

# Culturing of coral spawn

Analysis of the implementation of a scientific project  
into the community of Koh Tao, Thailand



By

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(The picture on the front page shows an *acropora* coral growing over a glass bottle – Leuba, 2010)

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An advice, written within the frame of an orientation internship, over the implementation of the coral spawning project at the New Heaven Reef Conservation Project

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## Summary

In February 2010 the New Heaven Reef Conservation Program, part of the marine branch of Save Koh Tao in Thailand, started a new project, developed by Dr. James True, in order to increase the genetic diversity of corals on the reefs around Koh Tao by using sexual reproduction. This report is a restoration technique study over this “Coral Spawning Project”, which pays special attention to and gives advices over the implementation of this project. The aim here is to improve the efficiency of this scientific project for community-based organizations, like the NHRCP and others. In order to analyze this project it is combined with another project: the “Sediment Trap Project” is another project of the NHRCP and aims to measure the differences in sedimentation in space and time around Koh Tao. The cultivation process of this project is straightforward. In a certain night, when corals are spawning, divers collect the coral spawn, which is cultivated in buckets on land in order to protect it against several natural and anthropological threats. After the corals settled onto prepared settlement plates, they are moved out onto coral nurseries in the sea. The techniques therefore have been developed over many years, but the real challenge is to adapt them to community-based standards. During the implementation there were several problems faced and some are already solved. Water quality testing of the cultivation buckets showed that having good quality water for the coral spawn is difficult to accomplish, but not impossible. Therefore, water changes have to be done constant and more frequent. The water for the water changes has to be chosen carefully as well as the place for the coral nurseries due to fluctuation of water quality and sedimentation around the island. Unfortunately, it seems that until now no new corals are growing on these nurseries. A reason for this can be this year’s mass bleaching event due to increased ocean temperatures. Because of this, it is difficult to say anything about the approach and success of this project. However, there was already a lot of progress done and more improvements can be implemented easily. Because of this all, the Coral Spawning Project has to be carried out again under normal conditions.

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## Introduction

A majority of the historical coral reef ecosystems worldwide are now listed as depleted, rare, or extinct (Goreau 2005; Pandolfi et al. 2003; Wilkinson 1999). The biggest threats to reefs are natural ones, such as tsunamis and cyclical ocean warming. Though these events can almost completely decimate an entire reef, they only occur infrequent and localized, allowing the reef to regrow between events. In contrast, anthropogenic or man-made threats are less dramatic than natural ones, but are more frequent, widespread and sustained, leaving no time for reef regeneration. Impacts like introduction of organic and inorganic pollutants (i.e. waste water), increased sedimentation and exploitation of reef resources (i.e. fishing) slowly decrease coral health. Furthermore, structural damage caused by boats and anchors, fishing nets and divers destroy many corals. However, coral reefs play an important role in human economies. Reefs directly benefit humans by providing food (world average of about 220 U.S. dollars per hectare per year), raw materials (\$27 US/ha/yr), recreation (\$3008 US/ha/yr) and biological materials used in medicines and scientific research. It was estimated that in 1994 the earth coral reefs provided about \$10.6 trillion U.S. dollars to world economies (Costanza 1997).

Although knowing the importance of coral reefs, these treasures ecosystems are systematically destroyed. Many scientists have shown through modeling and trajectory patterns based off historical data that the world's reefs may be close to extinction within the next few decades; indicating that passive protective measures may not be enough, and restoration efforts must be attempted (Pandolfi et al. 2003; Goreau 2005; Wilkinson 1999). Restoration ecology is an active instrument, implying that at least some proportion of habitat goods and services are recoverable through anthropogenic manipulations (Rinkevich, 2008). Most coral reef restoration projects around the world concentrate on increasing the amount of corals in an area through propagation, or breaking apart a healthy coral colony to make many smaller colonies. However, the disadvantage of this technique is the loss of genetic diversity, because the new colonies are only clones of the mother colony. This will lead to higher susceptibility to diseases, changing water conditions, and other disturbances. One new method recently created to increase the abundance and genetic diversity of corals is the coral cultivation technique by using sexual reproduction (eggs and sperms are fertilized) and the transplantation of larval settlement into the degraded reefs.

## ***Koh Tao***

Koh Tao (literally translated “Turtle Island”) is a 21km<sup>2</sup> island located in the Gulf of Thailand (fig. 1) and is famous for being the cheapest place in the world to learn diving. In order to fulfill the needs of the divers, accommodation, restaurants, etc are built. Because of its flourishing diving industry it is apparent that Koh Tao depend largely on its diving grounds and thus coral reefs. But these reefs are threatened by many human activities; especially addressable are fishing and diving.



**Figure 1: Map of Thailand & Koh Tao (Google Maps, 2010; changes done by C. Hoppe)**

Therefore, the New Heaven Reef Conservation Program (NHRCP), part of the Marine Branch of the Save Koh Tao Community Group, has made it to their goal to monitor and evaluate the health and biodiversity of Koh Tao’s coral reefs in order to preserve and restore the coastal ecosystems. Furthermore, through the use of passive and active restoration techniques a reducing of human impacts is aimed. The most recent project of the NHRCP is the Coral Spawning Project and is one of the first of its kind in Thailand. This project is an attempt to increase the genetic diversity by using sexual reproduction and is being conducted along side with Dr. James True of the Prince of Songkla University in Hat Yai. The techniques have been developed over many years in Australia and are therefore “mature” in an academic sense (True, 2009). However, the community of Koh Tao has different interests as well as a different background and budget for this project. Because of this the challenge here is to adapt the techniques of scientist to the needs, knowledge and skill levels of community-based organization (True, 2009). Additionally, another recent project of the NHRCP is combined with the Coral Spawning Project in this report. The Sediment Trap Project aims to depict the differences and fluctuations in sedimentation in several bays around the island. Since corals are restricted to waters with low sedimentation, the determination of convenient “low-sediment” areas is necessary. The outcome of the Sediment Trap Project is combined with the results of Coral Spawning Project.

Thus, this report aims to improve the efficiency of the quite scientific Coral Spawning Project by giving advices over the implementation in order to meet the needs of a community, like Koh Tao.

Because of this it can be seen as a guidebook for proper implementation, but also as a restoration technique study for community-based organizations. Underlying this, the main research question arises:

What aspects of the scientific Coral Spawning Project need to be modified in order to implement it efficiently into a community like Koh Tao?

This main research question is answered with help of a few sub questions:

What is the Coral Spawning Project about? - (the biological and cultivation aspect)

How is it implemented?

What problems appear?

How can these problems be solved?

What is the outcome of the project until now?

Thus concerns this report the following: first, the biological background of corals and their sexual reproduction; second, a description of the whole cultivation process. After this, monitored data and problems (troubleshooting) are analyzed in order to give possible solutions to improve the efficiency of the process. In the end, the outcome of the project is shown, followed by a discussion over the course of the project and a conclusion and an advice.

## ***Methodology***

In the time from March to July the Coral Spawning Project was carried out twice by the NHRCP and several volunteers. During these implementations many (small) problems appeared throughout the whole process. Because of this the whole process was monitored and problems were analyzed in order to find solutions. Therefore, problems were discussed with the project coordinator Chad Scott and Devrim Zahir as well as with helpers of the coral spawning project who were interviewed afterwards and had the chance to give their comments over the process. Their opinions and ideas were used to find solutions. Some ideas go beyond the scope of this research and were only added to the discussion section. Furthermore, the problems were also approached through literature research. Some problems could be solved easily and the solutions were already implemented when the Coral Spawning Project was carried out the second time. However, they are analyzed in this report as well. When the project was implemented the second time, the water quality (in the cultivation buckets) was tested by using a multimeter and a colorimeter in order to measure temperature, ph and nitrate and phosphate. The results were taken into account as well as the results of the Sediment Trap Project. This project took place in the same time as the Coral Spawning Project and provided much information over proper areas for coral to grow. At the moment there are three different sediment traps installed near coral nurseries in the bays of Chalok Baan Kao, Ao Leuk and near the Hin Fai-Rock. Each trap has three tubes which catch sediment. On land the sediment is filtered out of the water in the tubes and is, if dry, weighted with a scale. The weight of the sediment in all three tubes is added and then divided by the number of days since the last time the sediment trap was emptied. The outcome gives an idea of the sedimentation in the different bays per day

The actual cultivation process of coral spawn is described in the respective section and was developed by Dr. James True. However, several modifications were made. These are described in "cultivation process" section as well.

## Biological process

Coral reefs are certainly one of the most important natural attractions of our planet. High biodiversity and great ecological complexity are significant for them. They provide habitat and act as nurseries for fish and aquatic invertebrates, provide barriers from storms and waves to protect sea coasts, and help to regulate atmospheric gases. Though they only cover 0.1% of the ocean floor, they support over 25% of all marine life (Scott, 2010). Additionally, they are one of the earth's oldest ecosystems, made out of vast amounts of calcium carbonate (CaCO<sub>3</sub>), limestone, which are the remains of skeletons of several reef inhabitants, like mollusks, crustaceans and corals. The primary architects of coral reefs are the last one.

However, "corals" is a general term for several reef building organisms of the phylum cnidarians. The actual organism which is responsible for the growth of coral reefs are sessile animals called coral polyps. These polyps live in symbiosis with an alga called zooxanthellae, which lives inside the tissue of the polyp. Zooxanthellae perform photosynthesis and provide polyps with nutrients which they need for metabolic activities like producing their calcium-carbonate skeleton or in order to reproduce. The most important factors for corals to grow are temperature and light availability. Coral reefs are very sensitive to temperature and are healthiest in a temperature range of 22 to 28 degrees Celsius, and usually do not survive in waters that are not within an 18° to 36°C range (Hoegh-Guldberg 1999; Wilkinson 1999). In order to perform photosynthesis corals require light. Therefore, most hard corals only grow to a maximum depth of about 60 to 100 meters in tropical areas; increased latitude, sedimentation rates, or plankton growth will decrease this depth dramatically (Hoegh-Guldberg 1999; Wilkinson 1999). Another important factor in coral health is the water quality. Generally, corals are not able to survive in water with high sediment and pollution concentrations, which generates stress. Corals stand in direct competition with micro- and macroscopic algae, which favor nutrient enriched water and will overgrow corals under these conditions. Furthermore, pollution can increase the amount of marine diseases in many of the same ways it affects terrestrial life; currently there are over 12 previously unknown diseases attacking corals worldwide (Castro 2007).

Coral reefs form a very complex ecosystem based on a central theme of mutualistic, commensal, or parasitic symbioses. Many reef organisms are reliant upon another specific species of organism to survive. Corals not only rely on zooxanthellae to gain necessary nutrients, but also on fish and invertebrate grazer species which consume competitive algae. The elimination of any species in a reef has far reaching effects throughout the coastal ecosystem.

Corals reproduce both, asexually and sexually. The asexual process can be described as multiplication of existing polyps. The parent colony is divided to form new coral colonies which are then genetic clones of the old one. This results in relatively rapid growth but without contribution to the genetic pool. In contrast, sexual reproduction increases biodiversity. During the sexual reproduction corals can be either hermaphroditic (both sexes present) or gonochoric (produce either eggs or sperm). Furthermore, corals have two different strategies to reproduce, the brooders and the broadcasters. Brooders are mostly hermaphrodites and self-fertilize within their gastrovascular cavity, releasing multiple times per year planula larvae into the water when they are ready (Borneman, 2001).

Broadcasters, in difference, do not self-fertilize, but release billions of little bundles, which may contain both eggs and/ or sperm, into the sea. The process is similar and analogous to the wind-pollination of terrestrial plants (Borneman, 2001). This project focuses only on the sexually reproduction of broadcasters.

Spawning only occurs once or a few times per year in the night and is generally done *en masse*. In a certain night corals from all over a reef release billions of their gametes at the same time into the water. Therefore, the corals must be in good conditions and have excess energy in order to produce and release gametes in form of bundles of eggs and sperm. Furthermore, other factors, such as water temperature and the lunar phase are very important for the corals to spawn. While a critical temperature prepares corals for an upcoming spawning event, the moonlight level seems to be the actual timing trigger for the release of the gametes. When the corals are about to spawn, the



gametes are transported from the gastrovascular cavity, where they are produced, to the oral disc, where they are released. The oral disc surrounds the mouth and gametes are set there for up to an hour before actual spawning. It is possible to see them there with the naked eye (see figure 2). Once released, the gametes start rising to the water surface where they break up, allowing sperm and eggs to mix. They form slicks of fertilized eggs and drift with currents for the next 4 to 5 days. Already after 1 to 2 hours after spawning, cell division is initiated and planulae larvae hatch during the next day (Borneman, 2001).

**Figure 2: Coral Spawning (True, 2009)**

The coral spawning is a food feast for many other reef inhabitants and attracts different species of fish and invertebrates. Even other adult coral polyps feed on coral spawn. By spawning in mass, the coral gametes have a higher chance to survive and settle down successfully than if they were released at random times. However, the gametes are not only threatened by predators during their floating and drifting stage, but also by many other factors. Actually, the mortality rate for the corals prior to settlement has been estimated in excess of 90% (Borneman, 2001). Coral spawn is washed up on the beach or is lost at sea, but is also killed by slightly disturbances in the quality of the surface water. Oil leakages cause a film on the surface and heavy rainfalls reduce the salinity of the surface water, which both easily can kill the floating larvae. The planulae larvae lead a planktonic life on the surface until they settle down onto any useful substrate on the reef, with prefer of solid and stable substrate, like rocks or dead corals. A number of chemical cues present in the water induce the settlement of planulae, as well as biochemical and metabolic factors variably present in the larvae (Wilson & Harrison, 1998).

When the larvae are about to settle down, they start to actively swimming down in search for a proper substrate. The texture of the substrate is at this point very important. Coral larvae prefer a complex topography as substrate. Furthermore, chemical signals emitted by the biofilm, a coating of bacteria and microbes that form a film on substrates, induce settlement as well. Especially coralline algae in the biofilm on the substrate seem to be the most important factor for coral settlement. Of course factors which are important to adult corals, like light, water depth, temperature, sedimentation, and symbiosis, are also important for coral larvae.

If the larvae were able to settle down and attach to a proper substrate, polyps begin to develop and lay down a skeleton. However, the mortality rate of juvenile corals in this stage is still 60 to 90% due to heavy predation and competition with other species (especially algae which are directly competing with corals). Another 30 to 40% will fail to reach maturity (Borneman, 2001). But if successful, a colony begins to grow through coral budding (a form of asexual reproduction). The colony may take 4 to 5 years to sexually mature and in order to reproduce sexually again.

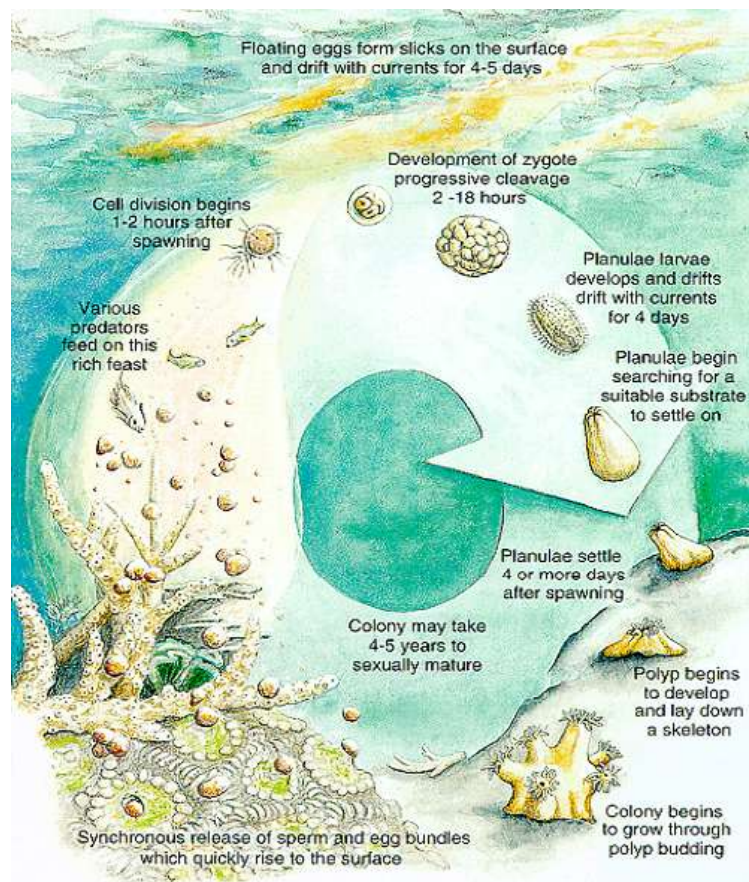
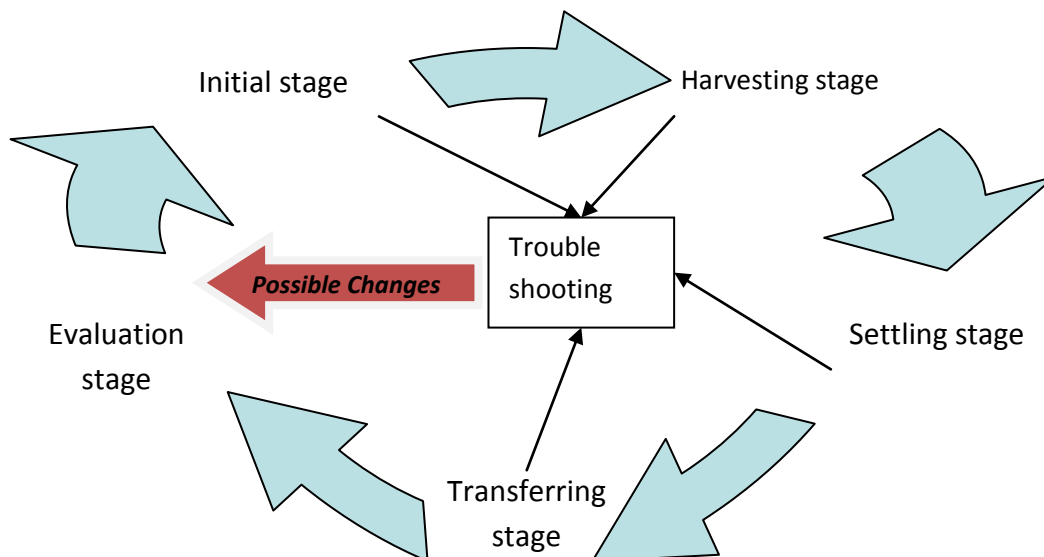


Figure 3: Spawning cycle of corals (True, 2009)

## Cultivation process

As already stated, the Coral Spawning Project is a coral cultivation technique by using sexual reproduction (eggs and sperms are fertilized) and the transplantation of larval settlement into the reefs. In this way, it is aimed to a) increase fertilization success of widely dispersed colonies; b) preserve and increase genetic diversity and c) provide a sustainable source of young corals for transplantation onto natural and artificial reef structures.

The general process is quite simple and straight forward, but requires lot of care. First, the date of coral spawning (release of eggs and sperm into the water) is determined. Furthermore, buckets, settlement plates, etc. must be prepared beforehand (before the coral spawning). After collecting the sex cells in a certain night, they are moved into buckets of sea water on land to be in a safer environment. After a few weeks in the buckets, the hatched coral larvae are allowed to settle down on concrete plates shaped like mushrooms, and put out onto specially designed coral nurseries in the sea.



**Figure 4: Stages of the Coral Spawning Project (Hoppe, 2010)**

In order to simplify the analysis and evaluation of the process afterwards, it is (in this case) divided into 5 stages of involvement, according to the different activities which were taken (see figure 4). Together they form a “cycle”. The cycle starts with the initial stage and ends with an evaluation which is then again used for the next cycle. The following description of the process is exactly as it was carried out by the NHRCP for the first time and as it was recommended by Dr. True. Until now there were two cycles of coral spawning.

The **initial stage** consists of monitoring coral colonies to determine the date of spawning. Therefore, Dr. James True visited the island in February 2010 to break off small parts of corals in order to look inside the mesenteries (place where polyp is attached to skeleton) weeks before. If the coral is going to reproduce eggs and sperm, they can be seen in the mesenteries and the date of spawning can be predicted. Corals develop the sex cells in the course of 9 months. During the first 6 months the sex cells cannot be seen, while they turn macroscopic in the next months. When they change from white to red, it is apparent that they are going to reproduce after the next full moon. The number of days between the full moon and the coral spawning is always the same. That means that if the exact date of spawning is known in an area, it will be the same in the next year. The corals which seemed to reproduce were marked. In addition, the initial stage comprises preparations for the proceeding stages; nets for the second, buckets and settlement plates for the third and coral nurseries for the fourth stage must be prepared.

The nets for the second stage are actually modified butterfly nets with the top of a plastic bottle glued to the cut off top of the butterfly net. Additionally, a jar or similar must be provided to collect the coral spawn.

The preparation of the settlement plates (figure 5) requires a little bit of time. An easy settlement substrate can be manufactured out of concrete. It is important that the artificial substrate has an attachment point in order to bring it onto a coral nursery, as well as a complex surface topography where coral larvae can easily attach to. In the end they look like concrete mushrooms. The base for it is prepared with a piece of wire looped around a piece of drinking straw to provide a convenient reinforced attachment point, which is put into a small round mould. This mould is filled with concrete, mixed out of sand and cement (1:2 ratios). The same concrete mix is poured into an egg carton mould, which has a complex surface topography. The dry bases are pushed into the wet cement of the top piece in such a way that it is ensured that the base is horizontally and the metal reinforcing is covered (see figure 5).



**Figure 5: Preparation of settlement "mushrooms" (True, 2009); one mushroom is ca. 6cm high and has a diameter of 2.5 cm to 4 cm**

When the mushrooms were dry, they were placed into the seawater (near the nurseries) for 1 week during the first cycle and for 2 weeks during the second cycle, to form a biofilm with coralline algae on it.

The buckets, which are used as cultivation units, are simple food-grade plastic buckets. The water in the tanks will be gently aerated with aquarium aerator to enhance circulation.

The coral nurseries are built out of PVC-pipes, which are glued together to form a frame. The inside forms a mesh out of strings, knotted to the frame. These nurseries are moved out to the sea and are attached underwater. All preparations were finished before the second stage.

During the **harvesting stage**, when the corals are spawning, divers collect the released sex cells in the nets and jars and move them into the buckets of sea water on land. The top of the nets (plastic bottle) is kept positive buoyant. Coral that are ready to spawn will “set” the egg-sperm bundles at the oral disk for up to an hour before actual spawning. This is the sign for the divers to get ready. Because of the fact that the bundles ascend to the surface, they can be caught easily by divers hovering with the net over the coral (see figure 6).

When the net becomes too full they are emptied into the jars, carried by another diver. The first time of coral spawning and collecting took place on March 4<sup>th</sup> 2010. At this time branching and table corals of *Acropora* Corals were collected. The second time was on April 4<sup>th</sup> 2010 and corals of the species *infauida* (massive and brain corals) were spawning. After the collection, the coral spawn was put into a fertilization tank, a bucket of seawater, where they stayed for few hours, before they were moved into the prepared cultivation buckets. Two blue and two clear buckets were used in order to assess at the effect of differences in temperature and light inside the buckets.



**Figure 6: Collecting coral spawn (Leuba, 2010)**

For the next days the corals must be provided fresh seawater until they settle down on the prepared substrate. This is the **settling stage**. In the first few days it is necessary to change the water 2 times per day, after this 1 time per day is sufficient (True, 2010), because especially in the first days the breakdown product of unfertilized eggs or just dying organic matter will make the water quality go down. The fresh seawater was taken out of the bay in Chalok Baan Kao and was carefully poured into the buckets, while the “old” water was siphoned out with a hose. Aeration and shade was supplied. No additional material was used to configure temperature, salinity or any other abiotic factor.

After the coral larvae settled down they were put out by divers onto the designed coral nurseries in the sea (**transferring stage**). The nurseries can be found in Ao Leuk and in Chalok Baan Kao (see figure 1). There they are monitored for the next year until the corals are big enough to move out onto the natural reef or onto an artificial reef.

After this all the whole process of the first cycle was evaluated in order to improve it for the next time (**evaluation stage**). This part is discussed in the following section.

## **Analysis and evaluation of the process**

During the first completion of the coral spawning project many problems appeared. These problems are divided now according to the respective stage. Some problems were already solved and the solutions implemented in the second cycle. These improvements are analyzed and evaluated in this section as well as possible solutions for other problems, which are not implemented yet.

Furthermore, helpers of the coral spawning project were interviewed afterwards and had the chance to give their comments over the process. These comments are as included as the monitoring data of the coral nurseries (with settlement plates) and testing of the water quality during and after the second cycle.

### ***Initial stage:***

After the first coral spawning, it became apparent that a few problems arise already out of the initial stage. The marking of certain corals is unnecessary, because divers only gather around the marked corals, bumping into each other and disturbing each other. However, all corals of a certain species in a certain area release their sex cells all together in one and the same night. Instead of marking certain corals, it is rather recommendable to mark a whole area. This will spread divers over a wider area and additionally, spawn of many more different colonies is caught which raise the biodiversity. The method of preparation of the settlement plates seems to be satisfactory, because of the complex structure coral larvae can settle onto them. However, the coral larvae needed more time than expected to settle down (especially in the blue bucket). Especially coralline algae are an important factor for coral settlement. During the monitoring of the coral nurseries it became apparent that more coralline algae are on the settlement plates after more time has passed (4 weeks). Therefore, it is recommended to place the settlement plates into the water much longer before the coral spawning in order to have more coralline algae to induce the settlement of the coral larvae.

### ***Harvesting stage***

The most important question during the harvesting stage is: "Is enough coral sperm caught?" There are many possibilities to increase the catching quota. The use of stationary nets over certain coral colonies is recommended by Dr. James True. These nets are pulled over the coral and are kept there with the help of threaded cords. However, the NHRCP decided against this method, because of the damaging of corals and the fact that 100% of the spawn is caught, "there is no chance for corals to reproduce naturally" (Scott, 2010). Nevertheless, an increased amount of coral spawn is recommended, because of the high mortality rate of coral spawn and larvae (>90%). This can be reached not only with the help of stationary nets but also by increasing the number of divers and/or by improving the communication between the divers. The actual spawning (the release of the sex cells) only takes about a few minutes. Some divers were still on the boat while some corals have already finished spawning. Therefore, in order to coordinate the collecting better, every diver on the boat has to be ready all the time, from sunset on until the spawning is over. Additionally, one buddy-team (two divers) has to be under water all the time in order to dive in the marked area and check on several corals if they are already setting their egg-sperm bundles at the oral disc (they can be then seen as red dots). If not, the team goes back to the boat after half an hour and the next buddy-team goes into the water. But if the corals are about to spawn, one diver ascends to the surface and

informs the other divers who have to hurry to get into the water. Here it is also useful to establish an order in which the divers are going into the water (in order to prevent panic and confusion). Once in the water they directly start diving (always in teams of two!) and harvesting the coral spawn with help of the modified butterfly-net.

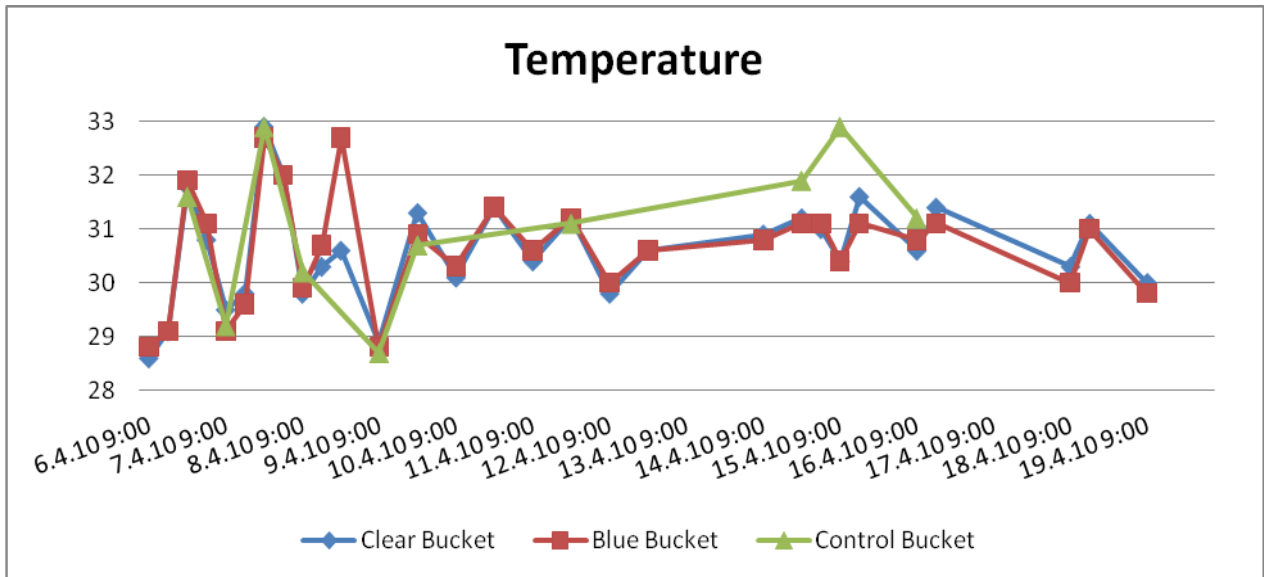
Another problem during the harvesting stage concern the fertilization buckets. The water was taken directly out of the sea without filtering. Because of that, small invertebrates which feed on coral spawn came into the bucket as well. Therefore, the water needs to be filtered with plankton mashing beforehand.

Furthermore, the nets only need to be emptied into the jars when the divers are about to ascend and go back to the boat. It happened this time that the nets were emptied many times, while the air out of the nets filled up the jar every time until the jar was only full of air.

### ***Providing stage***

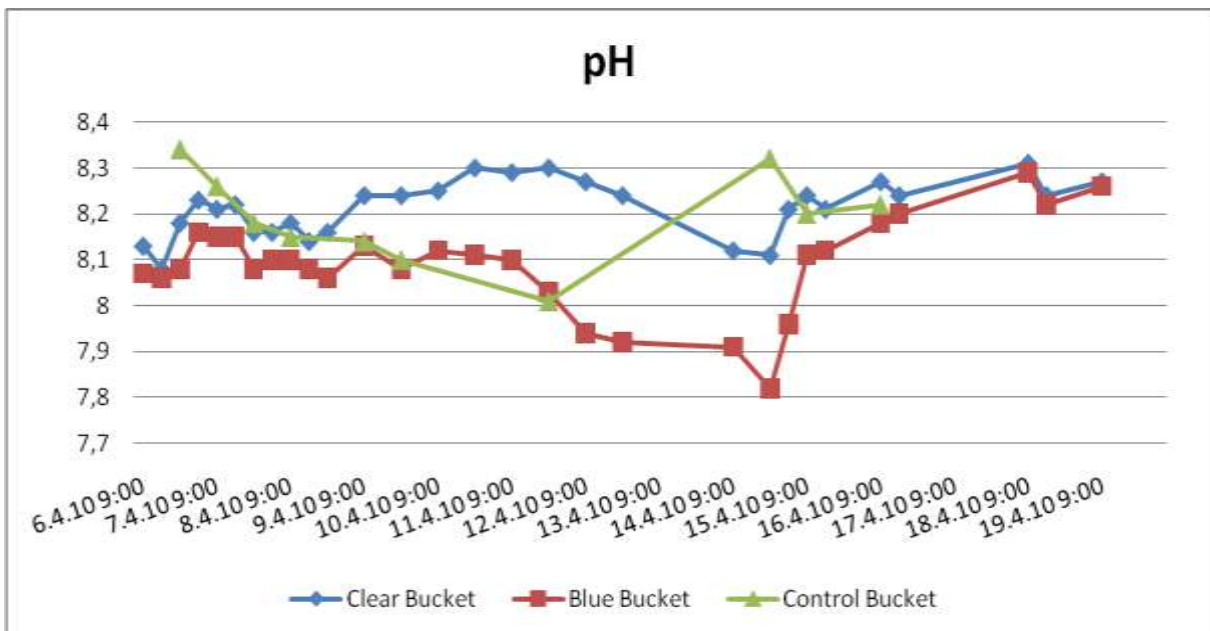
The providing stage is the most important and most difficult stage and brought up many problems, but has been improved a lot since the first coral spawning cycle. One of the most important improvements of the second cycle is the implementation of water quality testing before and after every water change. Temperature, pH were measured with a multimeter, while the turbidity, the concentration of phosphate ( $\text{PO}_4^{3-}$ ) and nitrate ( $\text{NO}_3^-$ ) were tested with a colorimeter. During the first three days there was a water change every morning and evening (6.-8. April 2010). According to Dr. James a water change was not necessary for the next couple of days. Because of this and the celebration of Songkran, the Thai-New Year, the water changing stopped (9.-13. April 2010). After this there was a water change once per day (14.-18. April 2010). The water of the clear and the blue bucket were tested as well as the fresh seawater, which was used for water changes (control bucket). This water was taken from the sea surface mostly right before the water change, but sometimes the water already standing in the bucket was used. After analyzing the water quality data of the second coral spawning cycle with Office Excel 2007, it becomes apparent that the water quality between the clear and the blue bucket does not differ very much. The results of the water testing can also be found in appendix I.

The temperature in both buckets is nearly equal throughout the whole providing stage (see figure 7). The average temperature in the blue bucket is with  $30.64^\circ\text{C}$  slightly higher than the average in the clear bucket ( $30.6^\circ\text{C}$ ). In contrast, the fresh seawater and especially the water already standing in the control bucket was often hotter. On the one hand that means that the shading for the other buckets is sufficient and the corals do not get too much sunlight, but on the other hands it shows the importance of getting water for the water change fresh and from a higher depth. The water on the surface is due to the sunlight just too hot. Because the temperature in the buckets changed throughout the whole day it would be helpful to measure the temperature constantly in order to figure out when it is the best time to change the water.



**Figure 7: Water quality testing of the cultivation buckets – Temperature**

The pH in the clear bucket was throughout the whole time higher than in the blue bucket (see figure 8). While the pH average in the clear bucket is 8.21, the average in the blue bucket is 8.09. Normally, the pH of seawater is between 7.9 and 8.3. If the water is getting hotter the pH goes down. However, for the project a higher pH (more basic water) is preferable for the corals to form their calcium skeleton more easily. If the water is too acidic, carbonate salts are dissolved and so do the skeletons of corals. Instead carbon-dioxide is released.



**Figure 8: Water quality testing of the cultivation buckets – pH**

The nitrate ( $\text{NO}_3^-$ ) concentration in both buckets is throughout the whole time very similar (see figure 9). The  $\text{NO}_3^-$  average in the clear bucket (0.92 mg/l) is slightly below the average in the blue bucket (0.96 mg/l).

The same can be said over the average of phosphate ( $\text{PO}_4^{3-}$ ) concentration in the buckets (clear bucket= 0.15 mg/l to blue bucket= 0.16 mg/l; compare with figure 10). But actually was the water quality in the blue bucket quite good until the water changing stopped for 5 days. It can be said that the water quality (pH,  $\text{NO}_3^-$  &  $\text{PO}_4^{3-}$ ) in the blue bucket goes down faster, if there is no water change over a longer time. Furthermore, the data of water quality depicts that the water change can be stopped up to a maximum of 3 days. Until then the water quality is quite stable, but goes down very fast if this limit is exceeded.

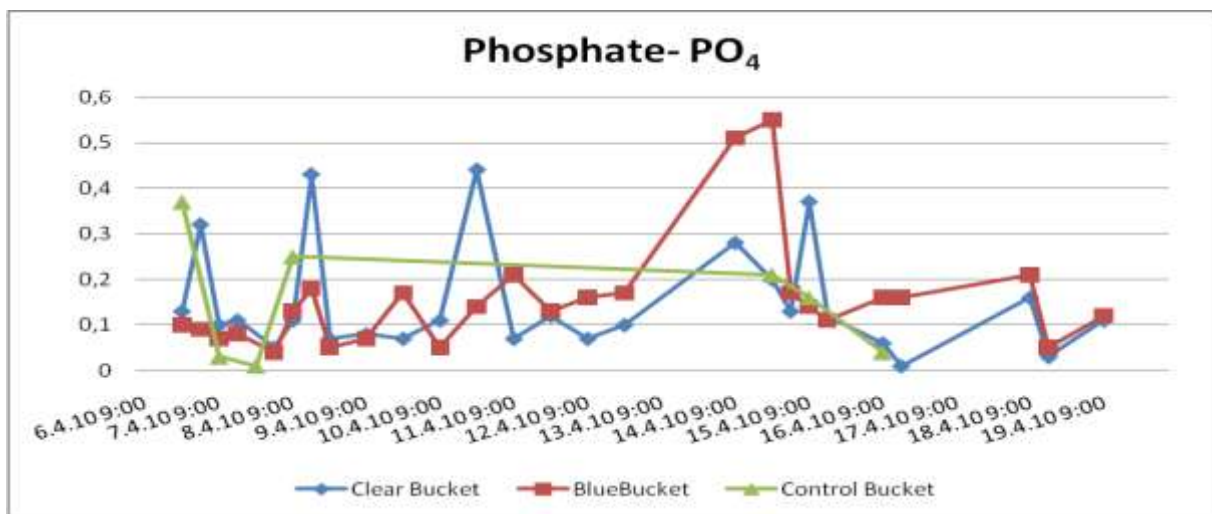


Figure 9: Water quality testing of the cultivation buckets – Phosphate

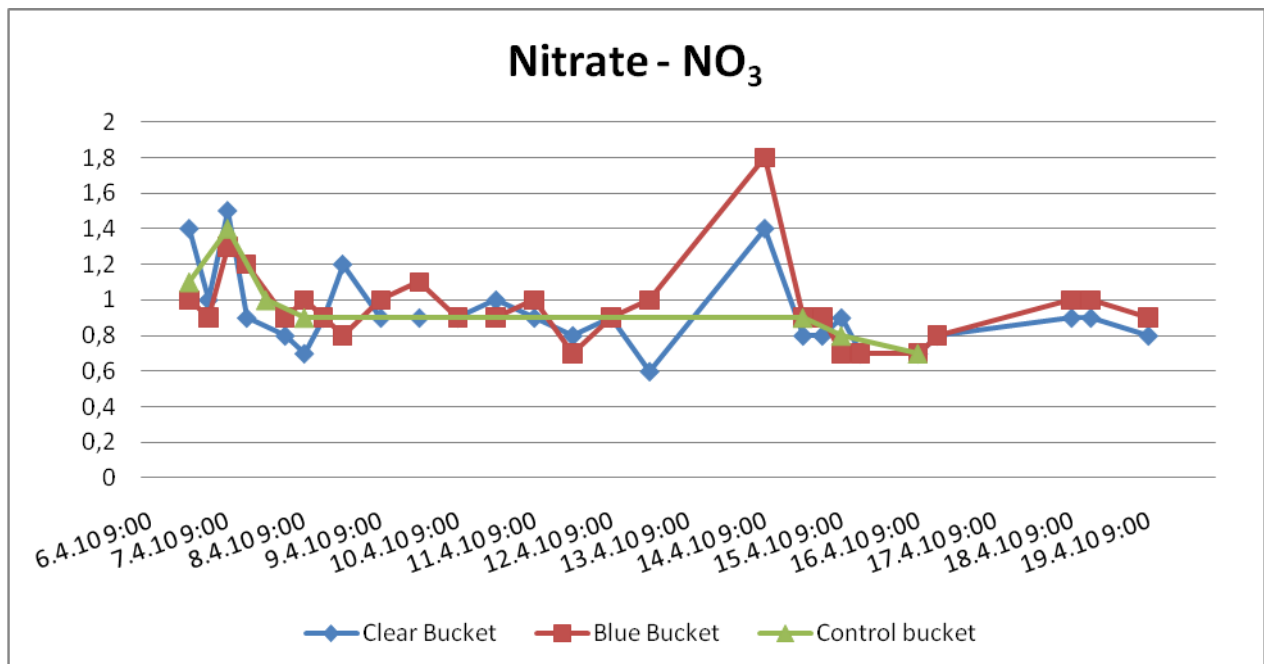


Figure 10: Water quality testing of the cultivation buckets – Nitrate

Because of this a better planning of water changes is highly recommended. Especially in the first days the water should be changed more often, because breakdown products of corals which do not survive are making the water quality go down and their proteins and lipids are floating on the surface in form of scum. Furthermore, even if the water quality stays stable for 3 days without water change, such long breaks are not recommended, because high water quality is very important for corals to grow.

However, the water quality of the fresh seawater in the control bucket changed very much. In the beginning the water was taken near the shore in Chalok Baan Kao where the quality fluctuates very much due to human activities in the bay and it happened that the water quality in the buckets was even worse after the water change. In the end the water was taken from further out and had less nutrients and a better quality. But testing the water near the coral nurseries showed that water quality here is much better and also cooler. Here the average concentration of phosphate is 0.04 mg/l and the nitrate concentration 0.7 mg/l, while the water quality in the control bucket has an average phosphate concentration of 0.15 mg/l and an average nitrate concentration of 0.97 mg/l.

In conclusion, the water quality between the buckets does not differ very much, but is slightly better in the clear bucket. Furthermore, while the larvae in the clear bucket settled down nearly as predicted (one week after spawning), the larvae in the blue bucket settled down a 4 to 5 days later. Reduced sunlight in the bucket can be the reason therefore. The corals might think that they are too deep and are waiting until the water is getting shallower. Because of all this, the clear bucket seems to be more useful as a cultivation tank than the blue bucket. Additionally, it is very important to plan the water changes very well and to get the fresh seawater more often and from further out and deeper in order to have a good quality. However, this would be very time and work intensive, but it is possible. Therefore, volunteers (and divers) have to be divided into groups. One group is in charge for the water change, while the other group takes care to provide fresh seawater. For each duty only two volunteers are needed. A schedule has to be made that all volunteers take turns.

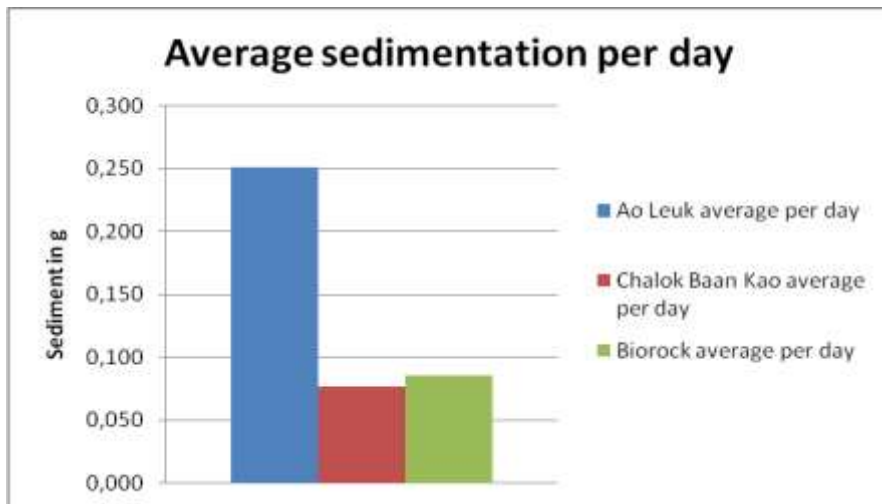
The water change itself made many problems as well. After each water change we lost many coral eggs which were still in the exchanged water. Because of this, we used a PVC-pipe with plankton mashing during the second cycle to filter the eggs out of the "old" water and put them back in the bucket with the fresh seawater. Furthermore, during the first coral spawning cycle the water change was done by siphoning the water out of the buckets with a small hose. This method is very time-consuming. Because of this, for the second cycle there were valves added to the buckets in order to simplify and speed up the water change.

## ***Transferring stage***

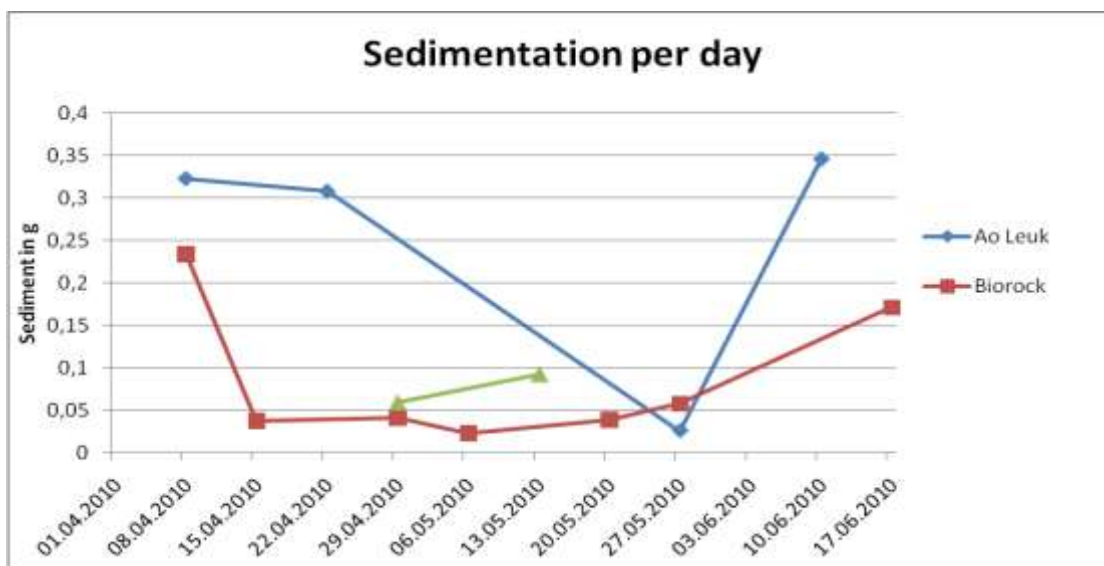
The transferring of the settled coral larvae onto the coral nurseries in the sea can be easily improved. During the first time the divers were not really gentle with carrying the buckets onto the boat and attaching the settlement plates onto the nursery. However, even if the coral larvae are attached to the settlement plates now, they have not built up a skeleton yet. Because of this they are not connected very strong and are easily disturbed. While putting them onto the coral nursery, divers are not allowed to touch the upper part of the settlement plates where coral larvae could have settled. Furthermore, the buckets and the settlement plates have to be transported very gently.

Divers have to be very gentle anyway, not only during this stage but during the whole process (f.e. during the water change). Therefore, it is necessary to give the divers and other helpers a proper briefing beforehand about the importance of the project, the tasks and how to carry them out. However, most of the helpers were only sometimes involved into the process and barely knew anything about the project. Because of this, it is necessary to give a briefing before every stage to bring everybody on the same level of knowledge. Additionally, there were many helpers for the dives to collect the coral sperm, but only a few people to help with the water change. It is important to encourage the divers to help with the water change as well. During the first coral spawning, many divers agreed to help with water change but never showed up. Because of this a written schedule has to be made in the same night as the coral spawning in order to know who is when in charge of water testing.

After the coral larvae were attached to the coral nurseries appeared another problem. Corals are reliant upon another specific species of organism to survive. They not only stand in symbiosis with zooxanthellae to gain necessary nutrients, but also with fish and invertebrate grazer species which consume competitive algae. However, while monitoring the nurseries, the fast growth of algae on them showed, there are neither fish nor invertebrates which could graze on these algae. The NHRCP has experienced that fish will definitely come to artificial structures after a longer time. But for the coral larvae they are already really important in order to compete with the algae. Thus, it might be useful to move the coral nurseries into waters with high fish abundance for the first weeks. Additionally, it is important to choose a good position for these nurseries since corals cannot grow very well in water with high sedimentation and/or high nutrient concentration. As the recent Sediment Trap Project of the NHRCP shows is the quantity of sediment in every bay different. At the moment there are three different sediment traps installed near coral nurseries in the bays of Chalok Baan Kao, Ao Leuk and near the Hin Fai-Rock. Each trap has three tubes which catch sediment. On land the sediment is filtered out of the water in the tubes and is, if dry, weighted with a scale. The weight of the sediment in all three tubes is added and then divided by the number of days since the last time the sediment trap was emptied. Even if the data is not reliable yet, because of a small database, it should not be ignored. Until now the data depicts that the average sedimentation per day in Ao Leuk is with ca. 0.25g sediment nearly three times higher compared to Chalok Baan Kao (0.076g) and the Hin Fai-Rock (0.086g) (see figure 11). Figure 12 shows the sedimentation per day in the course of time. A point on the line stands for one measurement. The results of the sediment traps can be seen in a table in appendix II.



**Figure 11: Result of sediment traps - average sedimentation**



**Figure 12: Result of sediment traps - sedimentation per day in the course of time**

However, it is important to take into count the surrounding of the nurseries here. While the nurseries in Chalok Baan Kao are surrounded by big brain and similar corals which inhabit several fish species, the ones in Ao Leuk are unprotected in the middle of a sandbar. Simply because of the fact that there are corals means that there is less sand and thus less sediment. Furthermore, a few corals near the nurseries in Chalok Baan Kao can catch the sand and prevent it from suspending again. This can be the reason for less sedimentation. However, if you swim closer to the coral reef in the bay of Ao Leuk, away from the nurseries, the visibility increases while the sedimentation decreases. Of course, coral nurseries are used to get to corals off the bottom into mid-water with less sedimentation, but in times like these they had to be lowered down because of too much sun and too warm water in mid-water. This all shows the importance of a good position of the coral nurseries, preferable near coral reefs, where sedimentation & pollution is low and where corals can stand in symbiosis with other species.

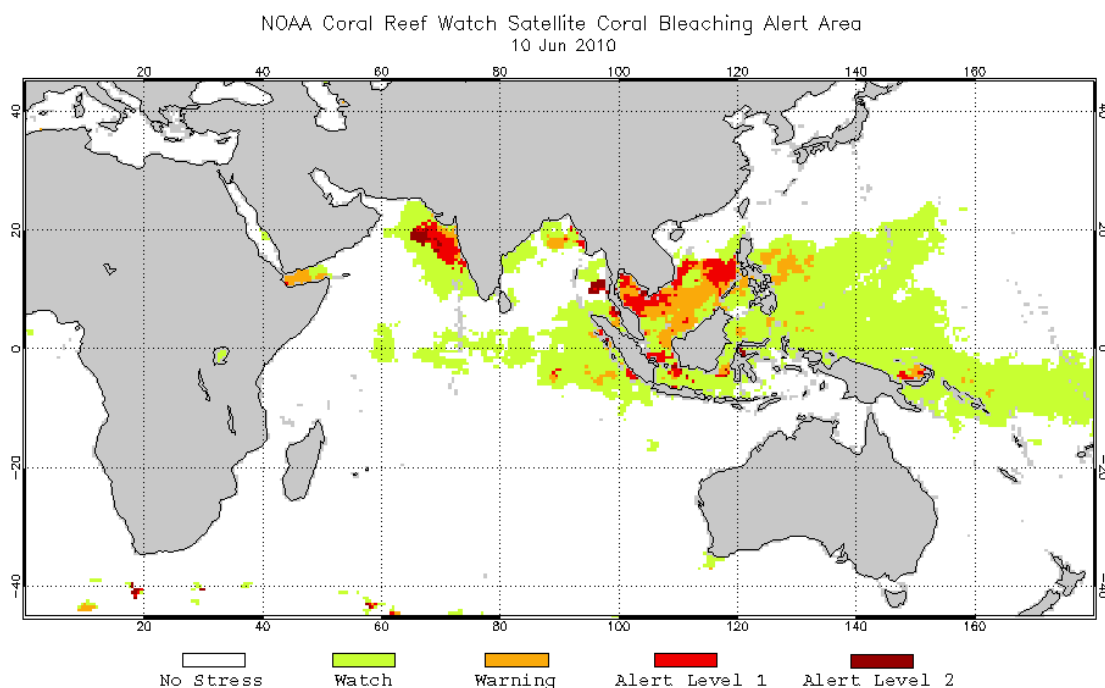
## Progress

The first coral spawning cycle took place on March 4<sup>th</sup>, the second time on April 4<sup>th</sup>. After 2 weeks of providing, the now settled coral larvae were moved out to the coral nurseries in the sea. There they were monitored and are still monitored. That means that divers were looking for corals which already built up a skeleton, which can be seen with the blank eye. It is possible to see the first skeletons with the eyes after two month (True, 2010).

Two weeks after transferring to the sea, first sponges start to grow on the coral nurseries and the sediment plates. This is not a bad sign, because sponges filter the nutrients out of the water. However, already now the nurseries were covered in algae, because there are no fishes in the area which feed on algae. Now, after 3 months for the first coral spawning cycle and 2 months for the second, the situation has barely changed. More sponges are growing; the settlement plates have more coralline algae on them and there are still many algae on the nurseries and settlement plates. Unfortunately, it seems that until now no new corals are growing there. There are many reasons why this project was not successful

## Discussion

One main reason, why this project was not successful is the fact that this year the Gulf of Thailand heated that much that a mass bleaching event took place (see figure 13). Coral bleaching is a term used to describe the phenomena when the coral loses all or some of their symbiotic algae (zooxanthellae), which lives inside the tissue of the coral and provides it with nutrients. However, under stressful conditions (e.g., low salinity, pollution, unusually high or low water temperatures) the zooxanthellae starts to produce reactive oxygen which damages the cellular structures. Therefore, the coral starts to expel the zooxanthellae from their tissue – with the result that the white calcium carbonate skeleton becomes visible through the now transparent tissue layer. The coral can recover from this if the environmental conditions are changing again after a short period. If the conditions persist, the coral may die. Furthermore, because of the loss of zooxanthellae, the coral also loses 90% of its nutrients and energy budget, supplied by these algae.



**Figure 13: Progress of coral bleaching (NOAA Coral Reef Watch, 10.June 2010)**

Mass coral bleaching is primarily caused by unusually high sea temperatures. This year's bleaching event is caused by El Niño. El Niño is characterized by unusually warm ocean temperatures in the Equatorial Pacific, as opposed to La Niña, which is characterized by unusually cold ocean temperatures. El Niño is an oscillation of the ocean atmosphere system in the tropical Pacific having important consequences for weather around the globe

It is apparent that this year's mass-bleaching event and El Niño might be the main reason why this project failed. Because of these special circumstances it is difficult to say anything about the approach and success of the project. It has to be carried out again under "normal" conditions in order to say much more over this approach.

Of course there were also many mistakes during carrying out the project which can be fixed easily. It would be possible to have good water quality in the buckets if it was changed more often and with water from further out and deeper. Another option is a constant water change with the help of a pump. Because of the fact that the bay in Chalok Baan Kao is very shallow, this approach would be useless because the water has to be pumped over a long distance. In Ao Leuk, this method would be making more sense.

With the help of the water-quality data we already got a lot of information about the quality in the buckets. However, the data does not show the amount of zooxanthellae in the buckets. These algae are very important for corals and it should be measured if they are available for the corals in the buckets. Because if not, the larvae should be exposed to zooxanthellae just prior to settlement since experience has shown that uptake is highest around the period of settlement competency and in the short period after the formation of the first skeletal elements (2-3 days after settlement) (True, 2009).

Another point of discussion is, if there is a stage missing between the providing and the transferring stage. When the coral larvae have settled down, they are moved out to coral nurseries in the sea. However, until they have grown a little bit and built up their skeleton, they are quite weak and not resilient against outer disturbances. Because of this, another stage can be introduced between the providing and the transferring stage. During this stage, the coral larvae are moved out of the bucket into big tanks with seawater, where they are cultivated in an artificial created environment where control factors as temperature, pH etc are controlled. This will protect the corals against outer unpredictable stress effects, like bleaching events. They will stay in there until they have built up their skeleton. However, the benefit of this tank is tested at the moment by Dr. James True.

After carrying out the Coral Spawning Project twice and after discussing it, it is time to ask if this project meet the needs of the New Heaven Reef Conservation Program. Of course it is impossible to do everything the right way during the first trial and there can be many improvements done. However, as the water testing results of the buckets and open sea depicts, is the contrast between them still very big. The project has been tested only in laboratories or with high-tech equipment and without them it is hard to keep the water quality high and have success (Zahir, 2010). Apparently, this method is still too scientific and it is very difficult to translate it into the needs, knowledge, skills and especially budget of a community-based group. There is no need for them to have a laboratory. In contrast to scientists who want to study corals, they want to restore and create an environment that works (Scott, 2010). It has to work inside the sea. An approach for this is the settlement under the sea option. The difference here is the fact that the coral larvae are not transported to the land, but are cultivated on special coral nurseries, which keep the larvae captured, but in the open sea. In these nurseries the coral larvae will float for a few days until they settle down onto the settlement plates at the bottom of the coral nurseries. The open sea culture facilities are a very easy, low-maintenance method for growing corals, although vulnerable to adverse environmental conditions (True, 2009). This means that with this approach the coral larvae would not have survived this year's bleaching event either. However, this is a viable alternative to the construction of large ex situ facilities, particularly for local community groups (True, 2010).

## Conclusion and advice

The New Heaven Reef Conservation Program has now carried out this Coral Spawning Project twice. Unfortunately, it seems that until now new corals could not be cultivated - possibly due to failure in the implementation (i.e. constant water changes) and of course due to the mass bleaching event this year. However, that does not mean that the whole project was useless. There was already a lot of progress done after carrying out the Coral Spawning Project and more improvements can be implemented easily as well as the same faults can be avoided. With another approach (i.e. settlement under the sea) everything would start at zero again, therefore, the Coral Spawning Project should be carried out again.

Some of the most important issues and improvements are again listed here.

During the initial stage it is recommended to put the settlement plates into the water much longer before the coral spawning in order to have a proper biofilm with more coralline algae on them. Another important issue is the communication between the divers. By improving it, fewer divers could be used much more efficient during the harvesting stage. Furthermore, it is necessary to give the divers and other helpers a proper briefing beforehand about the importance of the project, the tasks and how to carry them out in order to bring everybody on the same level of knowledge.

The water quality data of the buckets shows that the water quality in the clear bucket is slightly better than in the blue one. Furthermore, the larvae in the blue bucket settled a few days later. Because of all this, the clear bucket seems to be more useful as a cultivation tank. It is also important to plan the water changes very well with help of a schedule. A reason for this is to be able to split the water changes up between more people and to have more security that the water changes are really carried out. The fresh seawater for the water change should be taken from further out and deeper in order to have a good quality.

After transferring the coral larvae onto the nurseries they need to live in symbiosis with fish and invertebrate grazer species which consume competitive algae. Because of this it is useful to move the coral nurseries into waters with high fish abundance. Furthermore, the water has to be low in pollution and sedimentation. Because of this a good position of coral nurseries is near coral reefs, where sediment is closed up by corals and nutrients filtered out of the water by sponges and giant clams.

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## List of Appendixes

- Appendix I:  
depicts the outcome of the water quality testing, divided into three tables corresponding to the three buckets (clear, blue and control bucket)
- Appendix II:  
shows the result of the Sediment Trap Project of the bays Chalok Baan Kao, Ao Leuk and the Biorock

## Appendix I

Water quality testing - Clear Bucket							
	Time	Temp in °C	pH	PO <sub>4</sub> <sup>3-</sup>	NO <sub>3</sub> <sup>-</sup>	Turbidity in FTU	
06.04.2010	6.4.10 9:00	28,6	8,13				
	6.4.10 9:30	29,1	8,08				
	6.4.10 16:30	31,8	8,18	0,13	1,4	1	
	6.4.10 17:00	30,8	8,23	0,32	1		
	07.04.2010	7.4.10 9:00	29,5	8,21	0,1	1,5	11
		7.4.10 9:30	29,8	8,22	0,11	0,9	
	7.4.10 16:30	32,9	8,16				
	7.4.10 17:00	32	8,16	0,05	0,8	2	
	08.04.2010	8.4.10 9:00	29,8	8,18	0,11	0,7	7
		8.4.10 9:30	30,3	8,14	0,43	0,9	0
	8.4.10 16:30	30,6	8,16	0,07	1,2	3	
	8.4.10 17:00						
	09.04.2010	9.4.10 9:00	28,9	8,24	0,08	0,9	1
		9.4.10 9:30					
	9.4.10 16:30	31,3	8,24	0,07	0,9	5	
	9.4.10 17:00						
	10.04.2010	10.4.10 9:00	30,1	8,25	0,11	0,9	0
		10.4.10 9:30					
	10.4.10 16:30	31,4	8,3	0,44	1	0	
	10.4.10 17:00						
	11.04.2010	11.4.10 9:00	30,4	8,29	0,07	0,9	0
		11.4.10 9:30					
	11.4.10 16:30	31,2	8,3	0,12	0,8	6	
	11.4.10 17:00						
	12.04.2010	12.4.10 9:00	29,8	8,27	0,07	0,9	1
		12.4.10 9:30					
	12.4.10 16:30	30,6	8,24	0,1	0,6	2	
	12.4.10 17:00						
	13.04.2010	13.4.10 9:00					
		13.4.10 9:30					
	13.4.10 16:30						
	13.4.10 17:00						
	14.04.2010	14.4.10 9:00	30,9	8,12	0,28	1,4	8
		14.4.10 9:30					
	14.4.10 16:30	31,2	8,11	0,2	0,8	5	
	14.4.10 17:00	31	8,21	0,13	0,8	3	
	15.04.2010	15.4.10 9:00	30,4	8,24	0,37	0,9	0
15.4.10 9:30		31,6	8,21	0,12	0,7	0	

	15.4.10 16:30					
	15.4.10 17:00					
16.04.2010	16.4.10 9:00	30,6	8,27	0,06	0,7	0
	16.4.10 9:30	31,4	8,24	0,01	0,8	0
	16.4.10 16:30					
	16.4.10 17:00					
17.04.2010	17.4.10 9:00					
	17.4.10 9:30					
	17.4.10 16:30					
	17.4.10 17:00					
18.04.2010	18.4.10 9:00	30,3	8,31	0,16	0,9	0
	18.4.10 9:30	31,1	8,24	0,03	0,9	0
	18.4.10 16:30					
	18.4.10 17:00					
19.04.2010	19.4.10 9:00	30	8,27	0,11	0,8	0
	19.4.10 9:30					
	19.4.10 16:30					
	19.4.10 17:00					
AVERAGE		30,6	8,2137931	0,1480769	0,9230769	

Water quality testing - Blue bucket						
	Time	Temp in °C	pH	PO <sub>4</sub> <sup>3-</sup>	NO <sub>3</sub> <sup>-</sup>	Turbidity in FTU
06.04.2010	6.4.10 9:00	28,8	8,07			
	6.4.10 9:30	29,1	8,06			
	6.4.10 16:30	31,9	8,08	0,1	1	7
	6.4.10 17:00	31,1	8,16	0,09	0,9	
07.04.2010	7.4.10 9:00	29,1	8,15	0,07	1,3	9
	7.4.10 9:30	29,6	8,15	0,08	1,2	
	7.4.10 16:30	32,7	8,08			
	7.4.10 17:00	32	8,1	0,04	0,9	2
08.04.2010	8.4.10 9:00	29,9	8,1	0,13	1	2
	8.4.10 9:30	30,7	8,08	0,18	0,9	0
	8.4.10 16:30	32,7	8,06	0,05	0,8	2
	8.4.10 17:00					
09.04.2010	9.4.10 9:00	28,8	8,13	0,07	1	0
	9.4.10 9:30					
	9.4.10 16:30	30,9	8,08	0,17	1,1	7
	9.4.10 17:00					
10.04.2010	10.4.10 9:00	30,3	8,12	0,05	0,9	0

	10.4.10 9:30					
	10.4.10 16:30	31,4	8,11	0,14	0,9	0
	10.4.10 17:00					
11.04.2010	11.4.10 9:00	30,6	8,1	0,21	1	0
	11.4.10 9:30					
	11.4.10 16:30	31,2	8,03	0,13	0,7	3
	11.4.10 17:00					
12.04.2010	12.4.10 9:00	30	7,94	0,16	0,9	2
	12.4.10 9:30					
	12.4.10 16:30	30,6	7,92	0,17	1	8
	12.4.10 17:00					
13.04.2010	13.4.10 9:00					
	13.4.10 9:30					
	13.4.10 16:30					
	13.4.10 17:00					
14.04.2010	14.4.10 9:00	30,8	7,91	0,51	1,8	8
	14.4.10 9:30					
	14.4.10 16:30	31,1	7,82	0,55	0,9	6
	14.4.10 17:00	31,1	7,96	0,17	0,9	6
15.04.2010	15.4.10 9:00	30,4	8,11	0,14	0,7	0
	15.4.10 9:30	31,1	8,12	0,11	0,7	0
	15.4.10 16:30					
	15.4.10 17:00					
16.04.2010	16.4.10 9:00	30,8	8,18	0,16	0,7	0
	16.4.10 9:30	31,1	8,2	0,16	0,8	0
	16.4.10 16:30					
	16.4.10 17:00					
17.04.2010	17.4.10 9:00					
	17.4.10 9:30					
	17.4.10 16:30					
	17.4.10 17:00					
18.04.2010	18.4.10 9:00	30	8,29	0,21	1	0
	18.4.10 9:30	31	8,22	0,05	1	0
	18.4.10 16:30					
	18.4.10 17:00					
19.04.2010	19.4.10 9:00	29,8	8,26	0,12	0,9	0
	19.4.10 9:30					
	19.4.10 16:30					
	19.4.10 17:00					
AVERAGE		30,641	8,089	0,155	0,958	

Water quality testing - Control Bucket						
	Time	Temp in °C	pH	PO <sub>4</sub> <sup>3-</sup>	NO <sub>3</sub> <sup>-</sup>	Turbidity in FTU
06.04.2010	6.4.10 9:00					
	6.4.10 9:30					
	6.4.10 16:30	31,6	8,34	0,37	1,1	2
	6.4.10 17:00					
07.04.2010	7.4.10 9:00	29,2	8,26	0,03	1,4	3
	7.4.10 9:30					
	7.4.10 16:30	32,9	8,18	0,01	1	4
	7.4.10 17:00					
08.04.2010	8.4.10 9:00	30,2	8,15	0,25	0,9	2
	8.4.10 9:30					
	8.4.10 16:30					
	8.4.10 17:00					
09.04.2010	9.4.10 9:00	28,7	8,14			
	9.4.10 9:30					
	9.4.10 16:30	30,7	8,1			
	9.4.10 17:00					
10.04.2010	10.4.10 9:00					
	10.4.10 9:30					
	10.4.10 16:30					
	10.4.10 17:00					
11.04.2010	11.4.10 9:00					
	11.4.10 9:30					
	11.4.10 16:30	31,1	8,01			
	11.4.10 17:00					
12.04.2010	12.4.10 9:00					
	12.4.10 9:30					
	12.4.10 16:30					
	12.4.10 17:00					
13.04.2010	13.4.10 9:00					
	13.4.10 9:30					
	13.4.10 16:30					
	13.4.10 17:00					
14.04.2010	14.4.10 9:00					
	14.4.10 9:30					
	14.4.10 16:30	31,9	8,32	0,21	0,9	0
	14.4.10 17:00					
15.04.2010	15.4.10 9:00	32,9	8,2	0,16	0,8	0
	15.4.10 9:30					
	15.4.10 16:30					

	15.4.10 17:00					
16.04.2010	16.4.10 9:00	31,2	8,22	0,04	0,7	0
	16.4.10 9:30					
	16.4.10 16:30					
	16.4.10 17:00					
17.04.2010	17.4.10 9:00					
	17.4.10 9:30					
	17.4.10 16:30					
	17.4.10 17:00					
18.04.2010	18.4.10 9:00					
	18.4.10 9:30					
	18.4.10 16:30					
	18.4.10 17:00					
19.04.2010	19.4.10 9:00					
	19.4.10 9:30					
	19.4.10 16:30					
	19.4.10 17:00					
AVERAGE		31,040	8,192	0,153	0,971	

## Appendix II

### Sedimenttrap

#### Ao Leuk

Date	1st pot	2nd pot	3rd pot	total	average	per day	average per day
01 April 2010	cleaning the pots						0,251
08 April 2010	0,8	0,59	0,87	2,26	0,753	0,323	
18 April 2010	1,03	1,02	1,03	3,08	1,027	0,308	
27 May 2010	0,41	0,2	0,42	1,03	0,343	0,026	
10 June 2010	1,4	1,21	2,23	4,84	1,613	0,346	

#### Chalok Baan Kao

Date	1st pot	2nd pot	3rd pot	total	average	per day	average per day
18 April 2010	cleaning the pots						0,076
30 April 2010	0,21	0,32	0,19	0,72	0,240	0,060	
17 May 2010	0,53	0,67	0,38	1,58	0,527	0,093	

0

#### Biorock

Date	1st pot	2nd pot	3rd pot	total	average	per day	average per day
30 March 2010	cleaning the pots						0,086
07 April 2010	0,57	0,67	0,63	1,87	0,623	0,234	
16 April 2010	0,15	0,1	0,09	0,34	0,113	0,038	
27 April 2010	0,16	0,14	0,15	0,45	0,150	0,041	
05 May 2010	0,06	0,06	0,06	0,18	0,060	0,023	
19 May 2010	0,25	0,17	0,12	0,54	0,180	0,039	
28 May 2010	0,17	0,18	0,17	0,52	0,173	0,058	
09 June 2010	cleaning the pots						
16 June 2010	0,45	0,37	0,38	1,2	0,400	0,171	