A Systematic Review. *Mar. Environ. Res.* **2023**, *183*, 105781. [CrossRef]
27. Shefy, D.; Shashar, N.; Rinkevich, B. The Reproduction of the Red Sea Coral *Stylophora pistillata* from Eilat: 4-Decade Perspective. *Mar. Biol.* **2018**, *165*, 27. [CrossRef]
28. Shefy, D.; Rinkevich, B. Stylophora pistillata—A Model Colonial Species in Basic and Applied Studies. In *Handbook of Marine Model Organisms in Experimental Biology*; CRC Press: Boca Raton, FL, USA, 2021; pp. 195–216, ISBN 978-1-00-321750-3.
29. Veron, J.E.N. *Corals of the World*; Australian Institute of Marine Sciences and CRR Qld Pty Ltd.: Townsville, QLD, Australia, 2000.
30. Zvuloni, A.; Mokady, O.; Al-Zibdah, M.; Bernardi, G.; Gaines, S.D.; Abelson, A. Local Scale Genetic Structure in Coral Populations: A Signature of Selection. *Mar. Pollut. Bull.* **2008**, *56*, 430–438. [CrossRef] [PubMed]
31. Monroe, A. Genetic Differentiation across Multiple Spatial Scales of the Red Sea of the Corals *Stylophora pistillata* and *Pocillopora verrucosa*. Master's Thesis, King Abdullah University of Science and Technology, Thuwal, Saudi Arabia, 2015.
32. Horoszowski-Fridman, Y.B.; Brêthes, J.C.; Rahmani, N.N.; Rinkevich, B. Marine Silviculture: Incorporating Ecosystem Engineering Properties into Reef Restoration Acts. *Ecol. Eng.* **2015**, *82*, 201–213. [CrossRef]
33. Douek, J.; Barki, Y.; Gateño, D.; Rinkevich, B. Possible Cryptic Speciation within the Sea Anemone *Actinia equina* Complex Detected by AFLP Markers. *Zool. J. Linn. Soc.* **2002**, *136*, 315–320. [CrossRef]
34. Graham, D.E. The Isolation of High Molecular Weight DNA from Whole Organisms or Large Tissue Masses. *Anal. Biochem.* **1978**, *85*, 609–613. [CrossRef] [PubMed]
35. Banguera-Hinestroza, E.; Saenz-Agudelo, P.; Bayer, T.; Berumen, M.L.; Voolstra, C.R. Characterization of New Microsatellite Loci for Population Genetic Studies in the Smooth Cauliflower Coral (*Stylophora* sp.). *Conserv. Genet. Resour.* **2013**, *5*, 561–563. [CrossRef]
36. Geneious Prime (Biomatters, Auckland, New Zealand) Home Page. Available online: http://www.geneious.com (accessed on 10 January 2024).
37. Kearse, M.; Moir, R.; Wilson, A.; Stones-Havas, S.; Cheung, M.; Sturrock, S.; Buxton, S.; Cooper, A.; Markowitz, S.; Duran, C.; et al. Geneious Basic: An Integrated and Extendable Desktop Software Platform for the Organization and Analysis of Sequence Data. *Bioinformatics* **2012**, *28*, 1647–1649. [CrossRef] [PubMed]
38. Peakall, R.; Smouse, P.E. GenAlEx 6.5: Genetic Analysis in Excel. Population Genetic Software for Teaching and Research—An Update. *Bioinformatics* **2012**, *28*, 2537–2539. [CrossRef] [PubMed]
39. Peakall, R.; Smouse, P.E. GenAlEx 6: Genetic Analysis in Excel. Population Genetic Software for Teaching and Research. *Mol. Ecol. Notes* **2006**, *6*, 288–295. [CrossRef]
40. Pritchard, J.K.; Stephens, M.; Donnelly, P. Inference of Population Structure Using Multilocus Genotype Data. *Genetics* **2000**, *155*, 945–959. [CrossRef] [PubMed]
41. Earl, D.A.; vonHoldt, B.M. STRUCTURE HARVESTER: A Website and Program for Visualizing STRUCTURE Output and Implementing the Evanno Method. *Conserv. Genet. Resour.* **2012**, *4*, 359–361. [CrossRef]
42. Corander, J.; Marttinen, P.; Sirén, J.; Tang, J. *BAPS: Bayesian Analysis of Population Structure. Version 5.3*; Department of Mathematics, Åbo Akademi University: Turku, Finland, 2009.
43. Belkhir, K.; Borsa, P.; Chikhi, L.; Raufaste, N.; Bonhomme, F. *GENETIX 4.05, Logiciel Sous Windows TM Pour La Génétique Des Populations*; Laboratoire Génome, Populations, Interactions, CNRS UMR 5171; Université de Montpellier II: Montpellier, France, 1996.
44. Takezaki, N.; Nei, M.; Tamura, K. POPTREE2: Software for Constructing Population Trees from Allele Frequency Data and Computing Other Population Statistics with Windows Interface. *Mol. Biol. Evol.* **2010**, *27*, 747–752. [CrossRef] [PubMed]

45. Tamura, K.; Stecher, G.; Kumar, S. MEGA11: Molecular Evolutionary Genetics Analysis Version 11. *Mol. Biol. Evol.* **2021**, *38*, 3022–3027. [CrossRef] [PubMed]
46. Kalinowski, S.T.; Taper, M.L.; Marshall, T.C. Revising How the Computer Program CERVUS Accommodates Genotyping Error Increases Success in Paternity Assignment. *Mol. Ecol.* **2007**, *16*, 1099–1106. [CrossRef] [PubMed]
47. Takabayashi, M.; Carter, D.; Lopez, J.; Hoegh-Guldberg, O. Genetic Variation of the Scleractinian Coral *Stylophora pistillata*, from Western Pacific Reefs. *Coral Reefs* **2003**, *22*, 17–22. [CrossRef]
48. Warner, P.A.; Willis, B.L.; Van Oppen, M.J.H. Sperm Dispersal Distances Estimated by Parentage Analysis in a Brooding Scleractinian Coral. *Mol. Ecol.* **2016**, *25*, 1398–1415. [CrossRef]
49. Yund, P.O. An in situ Measurement of Sperm Dispersal in a Colonial Marine Hydroid. *J. Exp. Zool.* **1990**, *253*, 102–106. [CrossRef]
50. Oliver, J.; Babcock, R. Aspects of the Fertilization Ecology of Broadcast Spawning Corals: Sperm Dilution Effects and in situ Measurements of Fertilization. *Biol. Bull.* **1992**, *183*, 409–417. [CrossRef]
51. Grosberg, R.K. Sperm-Mediated Gene Flow and the Genetic Structure of a Population of the Colonial Ascidian *Botryllus schlosseri*. *Evolution* **1991**, *45*, 130–142. [CrossRef]
52. Rinkevich, B.; Loya, Y. The Reproduction of the Red Sea Coral *Stylophora pistillata*. I. Gonads and Planulae. *Mar. Ecol. Prog. Ser.* **1979**, *1*, 133–144. [CrossRef]
53. Rinkevich, B.; Loya, Y. The Reproduction of the Red Sea Coral *Stylophora pistillata*. II. Synchronization in Breeding and Seasonality of Planulae Shedding. *Mar. Ecol. Prog. Ser.* **1979**, *1*, 145–152. [CrossRef]
54. Shefy, D.; Shashar, N.; Rinkevich, B. A Vital Staining Practice That Discerns Ancestry within Groups of Settling Larvae of a Brooding Coral. *J. Mar. Sci. Eng.* **2021**, *9*, 616. [CrossRef]
55. Amar, K.; Chadwick, N.; Rinkevich, B. Coral Planulae as Dispersion Vehicles: Biological Properties of Larvae Released Early and Late in the Season. *Mar. Ecol. Prog. Ser.* **2007**, *350*, 71–78. [CrossRef]
56. Berenshtein, I.; Gildor, H.; Fredj, E.; Amitai, Y.; Berman, H.; Kiflawi, M. *Israel Nature and Parks Authority Report: Connectivity in the Gulf of Aqaba/Eilat*; Israel Nature and Parks Authority: Eilat, Israel, 2018; p. 27.
57. Figueiredo, J.; Baird, A.H.; Connolly, S.R. Synthesizing Larval Competence Dynamics and Reef-scale Retention Reveals a High Potential for Self-recruitment in Corals. *Ecology* **2013**, *94*, 650–659. [CrossRef] [PubMed]
58. Biton, E.; Gildor, H. The General Circulation of the Gulf of Aqaba (Gulf of Eilat) Revisited: The Interplay between the Exchange Flow through the Straits of Tiran and Surface Fluxes. *J. Geophys. Res. Ocean.* **2011**, *116*. [CrossRef]
59. Glassom, D.; Zakai, D.; Chadwick-Furman, N.E. Coral Recruitment: A Spatio-Temporal Analysis along the Coastline of Eilat, Northern Red Sea. *Mar. Biol.* **2004**, *144*, 641–651. [CrossRef]
60. Afargan, H.; Gildor, H. The Role of the Wind in the Formation of Coherent Eddies in the Gulf of Eilat/Aqaba. *J. Mar. Syst.* **2015**, *142*, 75–95. [CrossRef]
61. Katz, T.; Ginat, H.; Eyal, G.; Steiner, Z.; Braun, Y.; Shalev, S.; Goodman-Tchernov, B.N. Desert Flash Floods Form Hyperpycnal Flows in the Coral-Rich Gulf of Aqaba, Red Sea. *Earth Planet. Sci. Lett.* **2015**, *417*, 87–98. [CrossRef]
62. Rinkevich, B. What Do We Know about Eilat (Red Sea) Reef Degradation? A Critical Examination of the Published Literature. *J. Exp. Mar. Biol. Ecol.* **2005**, *327*, 183–200. [CrossRef]
63. Abelson, A.; Shteinman, B.; Fine, M.; Kaganovsky, S. Mass Transport from Pollution Sources to Remote Coral Reefs in Eilat (Gulf of Aqaba, Red Sea). *Mar. Pollut. Bull.* **1999**, *38*, 25–29. [CrossRef]
64. Fishelson, L. Ecology of Coral Reefs in the Gulf of Aqaba (Red Sea) Influenced by Pollution. *Oecologia* **1973**, *12*, 55–67. [CrossRef] [PubMed]
65. Walker, D.I.; Ormond, R.F.G. Coral Death from Sewage and Phosphate Pollution at Aqaba, Red Sea. *Mar. Pollut. Bull.* **1982**, *13*, 21–25. [CrossRef]
66. Kteifan, M.; Wahsha, M.; Al-Horani, F.A. Assessing Stress Response of *Stylophora pistillata* towards Oil and Phosphate Pollution in the Gulf of Aqaba, Using Molecular and Biochemical Markers. *Chem. Ecol.* **2017**, *33*, 281–294. [CrossRef]
67. Rosenberg, Y.; Doniger, T.; Levy, O. Sustainability of Coral Reefs Are Affected by Ecological Light Pollution in the Gulf of Aqaba/Eilat. *Commun. Biol.* **2019**, *2*, 289. [CrossRef] [PubMed]
68. Al-Taani, A.A.; Rashdan, M.; Nazzal, Y.; Howari, F.; Iqbal, J.; Al-Rawabdeh, A.; Al Bsoul, A.; Khashashneh, S. Evaluation of the Gulf of Aqaba Coastal Water, Jordan. *Water* **2020**, *12*, 2125. [CrossRef]
69. Wielgus, J.; Chadwick-Furman, N.E.; Dubinsky, Z. Coral Cover and Partial Mortality on Anthropogenically Impacted Coral Reefs at Eilat, Northern Red Sea. *Mar. Pollut. Bull.* **2004**, *48*, 248–253. [CrossRef]
70. Topel, E.; Lehahn, Y. Long-Term Loss of Coral Reef in the Gulf of Aqaba Estimated from Historical Aerial Images. *Remote Sens.* **2022**, *14*, 6305. [CrossRef]
71. Guerrini, G.; Yerushalmy, M.; Shefy, D.; Shashar, N.; Rinkevich, B. Apparent Recruitment Failure for the Vast Majority of Coral Species at Eilat, Red Sea. *Coral Reefs* **2020**, *39*, 1715–1726. [CrossRef]
72. Chou, L.M. Chapter 10—Sustaining Marine Biodiversity in Singapore's Heavily Urbanized Coast. In *Progress in Sustainable Development*; Ting, D., O'Brien, P., Eds.; Elsevier: Amsterdam, The Netherlands, 2023; pp. 265–282, ISBN 978-0-323-99207-7.
73. Rosenberg, Y.; Simon-Blecher, N.; Lalzar, M.; Yam, R.; Shemesh, A.; Alon, S.; Perna, G.; Cárdenas, A.; Voolstra, C.R.; Miller, D.J.; et al. Urbanization Comprehensively Impairs Biological Rhythms in Coral Holobionts. *Glob. Change Biol.* **2022**, *28*, 3349–3364. [CrossRef]

74. Carlson, R.R.; Crowder, L.B.; Martin, R.E.; Asner, G.P. The Effect of Reef Morphology on Coral Recruitment at Multiple Spatial Scales. *Proc. Natl. Acad. Sci. USA* **2024**, *121*, e2311661121. [CrossRef] [PubMed]
75. Lampert-Karako, S.; Stambler, N.; Katcoff, D.J.; Achituv, Y.; Dubinsky, Z.; Simon-Blecher, N. Effects of Depth and Eutrophication on the Zooxanthella Clades of *Stylophora pistillata* from the Gulf of Eilat (Red Sea). *Aquat. Conserv. Mar. Freshw. Ecosyst.* **2008**, *18*, 1039–1045. [CrossRef]
76. Serrano, X.; Baums, I.B.; O'Reilly, K.; Smith, T.B.; Jones, R.J.; Shearer, T.L.; Nunes, F.L.D.; Baker, A.C. Geographic Differences in Vertical Connectivity in the Caribbean Coral *Montastraea cavernosa* despite High Levels of Horizontal Connectivity at Shallow Depths. *Mol. Ecol.* **2014**, *23*, 4226–4240. [CrossRef]
77. Sturm, A.B.; Eckert, R.J.; Carreiro, A.M.; Klein, A.M.; Studivan, M.S.; Dodge Farelli, D.; Simões, N.; González-Díaz, P.; González Méndez, J.; Voss, J.D. Does Depth Divide? Variable Genetic Connectivity Patterns among Shallow and Mesophotic *Montastraea cavernosa* Coral Populations across the Gulf of Mexico and Western Caribbean. *Ecol. Evol.* **2023**, *13*, e10622. [CrossRef]
78. Slattery, M.; Lesser, M.P.; Brazeau, D.; Stokes, M.D.; Leichter, J.J. Connectivity and Stability of Mesophotic Coral Reefs. *J. Exp. Mar. Biol. Ecol.* **2011**, *408*, 32–41. [CrossRef]
79. Semmler, R.F.; Hoot, W.C.; Reaka, M.L. Are Mesophotic Coral Ecosystems Distinct Communities and Can They Serve as Refugia for Shallow Reefs? *Coral Reefs* **2017**, *36*, 433–444. [CrossRef]
80. Kramer, N.; Eyal, G.; Tamir, R.; Loya, Y. Upper Mesophotic Depths in the Coral Reefs of Eilat, Red Sea, Offer Suitable Refuge Grounds for Coral Settlement. *Sci. Rep.* **2019**, *9*, 2263. [CrossRef]
81. Horoszowski-Fridman, Y.; Rinkevich, B. Active Coral Reef Restoration in Eilat, Israel: Reconnoitering the Long-Term Prospectus. In *Active Coral Restoration: Techniques for a Changing Planet*; J. Ross Publishing: Plantation, FL, USA, 2020; pp. 341–364.
82. Epstein, N.; Vermeij, M.J.A.; Bak, R.P.M.; Rinkevich, B. Alleviating Impacts of Anthropogenic Activities by Traditional Conservation Measures: Can a Small Reef Reserve Be Sustainedly Managed? *Biol. Conserv.* **2005**, *121*, 243–255. [CrossRef]
83. Rinkevich, B. The Active Reef Restoration Toolbox Is a Vehicle for Coral Resilience and Adaptation in a Changing World. *J. Mar. Sci. Eng.* **2019**, *7*, 201. [CrossRef]
84. Amar, K.O.; Rinkevich, B. A Floating Mid-Water Coral Nursery as Larval Dispersion Hub: Testing an Idea. *Mar. Biol.* **2007**, *151*, 713–718. [CrossRef]
85. Horoszowski-Fridman, Y.B.B.; Izhaki, I.; Rinkevich, B. Engineering of Coral Reef Larval Supply through Transplantation of Nursery-Farmed Gravid Colonies. *J. Exp. Mar. Biol. Ecol.* **2011**, *399*, 162–166. [CrossRef]
86. Nadeau, C.P.; Hughes, A.R.; Schneider, E.G.; Colarusso, P.; Fisichelli, N.A.; Miller-Rushing, A.J. Incorporating Experiments into Management to Facilitate Rapid Learning about Climate Change Adaptation. *Biol. Conserv.* **2024**, *289*, 110374. [CrossRef]