

Effect of the use of artificial reefs as substrate on outplanted *Acropora cervicornis* survival and growth in comparison with outplanting on natural reefs



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Abstract:

Knowledge gaps remain regarding the drivers of active coral restoration success that may impede our ability to effectively restore coral reef communities. Publications about ecological development of *Acropora* outplants on natural reefs reveal that long term survival is low and that growth and survivorship is negatively correlated with increased density, competition by other species and sedimentation. Freshly deployed artificial reefs have the potential to relieve corals from some of the stressors by facilitating a clean, competitor free environment. Especially the relieve from the prominent *Dictyota* macro-algae species which we assumed to be a main competitor, might benefit the corals. Here, we conducted a field experiment of one year to investigate growth and survival of nursery-raised *Acropora cervicornis* corals outplants for two types of reefs as substrate: natural patch reefs and originally clean algae-free artificial reefs, and simultaneously investigated the effects of depth and position of attachment to the reefs on these variables.

Significant differences in survival between reef types up to 53 days after outplanting indicated that corals did benefit from clean artificial reef substrate in the early stages. However, after that time, survival of corals on both reef types converged so that no significant differentiation could be made. Final survival after a year was $50\% \pm 10.20\%$ on natural patch reefs, while the survival on the artificial reefs plots decreased to $49.17\% \pm 13.86$. Initial cleanliness of the substrate seemed to be not important to survival in the long run in the current study. However, in combination with other survival improving practices, overcoming initial losses of outplanted corals by using clean artificial substrates has potential to improve survival in the long run.

Instantaneous growth rates of *Acropora cervicornis* were $.00379 \text{ day}^{-1}$ (sd = $.00129 \text{ day}^{-1}$) and $.00636 \text{ day}^{-1}$ (sd = $.00100 \text{ day}^{-1}$) on artificial reefs and natural patch reefs respectively. These rates were affected by all variables under scrutiny. *Place of attachment* within the artificial reefs had the strongest effect, followed by the *reef type* for which confidence intervals were wider. *Depth* was found to have an effect but only very minimal over the small range of 15 to 18.6 meters. *Acropora cervicornis* grew faster on the natural patch reefs. The hypothesis that the absence of an established benthic community on artificial reefs might have a positive effect on coral growth rate cannot be supported. Differential growth rates are possibly induced by differences in water flow rates within reefs and are possibly the combined result of differential flow rates and overall nutrient availability between reef types. More investigation into the drivers for growth is necessary to confidently deduce the most important factors for growth on the two reef types.

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1. Introduction

Since monitoring efforts started in the 1960's, coral communities throughout the Caribbean region have on average shown a steady decline in size (Jackson, J., Donovan, M., Cramer, K., & DeBrot, 2014). While healthy reefs show the capacity to be resilient by resisting or naturally recovering from degradation and as such can maintain ecosystem goods and services (Mumby *et al.*, 2007), many reefs around the Caribbean lack this vital characteristic (Jackson *et al.*, 2014). A mix of chronic and repeated disturbances add to the struggles for natural recovery, not the least being the increased frequency, intensity and severity of mass coral bleaching and extreme weather events that limit time for recovery (Boström-Einarsson *et al.*, 2020). These disturbances, in combination with disrupted supply of corals by stressors affecting their life-cycle (Boström-Einarsson *et al.*, 2020), increase necessity of active reef restoration.

While reef restoration was initially dedicated to shoreline protection, active reef restoration practices with the focus on ecological restoration took a leap in the 1990's (Young *et al.*, 2012). The species largely invested in for assisted recovery of reefs are the reef building branching acroporoid corals *Acropora palmata* and *Acropora cervicornis* (Young *et al.*, 2012). Their branching morphologies contributed largely to reef complexity before their mass mortality in the Caribbean region (Alvarez-filip *et al.*, 2011) due to a cascade of disease and stressors. This and the accompanying mass mortality of the most important herbivore of algae in the early 1980's, the grazing sea urchin *Diadema antillarum*, has resulted in the transformation of reefs from coral- to macroalgal-dominated environments (Cramer *et al.*, 2020; Ware *et al.*, 2020). Characteristics making both coral species particularly suitable candidates for assisted recovery are their high growth rates and their exceptional branching morphologies, providing essential habitat for other reef organisms, in addition to their natural use of fragmentation for asexual reproduction, ability to heal rapidly from wounds, and high survivorship (Young *et al.*, 2012). Despite these advantages and the affiliated big application of these branching corals in restoration programs, articles documenting on ecological development of *Acropora* outplants on natural reefs reveal that long term survival is low (Forrester *et al.*, 2014; Ware *et al.*, 2020), stressing the need for investigation into variables of success of this genus on artificial reefs.

Research into the succes of *Acropora cervicornis* outplants in general reveal that growth and survivorship is negatively correlated with density, competition by other species and sedimentation (Johnson *et al.*, 2011; Ladd *et al.*, 2016). These negative effects can potentially be mitigated by outplanting coral fragments on clean artificial reef structures. There, due to the minimized interspecies competition with an established benthic community and stress, the coral fragments will be more likely to grow and survive (Tanner, 1997). Especially the lack of dominant algae cover may benefit colonization by *Acropora* (McCook, Jompa, & Diaz-Pulido, 2001; Nugues & Bak, 2006; Meltvedt & Jadot, 2014).

Active restoration of corals as well as the use of artificial reefs structures has increased in recent years, but knowledge gaps regarding practices for succes remain (Ladd *et al.*, 2016). Much of the body of knowledge is dedicated to insight into the effect of artificial reefs structures on fish assemblages, limiting the availability of peer-reviewed articles describing ecological development of corals outplanted on artificial reefs (Hylkema *et al.*, 2021). Furthermore, because aims of projects involved in active restoration can be very different, a lot of knowledge about mechanisms underlying the succes of coral outplanting strategies and performance of artificial structures is dispersed or lacking (Ferse *et al.*, 2021; Hylkema *et al.*, 2021). Moreover, the majority of coral outplanting is done on natural reef substrate and additionally, strategies are largely dependend on the location of deployment (Ferse *et al.*, 2021) and are therefore often not generically translatable to other

locations. When monitoring of ecological development of *Acropora* on artificial structures was in place, this was only the case over relatively short timeframes and with other coral species (Ortiz-Prosper *et al.*, 2001), or only artificial reefs were assessed and comparison was made with literature (Cummings *et al.*, 2015).

To add to the body of knowledge regarding variables of success of *Acropora cervicornis* outplants on artificial reef structures, we here compare growth and survival of *A. cervicornis* outplants on artificial reef structures and natural patch reefs within the same experiment. This is, to our knowledge, a practice not yet documented in scientific literature. We hypothesize that *Acropora* outplants have a higher growth and survival rate on artificial reefs compared to natural patch reefs. Furthermore, given that the named species is abundantly targeted for coral propagation and subsequent outplanting, investigation into the effect of such practices might be crucial to the revival of potent coral reefs. The research aim can be divided into two main questions and their respective sub questions:

- a) How does the use of artificial reefs as substrate affect the survival of *Acropora* outplants?
 1. What is the survival rate of *Acropora* outplants on the artificial reefs?
 2. What is the survival rate of *Acropora* outplants on the natural reefs?
 3. Regarding Survival, how do the artificial reefs perform compared to the natural patch reefs for the *Acropora* outplants?

- b) How does the use of artificial reefs as substrate affect the growth of *Acropora* outplants?
 1. What is the growth rate of *Acropora* outplants on the artificial reefs?
 2. What is the growth rate of *Acropora* outplants on the natural reefs?
 3. Regarding growth, how do the artificial reefs perform compared to the natural patch reefs as substratum for the *Acropora* outplants?

2. Methods

2.1 Experimental design

The field site was a sandy area bordering a sloping reef at an approximate depth of 15 meters, and was located offshore of the east coast of Caribbean island of Saba (17°36'51.4"N, 63°13'56.4"W), at a dive location called 'Big Rock Market' (Figure 1).



Figure 1: Map of the wider Caribbean region and of Saba, displaying the location of the experimental setup at Big Rock Market and the nursery location at Ladder Bay.

This sandy area, sheltered by natural reefs, is located in a marine protected area and allowed for the placement of 10 artificial reef structures. These structures were placed along the edge of the natural reef at three meters distance from the edge, with 10 meter intervals (Figure 2).

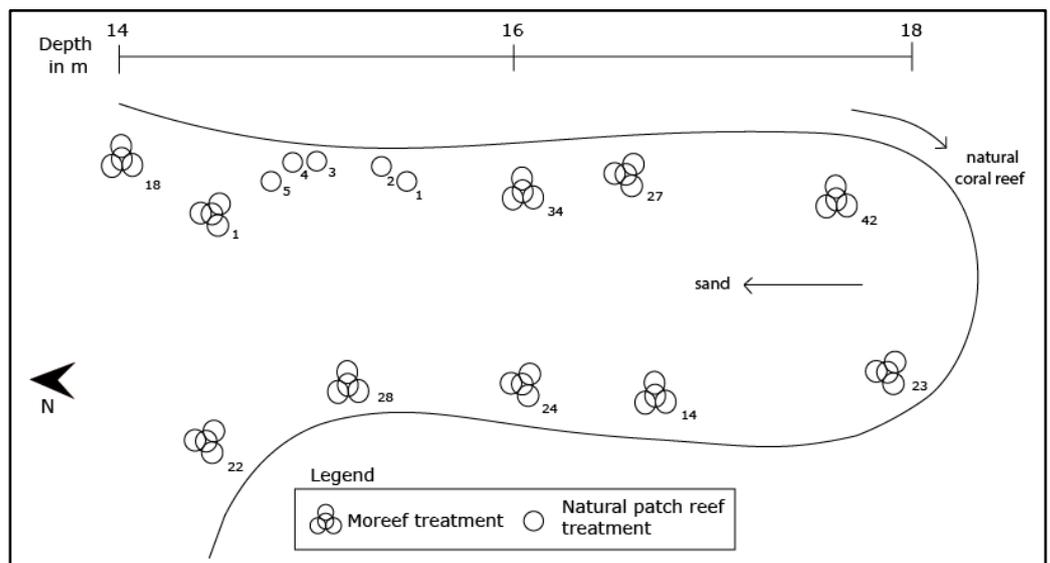


Figure 2: Sketch of location at Big Rock Market, where reefs were located.

Additionally, five natural patch reefs in proximity of these artificial reefs were selected. The surface area of these five small patch reefs was similar in size to the artificial reefs. Artificial reefs used for the experiment are stacks of so-called ‘Moreefs’: modular structures that are designed to provide maximum shelter for reef critters. Each artificial structure consisted of four Moreef modules with three modules on the sandy bottom and one on top of these three (Figure 3). These reefs were created in August 2020 and deployed on the 27th of August, when all reefs were one and a half weeks old at a minimum.



Figure 3: Sideview of modular artificial reefs structure used in the experiment. One module such as the one on the right, is placed on top of three modules at the bottom.

These two reef types, artificial reefs and natural patch reefs functioned as the main treatments in the experiment. Each reef in the experiment received 12 healthy nursery bred *A. cervicornis* fragments, consisting of ideally one branch of approximately 120 mm, so that after attachment branch length would be about 100 mm. The coral nursery is located off the shore at a dive location called ‘Ladder Bay’, roughly four kilometres from the experimental set-up (Figure 1).

Corals were attached to the reefs on the 15th of September 2020, when the artificial reefs were approximately a month old. Corals were attached horizontally to the vertical surface by pouring epoxy in existing or chiselled holes and subsequently putting the base of the coral in these holes. Holes of opportunity were used on the natural patch reefs, while holes on the artificial reefs were of equal depth. Therefore, due to differences in the depth of the holes, not every coral had an initial length at $t=0$ of 100 mm (Table 1). Starting length of corals on the natural patch reefs was therefore significantly smaller than on the artificial reefs (Wilcoxon ranked sum, $W = 5148.5$, $p < .001$).

Table 1. Initial lengths at start of the experiment ($t=0$).

Reef type	Acropora mean length (sd)	Acropora length range
Artificial “Moreef”	96.6 (16.6)	61-142
Natural patch reef	81.3 (18.2)	26-119

On the patch reefs the points of coral attachment were prepared by clearing the algae -primarily seaweeds of the genus *Dictyota*- and sediment-bound turf with scrubbers in a radius of 20 cm around a corals base. For the artificial reef treatment, six corals were divided among the three bottom modules of a reef and six were put on the top module. Corals from each position of attachment received their own marking tie-wrap, so that the attachment position could be taken into consideration in later analyses. For the natural patch reef treatment, six fragments were placed at 40-50 cm from the bottom and six at 80 cm, to get a good comparison with the artificial reefs. We also aimed for vertical substrate. A total of 180 fragments were successfully outplanted on these structures.

2.2 Monitoring

Acropora survival, branch length and health status indicators were monitored for the duration of one year from 25th of September 2020 till 17th of September 2021. Monitoring was done monthly for the duration of half a year after which monitoring was continued in three month periods. Data collection was done via SCUBA-diving and followed a standardized protocol (Appendix I). Information on survival status (dead or alive), breakage, bite marks, disease, and length of each branch was monitored per coral fragment. Bleached corals were not counted as dead yet, because they may recover. As soon as algae settles on the fragment, the coral is not alive anymore and was considered dead. Additionally, all *Acropora* fragments were photographed during the first three monitoring periods to verify measurements and to visibly track the changes in the early stage of the research period. Fragments were also photographed one final time at the end of the experiment. Artificial reef plots and natural patch reefs were examined during every monitoring event regarding any abnormalities that may influence the experiment.

2.3 Data preparation & statistical analyses

Survival (Yes/No) of each coral was summed per reef per monitoring month and divided by the original amount of corals on each reef to come to a survival ratio. Subsequently, this ratio was multiplied by 100 to get a survival percentage per reef per monitoring event. Individual coral length (mm) was constructed by summing the length of the branches per coral per monitoring event and an instantaneous growth rate r was calculated for each coral using log linear regression as described by first order kinetics (Osinga *et al.*, 2011).

Dead corals or corals with obvious impactful breakage, bite marks or disease were not taken into consideration for statistical testing. While these health status indicators were used to subset the data, this information was not explored quantitatively. To give information on reasons behind possible differential instantaneous growth, the number of branches per coral per monitoring event was computed from the data, as well as the time at first branching.

Statistical analyses and graphs construction were performed with R, RStudio version 4.0.4. Survival percentage per treatment failed to meet both ANOVA assumptions of normal distribution and homogeneity. Instead, the Wilcoxon rank sum test using the formula *wilcox.test(paired=F)* was used to test if there were significant differences in survival per monitoring period. An independent sample t-test using the formula *t.test()* was used to test if instantaneous growth rate was different between the patch reef and artificial reef treatment over the 12 month period. P values of $< .05$ were treated as statistically significant. To analyse the bigger observed variability within the artificial reef treatment, and to make inferences on the causality of instantaneous growth rates, a linear mixed model was used with position of attachment, reef depth, and reef type as explanatory variables and with specific reef ID's as added random effect. The independent variable, instantaneous growth rate, showed obvious correlations with coral length at $t=0$, but this variable was disregarded from the model for reasons explained in the results. The effects of the variables under investigation were estimated using the *aov()* and *lmer()* routines in R (Appendix II).

Not necessarily all cases of stunted growth were disregarded. Many cases of stunted growth could well be measurer error on a small scale (e.g. missing tiny new branches of about 5 mm) or could have occurred because decolouration of the colour tags made the observer ascribed a length to the wrong coral. In case stunted growth could be retraced to such occurrences, they were corrected when possible. In case stunted growth was only very minor, for instance because it occurred in an early monitoring period or only affected a tiny branch, the datapoint of that particular monitoring period was excluded from the analysis, but other datapoints were still used. Also, for corals that

experienced considerable breakage early on, datapoints before that moment were disregarded in the growth measurements and the experiment was simply reset. Remaining corals with stunted growth were not used in the analysis of growth and branching, but could be used in the analysis of the survival.

In the analysis the instantaneous growth rates of corals were assumed to be affected by microhabitats from the reef they were on. In this way instantaneous growth rates per reef were assumed to cluster around the a mean per reef instead of solely around the mean of the complete population of corals. Several test also hinted at this behaviour. Normality per reef was tested and found to be normal, and reefs exhibited equal variance (Levene's test, $df=9$, $p=.120$), which allowed for an ANOVA test. When filtering for artificial reefs only, mean instantaneous growth rates of some reefs were found to be significantly different from each other (Anova, $df= 9$, $p=.01$). Additionally, mean instantaneous growth rates of reefs in general are normally distributed around their mean (Shapiro-Wilk, $W=.91$, $p=.158$). These findings underpin the idea that the growth rates tend to cluster per reef and that reefs represent a greater population of reefs. For this reason, data was modelled using a mixed effects model.

3. Results

3.1 Survival

In total, 89 of 180 *Acropora* outplants survived over a monitoring period of approximately 12 months (357 days). Of the outplanted coral fragments 30 of 60 survived on the patch reefs, and 59 of 120 on artificial reef plots. After the 12 month period, this survival amounts to a mean survival on the artificial reef plots of $49.17\% \pm 13.86\%$ and a mean survival on the natural patch reef plots of $50\% \pm 10.20\%$. At the end of the experiment, the survival of *Acropora* fragments was not significantly different between the two treatments (t-test, $df = 10.726$, $p = .898$).

The only significant difference in *Acropora* survival between the treatments was found in the earliest monitoring period (after 25 days), where the survival was higher on the artificial reef plots (Moreef) than on the natural patch reefs (Wilcoxon ranked sum, $W= 42$, $p = .033$) (Figure 4).

After the first month, survival rates on patch reefs visually plateaued till around month six (day 165) (Figure 4). Then a slight drop in survival could be perceived, but a significant difference between survival on the patch reefs at day 165 and day 276 could however not be detected (Wilcoxon ranked sum, $W= 15.5$, $p = .588$). Especially survival on one patch reef, 'Reef 2', was high and stayed high until the ninth monitoring month.

Meanwhile, survival on the artificial reef plots steadily declined over the months. No significance differences in survival between any successive monitoring months were found. Significant differences were however found between the start of the experiment and 3th monitoring month (81 days) (t-test, $df = 9$, $p = .006$), between the 3th and 6th monitoring month (day 81-day 165) (t-test, $df = 17.865$, $p = .028$) , between the 9th and 12th monitoring month (day 276 – day 357) (t-test, $df = 17.207$, $p = .002$), but then none between the 6th and 9th monitoring month (day 165 – day 357) (t-test, $df = 17.944$, $p = .075$).

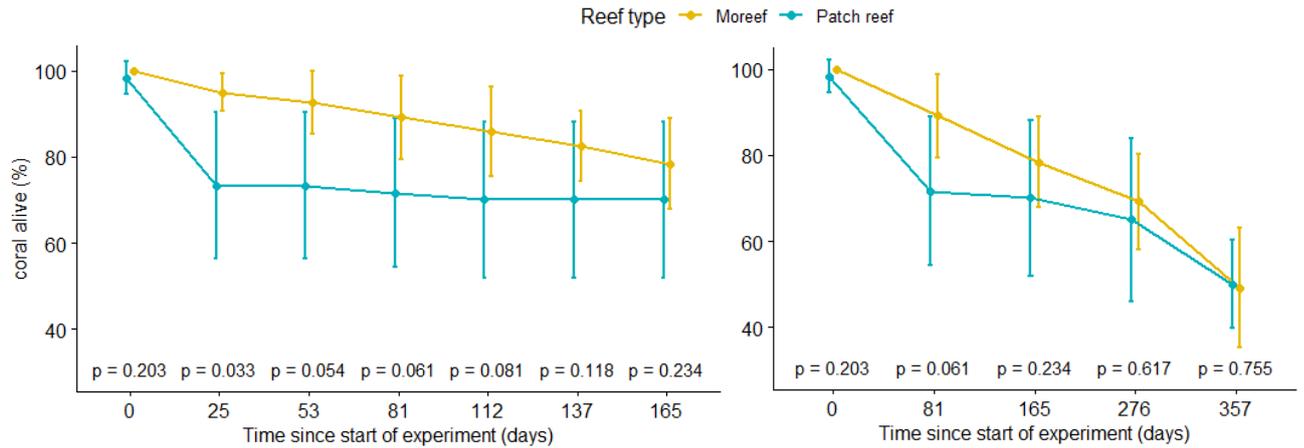


Figure 4: Mean percentage of corals alive of original 12 corals per reef over time. The left graph displays the first six months since the start of experiment. After six months, monitoring was continued every third month. The complete timeframe with observations of every third month is displayed in the right graph for ease of comparing survival rates. Evaluated differences between treatments are displayed as p-values below each observation (test= Wilcoxon ranked sum).

The sharpest percentual drop in survival on the patch reefs occurred at the start of the experiment, where the mean percentage of live corals dropped by 26.67 % in the first month. Overall the highest amount of corals died over the last three months of the experiment (Table 2) ; both patch reefs and artificial reefs experienced relatively low survival rates of .769 and .711. Survival rates for corals on artificial reefs over the successive months are fairly similar, except during the last three months (Figure 4).

Table 2. Absolute coral losses per treatment per monitoring period. Note that the last two monitoring periods display losses accumulated over three months.

Timeframe	Natural patch reefs		Artificial reefs	
	breakage	other	breakage	other
0	1	-	-	-
25	7	8	-	6
53	-	-	1	2
81	-	1	-	4
112	-	1	1	1
137	-	-	1	5
165	-	-	3	2
276	2	1	8	2
357	5	4	15	10

Table 3. Mean survival per treatment per three month period, expressed in percentages and corresponding survival rates over the 3 month periods.

Timeframe	Natural patch reefs		Artificial reefs	
	Mean % survival drop	Survival rate	Mean % survival drop	Survival rate
0-81	26.67	.7288	10.83	.8917
81-165	1.67	.9767	10.83	.8785
165-276	5.00	.9286	9.167	.8830
276-357	15.00	.7692	20.00	.7108

Corals that died during the experiment were significantly smaller at $t=0$ than corals that survived (Wilcoxon ranked sum, $W = 3291.5$, $p = .030$). More smaller fragments ended up on the natural patch reefs, but the same conclusion could be drawn when filtering for corals on artificial reefs only (Wilcoxon ranked sum, $W = 1387.5$, $p = .031$). Of the corals that died, some notoriously small corals (< 55 mm above substrate) died in the first month, but of corals above that length no particular correlation could be found between length at $t=0$ and the month of death ($r(85) = .11$, $p=.33$).

3.2 Growth & branching

Over the 12 month period, corals on artificial reefs on average grew to 393 mm (sd = 146 mm), while corals on patch reefs on average grew to 723 mm (sd = 235 mm) and had one outlier of 1548 mm. An example of the length accumulation is depicted in Figure 5. When clustering corals according to the two reef types only, a significant difference in *Acropora* instantaneous growth rates could be found between the types of reefs when performing a non-parametric alternative to the unpaired two-sample t-test (Wilcoxon ranked sum, $W = 224$, p -value = $<.001$). Instantaneous growth rates on artificial reefs ranged from $.00065 \text{ day}^{-1}$ to $.00689 \text{ day}^{-1}$, with a mean of $.00379 \text{ day}^{-1}$ (sd = $.00129 \text{ day}^{-1}$) and an outlier of $.00749 \text{ day}^{-1}$. Instantaneous growth rates on natural patch reefs ranged from $.00420 \text{ day}^{-1}$ to $.00807 \text{ day}^{-1}$, with a mean of $.00636 \text{ day}^{-1}$ (sd= $.00100 \text{ day}^{-1}$) and one outlier at $.00190 \text{ day}^{-1}$.

However, several factors influence the growth rates of the outplanted corals. First of all, as mentioned in the methods, corals from the same reef were found to have their instantaneous growth rates clustered to a certain extend, underpinning the assumption that each reef has an individual microhabitat affecting growth rates. Analysis of this behaviour was possible when filtering for artificial reefs. While for artificial reefs, the average squared differences from the mean are not found to be statistically dissimilar between reefs (Levene's test, $df=9$, $p=.120$), standard deviations per reef can be considerable. Mean instantaneous growth rates for these artificial reefs vary from $.0029 \text{ day}^{-1}$ to $.0050 \text{ day}^{-1}$, while standard deviations range from $.45 \text{ day}^{-1}$ to 1.65 day^{-1} , informing us that substantial overlap between instantaneous growth rates of corals on different reefs is present. Nevertheless, because of the result of the tests as described in the methods, random variance of the instantaneous growth rate for some



Figure 5: Length accumulation of an *Acropora* fragment on an artificial reef per three month period. This particular coral grew to 591 mm in the 12 month period and started at 137 mm at $t=0$.

random coral is assumed to be influenced by the specific reef and residual unexplored variance. In model form, this random effect of reefs on instantaneous growth rates of individual corals can be written as $Instantaneous\ growth\ rate_{i,j} = \mu + fixed\ effects_i + R_j + \epsilon_{i,j}$, where R_j is the variance component explained by reefs and $\epsilon_{i,j}$ the residual variance.

Several fixed effects thought to influence instantaneous growth rates were investigated. Apart from reef type, also reef depth, place of attachment to the artificial reefs and starting length have been put under scrutiny.

3.3.1 Starting length

While length at first branching is significantly different for corals on natural patch reefs and artificial moreefs (Wilcoxon ranked sum, $W=1386$, $p=.027$), this is most likely an artefact of the method of attachment. Corals chosen to be part of the experiment were measured at 120 mm, but during attachment to the reefs, some ended up deeper in holes than others. This was especially the case for the ones on the patch reefs, so that their average length at $t=0$ ended up being shorter, accounting for the variability in the data (Table 1). When investigating the relation between starting length and length at first branching with a

linear model, for all corals first branching seems to occur after approximately 25 mm of growth, almost completely irrespective of length at $t=0$ (Figure 6). So length at $t=0$ does not seem to play an important role in the growth pattern.

What however does seem important is that this length accretion was achieved at different times after the start of the experiment for corals on both reef types. About 60% of corals on patch reefs grew their first branch already in the second period from the start, while for the moreefs time of first branching was much more spread out (Figure 7).

These observations hint that first branching is originally dependent on growth rate and not vice versa. For that reason, starting length was digarded as fixed effect in the model and the three variables that remained were reef type, reef depth and

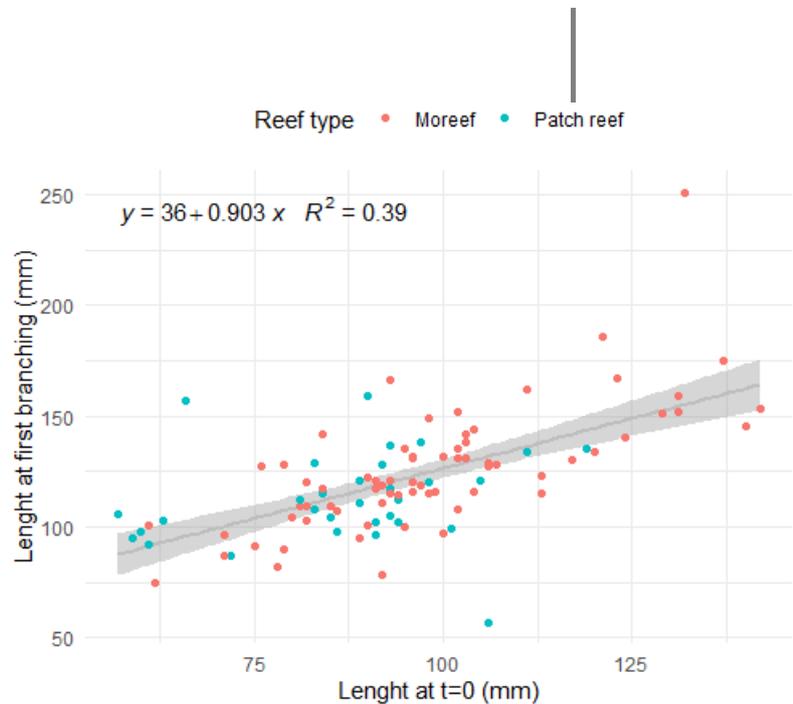


Figure 6: Length at first branching vs length at $t=0$. There is a positive correlation between the two variables. First branching occurs after approximately 25 mm of growth.

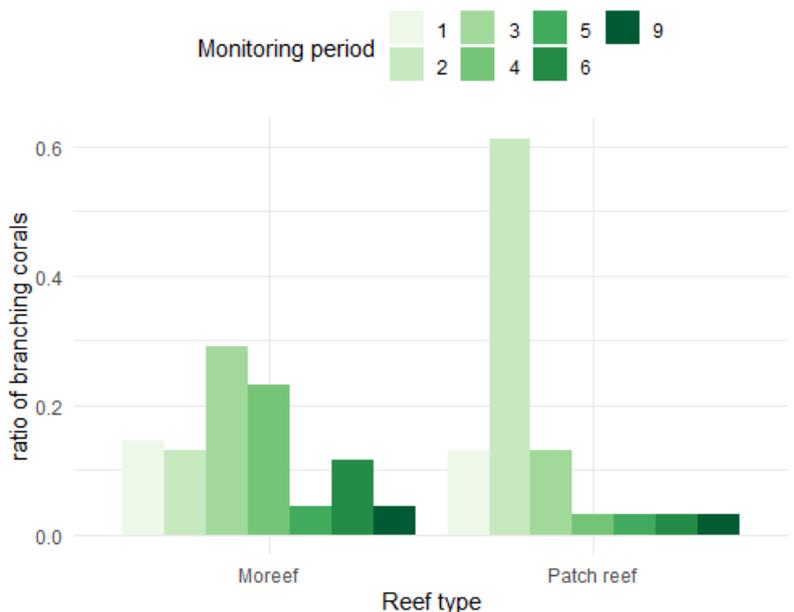


Figure 7: Ratio of total corals having their first branch during a particular monitoring period, separated by reef type. The amount in period 9 sums all corals from period 7, 8 and 9. No coral had its first branch later than period 9.

place of attachment. Parameter values from the resulting model: $Instantaneous\ growth\ rate_{i,j,k,l} = \mu + reef\ type_i + reef\ depth_k + place\ of\ attachment_l + R_j + \epsilon_{i,j,k,l}$, where investigated using the aov() and lmer() routines in R.

3.3.2 Reef type

The variable *reef type* was found to have a significant effect (aov, $p < .001$) on instantaneous growth rate, with natural reefs facilitating higher growth rate. The value of the parameter was estimated at $.0011578\ day^{-1}$ with an estimated standard error of $.00052\ day^{-1}$ (Table 4). This effect can be regarded as a relatively strong effect considering that instantaneous growth rates ranged from $.000651\ day^{-1}$ to $.008070\ day^{-1}$. However, the estimate is quite uncertain; the lower limit of the effect is estimated at $.0001386\ day^{-1}$. Then, when taking the standard deviation of the variance accounted for by the random effect of reefs into consideration, which is $.0005274\ day^{-1}$, the effect can also be relatively unimportant. Overall, the effect appears to be strong, but confidence intervals are wide. We cannot exclude that it is much weaker or stronger.

Table 4. Fixed effect estimates and confidence interval, as well as the random effect standard deviation of the variation induced by the individual reefs.

parameter name	Estimated value	lower-95 confidence limit	upper-95 confidence limit	random effect of reef (SD)
<i>intercept</i>	.0108196	.0077762	.0138630	.0005274
<i>reef type</i>	.0011578	.0001386	.0021770	" "
<i>reef depth</i>	-.0003799	-.0007317	-.0000281	" "
<i>place of attachment</i>	-.0012941	-.0018043	-.0007839	" "

3.3.3 Reef depth

Reef depth varied from 15 meters to 18.6 meters below the surface. The variable was found to have a significant effect on instantaneous growth (aov, $p = .023$). The effect was estimated at $(-.0003799\ day^{-1})m^{-1}$, with an estimated standard error of $.0001795\ day^{-1}$, creating an upper limit of the effect at $(-.00002808\ day^{-1})m^{-1}$ very close to 0. The effect appears to be a weak effect, and while confidence intervals are not particularly wide, its upper limit reveals the effect can be very small. On average the effect is weak, but there is also considerable variation between reefs rendering the effect possibly negligible over the reef depths in the experiment.

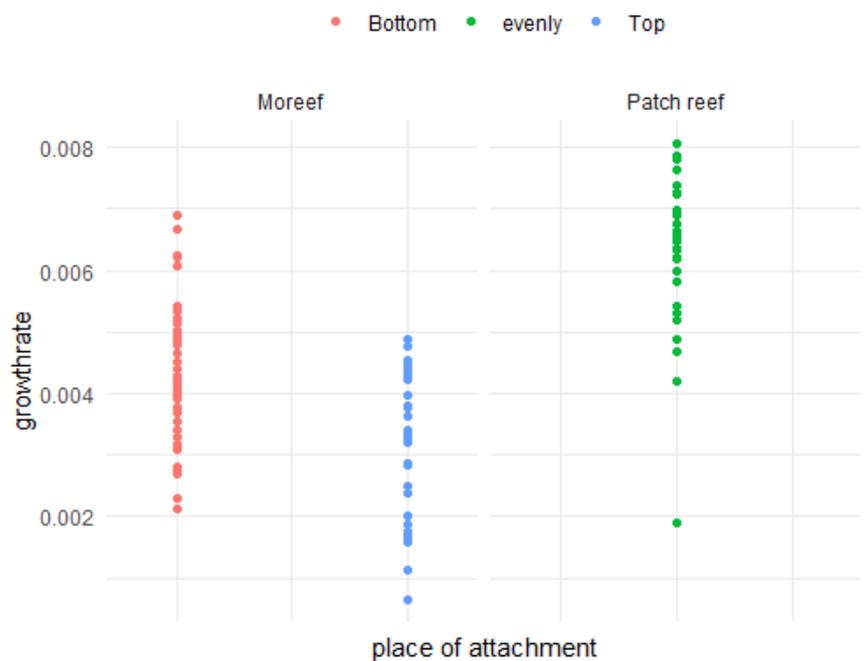


Figure 7: Variation in growth rates for the different places of attachment (Top, Bottom). This distinction was not made for corals on the patch reefs, hence 'evenly'.

3.3.4 Place of attachment

Within reefs *place of attachment* was found to have a significant effect on the instantaneous growth rate of corals (aov, $p < .001$). Its value was estimated $-.0012941\ day^{-1}$ and had an estimated standard

error of $.00026 \text{ day}^{-1}$, making this generally a strong effect. Additionally, its upper and lower limit revealed that even when taking the variation between reefs into consideration, the effect is always higher than 0, concluding that variability in the data is significantly explained by a corals position on the moreef module. The result of the effect can also be seen in Figure 8.

3.3.5 Branching strategy

Another remarkable finding is that differences in instantaneous growth rates coincided with differences in growth strategy. Corals on natural patch reefs branched earlier than ones on the artificial moreefs (Figure 9). This first branching seemed to be the result of faster growth, but after this first branching, length accumulation is amplified because then there are more tips to grow from. The peak of first branching occurred in the second monitoring period for the patch reefs (Figure 7). In this and the following period, a significant difference in the mean number of branches of corals could be detected between patch reefs and moreefs (Figure 9) (t-test, $p = .004$ and $p = .019$ respectively). Then in period 4, 5 and 6, the corals on the moreefs caught up with the ones on the patch reefs, but in period 9 and 12 corals on the patch reefs had a second wave of branching, again driving significant differences in the amount of branches between the reef types (t-test, $p < .001$ and $p = .006$ respectively).

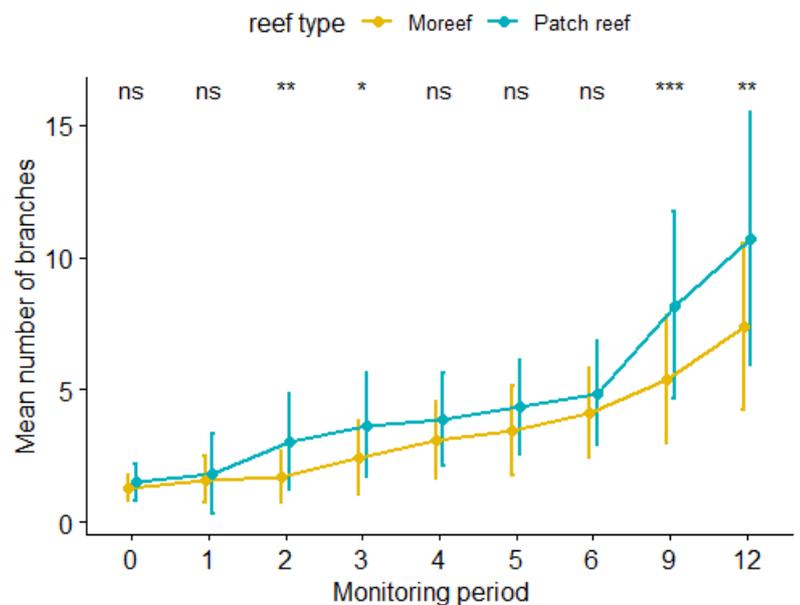


Figure 8: Mean number of branches per reef type over the monitoring periods. Significant differences occur in period 2, 3, 9 and 12.

4. Discussion & Conclusion

4.1 Survival

The type of reef corals are outplanted on seems to have notable impact on both survival and growth. While in the long run survival was the same between the reef types, most corals on natural patch reefs that died, died within 25 days after outplanting. After this period, deaths stagnated on these reefs, indicating that most of the mortality due to the transplantation process occurred early on.

A continually declining trend in survival is observable for corals outplanted onto artificial reef substrate during the period of stagnated death on the natural reefs. Only after percentage survival was relatively similar for both reef types, which was the case in the sixth monitoring period, survival rates became similar again. A similarity was also the sudden and quite synchronous loss over the last three monitoring periods. The survival rate over these months was .762 of the live corals on the patch reefs at the start of these three months and .711 of live corals on the artificial reefs. These rates contrast the previous survival rates, which had been $.884 \pm .067$ per 3 month period up to the ninth month for the artificial reefs. The survival rate on the natural patch reefs had been .929 during the last 3 month period. This decrease in survival rate might indicate a kind of hurdle in these last three months, that was not present in the previous months. There is a possibility that higher sea surface temperatures induced more deaths (Chilcoat, 2004), but we found no sign of bleaching during the experiment.

Other reasons could be more turbulent water in the hurricane season and/or the corals collapsing under their own weight. This thought is underpinned by the observed increase in breakage closer to the end of the experiment. This was especially obvious for corals on the artificial reefs. On these reefs there were eight deaths with breakage against two with other causes in the months 7 till 9, and 15 breakages as opposed to 10 with other causes occurred in the months 10-12. Also, the finding that three weeks after the end of the experiment, at the beginning of October, some additional corals had completely broken off, underpins the hypothesis. Reason for this sudden additional breakage could be the lack of proper natural attachment in time. Indeed It is easy to break when natural attachment is still weak or lacking.

Both regular deaths as well as breakages increased in month 10-12. But since no elaborate monitoring of the types of deaths encountered on the reefs had been performed, these non-breakage deaths cannot be pinpointed exactly to particular events or circumstances, leaving part of reason for the decrease in survival rate unanswered. In any case, further research could investigate a corals' center of gravity or status of natural attachment to find reasons for death by breakage and figure relative importance of this factor on survival.

Over the complete timeframe of the experiment, on average 50% of corals on patch reefs against 49.17% of corals on the artificial reefs survived. Directly making comparisons with literature is however hard because of the wide range of differences between experiments that could have brought about observed survival rate. Forrester *et al.* (2014), who transplanted fragments of *Acropora palmata* to a shallow patch reef (.4m - 1.6 m deep), reported 50% to 85% survival after 12 months for years without major storms. Fisk & Harriott (1995), reported 33% survival of transplanted *Acropora* fragments of 10-30 cm and 13.5% for *Acropora* smaller than 10 cm after a 7 month period. In that same experiment the shape of the survival curve of these smaller fragments closely resembles the survival curve in the current experiment, with a falling off in the mortality rate early after transplantation. The experiment confirmed that small fragments have higher mortality rates. Our data supports a similar finding; corals that died during the experiment were significantly smaller at $t=0$ than corals that survived. Of the corals that died, some notoriously small corals (< 55 mm) died

very early, but of corals above that length no correlation was found with the time of death. The same paper also reports high survival of 71% to 100% for transplanted fragments 7 months after transplantation, but then does not elaborate on fragment lengths. Johnson *et al.* (2011) reported low (10%) to very high (97.4%) survivorship in several case studies with *Acropora cervicornis*, but an elaborate documentation of the conditions on the outplanting site and monitoring timeframe was missing. In the abovementioned papers, corals were attached vertically to a horizontal surface. In the current experiment we attached the corals horizontally to a vertical surface, which might have had a negative impact on survival, for example via weaker natural attachment.

Apart from effects of water turbulence, disease, fragment size, and the effect seasonal temperature, there is also predation, competition and other disturbances by sea creatures, including different levels of corallivory, that could have negatively affected survival of the corals. Water turbulence, disease, and the effect seasonal temperature can be assumed to affect corals on both reef types similarly with the current experimental design. But predation, competition and other disturbances by sea creatures could differ between reef types. The foremost dissimilarity between reef types was the composition of the benthic community and any accompanying life. On the natural patch reefs *Dictyota* sp. were suspected to be the main algae competitors. The initial drop in survival of the patch reefs could have been the result of the combined effect of these stressors together with the stress of outplanting and acclimatization. This line of thinking is endorsed by the finding of Woesik *et al.* (2018), who found that survival of *Acropora cervicornis* was better on places with less *Dictyota* sp. cover. Any negative effect of the algae might have been worsened by small differences in attachment between reef types; on average the corals on patch reefs were placed in deeper holes, leaving their length at $t=0$ shorter than on the artificial reefs. In theory, this might leave them even more vulnerable to competition by the *Dictyota* sp. already present on those reefs, although the perimeter around a corals base was cleaned before attachment. The absence of a steep drop in survival on the artificial reefs at the start of the experiment and the steady survival rate hints that the use of clean artificial substrate is successful in reducing the initial stress corals experience after outplanting. To what extent the *Dictyota* sp. presence was responsible for the initial stress is however impossible to deduce within this study. Factors other than algae coverage that also correlate with the successional stages of a reef make it impossible to partition stressors on these corals into stress resulting from transplantation versus other uncontrolled stressors in the current study.

In conclusion, the use of clean artificial reef substrate does significantly benefit coral survival in the short run, but does not result in significantly better survival in the long run for this type of reef under the given circumstances. From the similarity in survival between reef types after 12 months ($49.17\% \pm 13.86\%$ and $50\% \pm 10.20\%$ for artificial and natural reefs respectively), it seems that coral survival in the long run is less affected by benthic community composition of substrate the coral is standing on, than it is by other factors. Therefore, investigation into other relevant factors of death is necessary to elevate coral survival. The use of artificial structures however is suitable for overcoming the initial loss of outplanted corals. This effect, in combination with other survival improving practices could result in significantly higher survival in the long run on clean artificial structures. This can happen if survival on these structures plateaus at a higher percentage than the percentage survival after the initial drop in survival on natural patch reefs.

4.2 Growth

The analyses revealed that all three parameters under scrutiny had an effect on the instantaneous growth rate. Place of attachment seemed to have the biggest effect within reefs followed by reef type, and reef depth seemed to have a weak to negligible effect. Our primary parameter of interest, reef type, had a notable effect on instantaneous growth rate, however, confidence intervals are

wide. Most importantly, the effect was found to be contrasting our prior beliefs about the positive effects of clean artificial reefs. It was assumed that *Acropora* outplants would have a higher growth on clean artificial reefs compared to natural patch reefs due to a reduction in interspecies competition and stress, mainly that of dominant algae cover on the artificial reef units. But in fact the higher growth rate was experienced by corals on natural patch reefs, despite the ample evidence suggesting that algae presence directly affects coral growth through competition (McCook, Jompa, & Diaz-Pulido, 2001; Nugues & Bak, 2006; Meltvedt & Jadot, 2014) at least in the early stages (Sandin & Mcnamara, 2012; Tamai & Sakai, 2013).

The observation that coral growth on the patch reefs, which had already established benthic communities, was actually higher than on the artificial reefs can mean several things about the effect of competition. Foremost, it could be that competition by algae might not have had a very strong effect on growth rate in the current experiment. Corals did not enter the habitat as complete juveniles but as healthy fragments of about 100 mm. Competitive advantage for corals is size dependent; adults are good competitors for space, but juveniles are particularly susceptible to algal competition via overgrowth and other mechanisms (Ferrari *et al.*, 2012; Sandin & Mcnamara, 2012). Tamai & Sakai (2013), observed that it were mainly encrusting turf algae that limited growth of small *Acropora tenuis* (≈ 2 cm in length), through inhibiting growth of the attachment base, which is a necessity for upward growth. At the start of the current experiment algae in direct proximity around a corals base were removed. Also, the *Acropora* fragments in our experiment were neither juveniles or adults, but they certainly exceeded the lengths mentioned in the study of Tamai & Sakai (2013) and might have surpassed the threshold to upward growth.

The observed early survivorship on natural patch reefs suggests an effect of initial composition of benthic community. If the effect was at least partly induced by competition for space by algae, the effect might just not have induced reduced growth rates. It should be kept in mind that no growth rate could be constructed for corals that died. Corals that had the most effect from stress by competition could have been the same corals that died and as such no information on growth for these corals is available, creating a bias in the data. In a similar way, it could be that mainly corals that manifested a high growth rate had the possibility of escaping the effect of competition or that corals that were actually not affected by direct competition - because they inhabited a cleaner spot - simply benefited from other characteristics, like possibly a higher nutrient availability, that the natural reef provided. In both these cases, the higher growth rates observed are manifestations of the deaths of slower growing or more susceptible corals on these reefs. Although the major noticeable difference between reef types was their difference in benthic community and with that in present competitors, no statement about the extent of this factor can be made. In any case, an interplay between reef characteristics has led to the observed instantaneous growth rates and positive properties of the natural reefs exceeded any negative effect of competition.

Next to a difference in competition, other factors that likely correlate with a reefs cleanliness are overall benthic community and presumed corallivory/disturbance by fish. These factors in effect have an outcome on growth via their respective contribution to the availability of nutrients. For example, fish-derived nutrient enrichment (Shantz & Burkepile, 2014) is thought to have positive effects on coral growth (Shantz & Burkepile, 2014; Meyer & Schultz, 1983; Meyer & Schultz, 1985). Due to this, nutrient availability is possibly higher on the natural patch reefs as opposed to the artificial reefs, which were originally devoid of life at the beginning of the experiment. However, issues of causality and correlation can only be hypothesized about, as these are confounding factors which make it harder to make any inferences about individual factors and because the relationship between growth and nutrient availability is complex and not well understood (Shantz & Burkepile, 2014). In the

current experiment, no data on the factors influencing nutrient availability was taken into consideration, but data on some of these factors has actually been gathered and a pilot investigation into the effect of benthic community and fish presence can be performed. Further research could thus look into the fish abundances and benthic community composition to narrow down the reef type effect more specifically.

Currently, the variable 'reef type' actually implies everything that is essentially different between the artificial reefs and natural reefs, and is not the effect of the other explored fixed effects like *reef depth* or random effects. The effect of reef type can thus further be explained through some additional factors. Such attributes driving differences in growth rate between the two reef types could be their difference in rugosity, overall shape or even the material used. Under the assumption that no prevailing difference in nutrients per water volume in the water column would occur within the study site, i.e., water quality is assumed similar, differences in growth rate induced by nutrient availability could partially originate from differences in water flow. While on the one hand the artificial reef used for the current experiment is intentionally designed with many crevices that could potentially stall water flow, its overall shape is domelike and smooth. Natural patch reefs on the other hand have a more rugose character, potentially stalling water flow more. At first thought, a higher flow rate potentially facilitates more growth via more nutrient influx, but low flow rates are often crucial because the increased flow rates can result in mechanical deformation of the exposed feeding surfaces resulting in suboptimal feeding (Wijgerde *et al.*, 2012). Additionally, *ex situ* experiments in aquaria have shown an important interplay between photosynthesis, light intensity and water flow rate. Generally, a higher flow rate would stimulate photosynthesis to the point of compensation, where there is no net change in O₂. However, from that point onward, a higher flow rate would only be beneficial under higher light intensities (Adams, 2006).

Light intensity is influenced by the depth of a reef, because of increased light attenuation with depth. While reef depth did seem to have a slight effect on growth; deeper reefs experience a lower growth rate, the depth range in the current experiment was not particularly wide and the effect was close to negligible. Therefore depth is assumed to not have induced much difference in light intensity between reefs. This perspective is furthermore underpinned by Ladd *et al.* (2021), who, contrary to the predictions of the light-attenuation paradigm, found no influence of water depth on coral growth for depths of 5, 10 and 17 meters. Therefore, under the assumption that not much difference in light intensity is present between the reefs because of the narrow depth range, the observed difference in growth between the reef types might be induced by higher flow rates around the overall lower situated artificial reefs. This could for instance have been because the higher flow rates were more detrimental than beneficial, as at these higher flow rates corals tissue could be more under stress of mechanical abrasion (Adams, 2006). The extend of the effect of flow rates would need further investigation, for example via the method suggested by Johansen, (2014).

Aside from reef type, place of attachment within a reef had a considerable effect on instantaneous growth rate. Corals attached to the bottom modules were situated about half a meter closer to the seabed than top ones. Furthermore, the bottom of the artificial reef was composed of three Moreef modules, possibly even having an increased sheltering effect. Both conditions have a decreasing effect on flow rate (Johansen, 2014). The finding that bottom corals had higher growth rates than top ones hint that the lower flow rates might have been beneficial for the corals. However, follow up studies need to take additional measurements to confirm the effect since no information on flow rates or light intensity has been gathered in the current experiment.

Continuing on the matter of the length of the corals at the start of the experiment, in the results section (Chapter 3) a strong case was made for not including 'Length at t0' as an explanatory variable

for growth rate. Indeed variation around the estimated slope in the linear model is similar for corals on both reef types (Figure 6), and corals on both reef types generally began branching after 25 mm of growth. While there may be an indication that the corals with smaller lengths at t0 needed slightly less length accumulation before branching, the main difference is that the 25 mm of length accumulation was already achieved for approximately 60 percent of corals on patch reefs in the second month. This in turn could then further drive differences of growth rates between reef types. The finding highlights that growth rates were already inducing these differences from an early stage and that they were not simply the result of differential branching moments for some arbitrary reason.

As a last note it is worthwhile to mention that the artificial reefs were only placed at the location of the experiment quite soon before the experiment started. The leaching of these concrete structures might have had a negative effect on growth rate. A study using cement as substrate to grow corals on in aquaria found a similar pattern. Cement plugs were not soaked for about six weeks before the experiment and thus were not chemically inert. Corals mounted on cement plugs initially grew slower than those mounted on ceramic, only overtaking ceramic plugs around 90 days into the experiment (Papke *et al.*, 2021).

In conclusion, the type of reef has a considerable effect on instantaneous growth rates of *Acropora cervicornis* between reefs and the location of attachment has a strong effect on instantaneous growth rate within reefs. Differential growth rates within reefs or between reef types are possibly induced by differences in overall nutrient availability (e.g. through differential water flow rates or fish presence induced nutrient enrichment). Depth was found to have an effect in this experiment as well, but only a minor and possibly negligible one over this range of 15 to 18.6 meters. The hypothesis that clean artificial reefs might facilitate higher coral growth rates cannot be supported by the data. Investigation into other components of coral growth is necessary to deduce which factor is most responsible for the lower growth rates on artificial reefs in the current study.

5. Recommendations

Any follow-up in-situ experiment should overcome the potential effect that leaching can have on coral growth by deploying the artificial reefs at least six weeks earlier than the start of the experiment. At time of translocation, corals need to be put in holes of similar depth to overcome any differences in competition that could arise through the method of attachment. Next to that, in the monitoring of health status of corals more variables can be taken into consideration in order to properly distinguish the type of deaths encountered. To figure the reason for types of death, the status of the attachment to the reef may be monitored visually or additional variables could be constructed to inform about the forces playing on a coral. A follow-up study would preferably also take variables as water quality parameters, wave exposure, light intensity and water flow rates into consideration. Not only would this make comparisons with literature more insightful, but the results from this study suggest that there are other factors than benthic community composition that play an important role in coral growth. While in ecological datasets a high degree of collinearity often complicates the estimation of individual parameters (Dormann *et al.*, 2013), hunting for additional predictors for growth in a follow-up experiment could result in more conclusive findings.

Regarding the research design, for a follow-up experiment it would be wise to use a more random design for the experiment than the current one. Here, all natural patch reefs were located at 15 meters under the surface, making it impossible to figure any differential effects of depth on reef

types. Also confounding factors such as fish presence and depth cannot be investigated with the current setup. Regarding the analysis, in the current experiment growth rates have been statistically deduced from a transformation of the total length of a coral accumulated over time. Another method could be to average the total accumulation per coral over the amount of branches for that coral or even follow the growth for each branch separately. This hopefully eliminates the effect of the amount of branches has on length accumulation. Additionally, this could give information on the growth per monitoring period.

From the current study it is clear that using uninhabited clean structures removes part of the simultaneous stress of outplanting and other stressors. However, initial cleanliness of the substrate does not seem to be the driving factor behind the survival of the corals in the long run in case no other measures to increase survival are taken. Therefore, more focus has to be put on overcoming hurdles for survival. In case measures effectively increase survival in the long run so that the eventual loss on the artificial reefs is less than the initial loss on the natural patch reefs induced by a combination of stressors, corals can benefit in the long term from relieving them from the cocktail of initial stressors. In that way artificial reefs can efficiently increase coral survival. In the same line of thinking, artificial reefs might be most efficient in locations that are less testing for corals were their long term survival is thus expected to be higher. Additionally, survival enhancing methods may even be easier to implement on artificial reefs than on natural patch reefs.

Even so, the benefit of increased initial survival with clean artificial substrates can possibly be mimicked by letting the corals acclimatize to the new environment on a nearby competitor-free structure first, before actually attaching them to any natural reefs. In this way, the need for artificial substrates can possibly be avoided. One would need to assess the similarity of the targeted environment to the nursery environment before translocation and conclude on the best method to minimize initial stress.

Literature and the finding that corals on the bottom of an artificial reef had higher growth rates than top ones suggests that that corals would benefit from specific flow rates. With modular artificial structures, the shape of the reef can easily be altered and specific flow rates can be facilitated. A follow-up study could investigate preferred shapes with these modular artificial reefs to maximize growth rates, in case that is a desirable trait. If slower growth rates would for instance correlate to denser and stronger corals, slower extensions rates could be beneficial for long term survival.

Finally, to limit death due to breakage, the corals could either be put more upright at time of outplanting or, in case of using modular artificial reefs, a shape should be chosen that shelters corals from disturbances.

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APPENDIX I

Acropora monitoring

Setup:

10 artificial reefs, each consisting of 4 Moreef modules
5 patch reefs are selected on the same location
12 coral fragments on every reef plot

Tie wrap color coding for outplants

1. Top unit corals receive one individual tie wrap, each one a different color. Tie wrap is placed just ahead of where epoxy is on the colony.
 1. White
 2. Green
 3. Blue
 4. Orange
 5. Pink
 6. Red
2. Corals on lower units receive the same 6 colors, however, each coral receives an additional black tie wrap.
 1. B-white
 2. B-green
 3. B-blue
 4. B-orange
 5. B-pink
 6. B-red

For patch reefs, there isn't a set method of placement as holes and crevices of opportunity were used. Therefore, the tie wraps and coloring will be randomly selected. However, the same pattern as mentioned above will be used, so make sure to follow the steps.

Coral measurement and health

Every month, each outplanted fragment will be monitored for growth and health.

1. Check every individual coral fragment for the health status. Remember every reef plot will be covered with 12 coral outplants.
2. Examine one coral fragment at a time and follow steps 3-8. Then proceed to the next coral fragment and repeat steps 3-8.
3. Firstly, observe the coral fragment and determine its overall health. Living coral fragments should be brownish to pale brownish in color and active polyps should be visible. You may sometimes encounter bleached fragments. Bleached corals are not dead, and they may recover. Therefore, make sure to look if coral tissue is still present. In all of these cases the coral is still alive.

As soon as **algae settles on the fragment**, the coral is not alive anymore and can be counted as dead. Look out for a fuzzy, tissue-like substance covering the coral fragment. When in doubt, note the color-code and note down the observation.

- After checking for health, proceed to check for breakage, bite marks and/or disease. Bite marks are not always visible because some fish predate the polyps. So, check if you see any spots that lack coral tissue and/or polyps. Disease can affect a small part of the fragment or affect the entire organism. Common diseases are white-band disease, white pox disease, black-band disease and Dark spots disease.
- Using calipers, measure the longest continuous branch starting at the front of the tie wrap to the tip. **Make sure to keep the caliper ends on the middle of the branch and measure to the nearest millimeter!**
- If the colony branches and forms a 'Y', and it is uncertain which branch is part of the longest; such as in figure 1, measure to the armpit and then measure the two branches separately.



Figure 1. fragment with uncertain branching measurement method

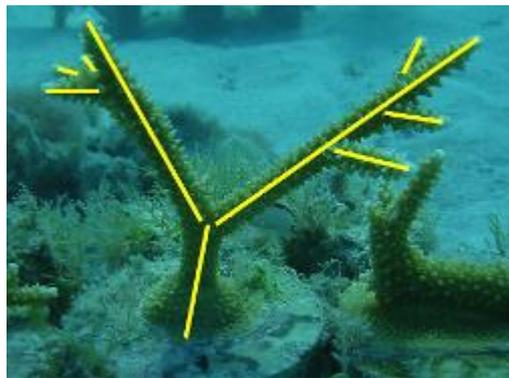


Figure 2. Measurement method of more complex individual.

- Measure each additional (side)branch as well, going from the tip to the middle of where the branch connects. If you draw a straight line from armpit to armpit for that branch, the middle of that line is the point to measure to. Every apical growth tip greater than 1mm is included. **Branches are considered as side branches when the expanding polyp has radial polyps.**



Figure 9. Side branch with axial polyp (growth tip) and radial polyps



Figure 4. Axial polyp which soon turns into a side branch due to the forming of radial polyps

- Make sure to write down reef number (number on side of top unit) and fill in the data for the correct color-code.

Materials needed:

- Slate with paper data sheet
- 2 pencils (one as backup)
- Caliper

Documentation

The following data sheet will be used underwater. Make sure to fill in who conducts the survey, on which date and which reef plot you are monitoring (number on side of top unit). **Note that the color codes are already filled in.** The order is not important, just make sure you fill in the data for the correct color code. The length section is divided into 4 slots. Slot 1 has to be filled in with the longest

Researchers:		Date:				Length in mm			
Reef ID	Colorcode:	Alive (1) Death (0)	Breakage?	Bite marks?	Disease?	1	2	3	4
	White								
	Green								
	Blue								
	Orange								
	Pink								
	Red								
	BWhite								
	BGreen								
	BBlue								
	BOrange								
	BPink								
	BRed								

continues branch measurement. The following numbers can be used for additional branches.

Pictures of coral

Every 3 months, an overview photo of each fragment is taken. The picture is taken with a size reference and in such a way that, if possible, all size branches are included. If that is not possible, make sure to make extra photos of the branches. It is important that all pictures include the tie wrap(s) to be able to identify them. Before fragment pictures are taken, take a photo of the reef number (number on top module), and repeat for each subsequent reef.

For the patch reefs, take a photo of the patch reef number on the rebar (1-5 tie wraps).

After returning from dive, check photos and confirm under water recorded coral health and mortality assessment. Check if all corals have been photographed by ensuring that all 15 reef plots have a matching number of 13 or above.

Materials needed:

- Camera

Data entry

1. Photograph survey sheet(s).
2. Rename the photos according to date.
3. Upload photos into Acropora survey folder on OneDrive.
4. Fill survey data into datasheet.
5. Store all data according to the data management plan
6. Clean survey sheet.

Please try to always put in the data on the same day of collection! Otherwise you risk not remembering certain aspects and this might mess with the data validity.

APPENDIX II

Report of the fixed and random effects estimates

1) Formulation of the linear model as used in R:

```
growthrate ~ reef.type + place_attach + Reef.depth + (1 | Reef.ID)
```

2) Results of the lmer() routine:

Random effects:

Groups	Name	Variance	Std.Dev.
Reef.ID	(Intercept)	2.782e-07	0.0005274
	Residual	1.147e-06	0.0010710

Number of obs: 102, groups: Reef.ID, 15

Fixed effects:

	Estimate	Std. Error	t value
(Intercept)	0.0108196	0.0030434	3.555
reef.typePatch reef	0.0011578	0.0005200	2.227
place_attachTop	-0.0012941	0.0002603	-4.972
Reef.depth	-0.0003799	0.0001795	-2.117

3) Results of the Aov() routine:

Error: Reef.ID

	Df	Sum Sq	Mean Sq	F value	Pr(>F)	
reef.type	1	1.210e-04	1.210e-04	43.353	3.95e-05	***
place_attach	1	2.670e-06	2.670e-06	0.956	0.3492	
Reef.depth	1	1.952e-05	1.952e-05	6.997	0.0228	*
Residuals	11	3.069e-05	2.790e-06			

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Error: within

	Df	Sum Sq	Mean Sq	F value	Pr(>F)	
place_attach	1	2.608e-05	2.608e-05	22.58	8e-06	***
Residuals	86	9.934e-05	1.155e-06			
