Global Environment Research Fund of the Ministry of the Environment

"Study on the disturbance and restoration of the coral reef"

Manual for restoration and remediation of coral reefs

Manual for restoration and remediation of coral reefs

Edited by Makoto Omori and Shuichi Fujiwara

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Ministry of the Environment

Nature Conservation Bureau

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Forward

Coral reefs have been called “tropical forests of the sea”, with high productivity and biodiversity underpinning their strong economic and social values as sources of food, places for recreation and with recognition also of their importance as a reservoir of genetic resources. The protection that reefs provide tropical coastlines is evident during storms and tsunamis, where reefs dissipate wave energy quickly and efficiently. However, because environmental conditions affecting reefs in many tropical and subtropical regions has deteriorated significantly during the past four decades, due to causes such as outbreaks of the predatory crown-of-thorns starfish (*Acanthaster planci*), sedimentation by terrestrial run-off, and destructive fishing by dynamite or poison, conservation of the reef has become an urgent international issue. Consequently, Japan, USA, Australia and other countries concerned about coral reefs collaborated to establish, in 1995, the International Coral Reef Initiative, which Japan has driven forward proactively.

Japan has approximately 96,000 ha of coral reefs, distributed mainly around the Ryukyu Islands. Since Japan also is one of the few developed countries with significant reef areas, we have a major role to play in the fields of reef research and conservation to provide a positive contribution in tropical and subtropical waters. In pursuing such objectives, the Ministry of the Environment conducted studies on the disturbance and restoration of coral reefs via the Global Environment Research Fund project during fiscal years 2000 to 2002. The Ministry actively promoted new research on sedimentation impacts and also the process of coral settlement and recruitment with regard to the coral transplantation, to obtain fundamental knowledge of transplantation. In addition to the Ministry's activities, other research bodies such as Akajima Marine Science Laboratory and Japan Marine Science and Technology Center (JAMSTEC) have made efforts, including various investigations and experiments for the purpose of the coral reef restoration, to significantly advance our knowledge.

This document has been prepared in recognition of an increasing need for information on restoration of degraded coral reefs, through means such as transplantation, in order to provide information that may be helpful for coral reef restoration activities worldwide. It is a cooperative effort of the Japanese scientists who are playing principal roles in the research, who have summarized their accomplishments and knowledge for this manual.

We express our heartfelt gratitude to those who have been fully cooperative with us for the preparation of this document, and desire earnestly that it is utilized widely to be helpful for the regeneration and restoration of the coral reefs.

Nature Conservation Bureau

Ministry of the Environment, Japan

Editors

Makoto Omori

Akajima Marine Science Laboratory

Shuichi Fujiwara

Metocean Environment Inc.

Authors

Shuichi Fujiwara

Metocean Environment Inc.

Masayuki Hatta

Ochanomizu University

Seiichi Hosoya

Metocean Environment Inc.

Kenji Iwao

Akajima Marine Science Laboratory

Satoshi Nojima

Amakusa Marine Biological Laboratory, Kyushu University

Mineo Okamoto

Tokyo University of Marine Science & Technology

Nami Okubo

Tokyo Institute of Technology

Makoto Omori

Akajima Marine Science Laboratory

Hiroki Taniguchi

Akajima Marine Science Laboratory

Contents

1. Conservation, restoration and remediation of coral reefs: Background and significance

Deterioration and destruction of global coral reefs have caused increasing anxiety for several decades. For the regeneration and restoration of coral reefs, the maintenance and recovery of acceptable environmental conditions are essential, thus requiring social recognition for this serious issue, including upgrading of popular support, and legal controls, recommendations and cooperation that are supported by the administrations willing to protect the coral reefs. Natural death of corals from typhoon or abnormal rise of water temperature can be corrected within several years or in a decade if their rearing environmental conditions become acceptable. However, because reefs can be degraded far faster than they can grow back, due to human caused stresses such as eutrophication and terrestrial run off, we have seen a net reduction of reefs for decades.

In the environment marked by such circumstances, various techniques have been developed for restoration of the coral reefs. But, the development of such techniques should not merely assume that the destruction of the coral reefs from the growth of human activities is inevitable. Restoration techniques should always facilitate the improvement of the surrounding environment as well as the reefs. This is a point to be noted in the beginning. In Okinawa where restoration of the coral reef is the most urgent demand, many reefs are still sacrificed and lost by human development activities.

As one of the coral reef restoration techniques, the transplantation of coral fragments has been implemented in Japan from the 1990s in various locations. On one hand, when a coral reef was removed, due to the necessity of port and harbor or other underwater construction, the transplantation (relocation) of whole colony or a part of the coral reef that is cut off to other location was performed. However, many of the past transplantation projects were performed without sufficient scientific verification and/or long term monitoring of the results. The projects did not always lead to advance of the restoration techniques because their achievements were not analyzed. Some restoration projects and development of techniques are in the stage of experiment or trial.

In this circumstance, we have decided to include the preparation of this manual for coral reef restoration in the process of study of the promotion of restoration of the coral community, one of the Ministry of the Environment’s Global Environment Research Fund Projects, to disclose the achievements and knowledge with regard to the restoration techniques that have been developed mainly in Japan. This document includes the collection of the achievements that were already published and those considered disclosable by the author of each chapter. For the achievements that are in the stage of trial or experiment, that is, state of the art, or those contained in patents, they are simply outlined. Research and development for the coral reef restoration have been advanced by individual institutes independently. The editors believes that the publication of this document that collects information about the present situation and outlook of the restoration of coral reefs will lead to greater mutual understanding of the relevant persons, and then, the exchange of the information

can cause further development of their activities.

For the compilation of this document, the editors acquired advice and instructions from many persons including Mr. S. Yamamoto, Ecoh Inc. and Dr. T. Hayashibara, Ishigaki Tropical Station, Seikai National Fisheries Research Institute. We would like to acknowledge these persons for their special favors. We would like to express our special thanks to Mr. G. Sweany, U.S. Foreign Service (ret.), and Dr. A. Heyward, Australian Institute of Marine Science, for their kind reading of the English manuscript and for providing many useful suggestions.

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2. Previous research and undertaking of coral reefs restoration

This documents collects the methods, achievements, and problems of measures including 1) seeding production and settlement induction by utilizing the coral sexual reproduction, 2) transplantation of coral fragments by utilizing asexual reproduction, 3) transplantation of colonies or entire reef, and 4) management of settled seeding, transplanted colonies and coral communities.

For the hermatypic corals, their larvae spawned by the sexual reproduction settle on the substrate and metamorphose in due time, and when their polyps are nearly completed, they start to make clonal polyps to form colonies. The colonies continue to bud to grow while increasing the polyps, and a part of the colonies broken by the force of the wave from storm or other sources may settle on the substrate again, where they grow by the asexual reproduction. An attempt to restore the coral reefs through transplantation of the coral fragments using layman's knowledge has been performed from the 1980's elsewhere than Japan, and from the 1990's in Japan. Regarding transplantation of the fragments, they can be fixed directly on the base rock in destination waters or settled on the substrate until they grow and then relocated to destination waters. There is another, patented a technique that hangs a piece of rope with the fragments into the sea where they have grown (Japanese published application 06-303875). However, the past attempts so far have not sufficiently investigated elements of fragment relocation such as suitable species, sites, methods and the size of fragments, and no long term monitoring has been made much for the state of the corals after the transplantation and ecological change in the periphery of the transplantation sites. Since it is expected that the advance of the transplantation techniques will lead to the restoration of coral reefs, creation of artificial coral reefs, and production and rearing of ornamental coral colonies that can possibly cause reduction of collection of the natural corals, the information should be enriched. In Japan, however many of the transplantations were carried out without sufficient scientific planning and the knowledge obtained from the activities is not collected, and thus, positive activities did not necessarily contribute to the advance of the transplantation techniques. The method is accompanied by big problems including criticisms against the method that obtains the fragments by breaking the live coral colonies (hereafter called "donor"), difficulty of standardizing of the method, and high need labor and other costs for large scale transplantation.

The followings are review of articles in the world with regard to transplantation of coral fragments based on literatures regarding 71 species in 12 families (revised after Okubo and Omori 2001). Those that were transplanted the most was the species of the genus *Acropora* of the Family Acroporidae (see Table 2-1). In Japan, *Acropora formosa*, which is generally seen in the moats that are affected by relatively strong waves, and branching corals such as *A. nobilis* which is seen in the moats and shallow places of reef slopes that are affected by weak waves are most commonly transplanted. Results of the fragment transplantation (by species) that were conducted by the Marine

Parks Center of Japan (1995) in the Sekisei Lagoon and Okinawa General Bureau (unpublished) at Naha Harbor are shown in Figs. 2-1 and 2-2.

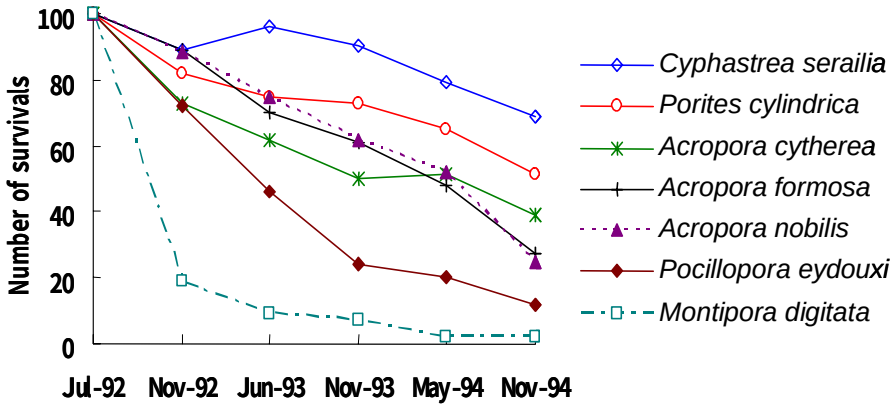

Fig. 2-1. Change in the number of survivors of seven species transplanted on the St. 9 off the north of Kuroshima Harbor, Okinawa (Marine Parks Center of Japan 1995).

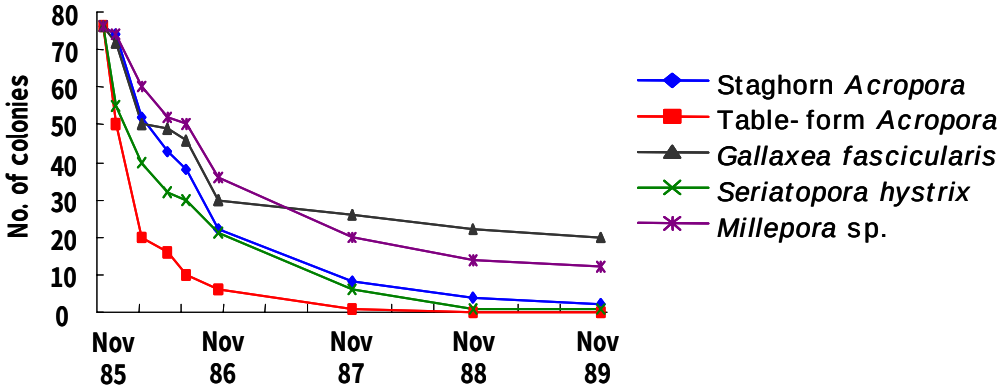

Fig. 2-2. Change in the number of survivors of different species transplanted at Naha Harbor, Okinawa (Okinawa General Bureau, unpublished data)

Results are generally not very outstanding. Although records of long term monitoring are not many, average survival rates (hereafter referred to as the rate) after 4 years are about 20 %. There is high rate for *A. formosa*, however. The fragments were transplanted two times and the rates were 69% at 43rd month after the first transplantation and 100% at 17th month after the second transplantation (Okinawa General Bureau, Okinawa Development Agency 1997). It is reported that the rate of *A. nobilis* at 18th month after the transplantation was 82% (Japan Marine Science and Technology Center 1991). In both species higher survival rates were those transplanted in shallow waters with strong tidal current, good water quality, and little ocean waves. Lower rates were recorded in turbid areas with weak tidal current that are separated from open sea at low tide (Marine

Parks Center of Japan 1993, 1994, 1995). For the branching form *A. microphthalma*, the rate was 100% at the 17th month, being the species with high aptness for transplantations (Okinawa General Bureau, Okinawa Development Agency 1997). The rate of the corymbose form *A. prominens* was 51.8% at the first year (Auberson 1982); for *A. humilis* 79.9% at the second year (Clark and Edwards 1995). For *A. tenuis* that was transplanted by 1, 2 and 4 colonies separately in nearly the same environment, the rate at second year varied to 100%, 50%, 25% respectively (Clark and Edwards 1995). The rates at three different locations using tabular coral *A. hyacinthus* were 24% at the first year (Japan Marine Science and Technology Center 1991), 44% at 17th month (Plucer-Rosario and Randall 1987), and 49% at the second year (Clark 1997). The rate of *A. divaricata* was 80.6% at the second year with considerably high growth rate (Clark and Edwards 1995). The rate of *A. cytherea* was 50% at the second year (Clark and Edwards 1995).

The rates of *Montipora foliosa* and *M. stellata* that were transplanted were both 100% at 15th month (Okinawa General Bureau, Okinawa Development Agency 1997). That of *M. digitata* was over 80% at the first year at a moat, but the rate was around 10% in the locations affected by strong ocean waves and tidal current (Marine Parks Center of Japan 1995).

Pavona frondifera of Agariciidae grew at higher rate and was tolerant against bleaching event (Yap et al. 1992). As compared to this, *Leptoseris gardineri* showed low rate at any experiment site ranging from shallow to deep waters (Plucer-Rosario and Randall 1987).

For Faviidae, the rate of *Echinopora lammellos* was 80% on coral rocks at the 3rd month (Kaly 1995). All *Leptastrea purpurea* and *L. transversa* died at 28th month later (Clark and Edwards 1995). *Galaxea fascicularis* of Oculinidae and *Pavona frondifera* and others also showed relatively high rate (Japan Marine Science and Technology Center 1991；Clark and Edwards 1995；Plucer-Rosario and Randall 1987).

For the rate of Pocilloporidae, *Pocillopora damicornis* were 100% at 7th month (Harriott and Fisk 1988) and 17th month (Okinawa General Bureau, Okinawa Development Agency 1997) respectively. This species is distributed widely in a variety of habitat environments. As a result of transplantation of *Seriatopora hystrix* to harbor, the rate was 13% at 43rd month (Okinawa General Bureau, Okinawa Development Agency 1997), and for *Stylophora pistillata*, the rate was 39% at 18th month (Japan Marine Science and Technology Center 1991). Both species are deemed to show low rate though they are pioneer species that establish in the shattered coral reef easily (Bouchon et al. 1981).

So far, the one that showed significantly higher rates was *Porites* spp. of Poritidae. The rate of *P. lichen* was 100%, *P. lobata* was 97%, and that of *P. nigrescens* was 84% (Clark and Edwards 1995). The rate of *P. lutea* was 92% at the 28th month (Clark and Edwards 1995). As a case of *P. cylindrical*, the transplantation to blocked water with more water flows showed good results (Marine Parks Center of Japan 1993, 1994, 1995). Another experiment in such water, the rate was 75% at the 5th month (Japan Marine Science and Technology Center 1991).

In the hermatypic corals other than Scleractinia, fragments of *Heliopora coerulea* of Helioporidae have been transplanted, and showed the rate of 100% at the first year (Auberson 1982). *Millepora dichtoma* of Milleporina, showed 68% at the first year, where as *M. platyphylla*, 58% at the first year (Auberson 1982).

In addition, a method has been proposed that electric current be applied to the wire which the fragments are attached for promoting deposition (mineralization) of the carbon ion and calcium ion contained in the seawater to facilitate the growth of the corals. Although there are some positive results in abroad (van Treek and Schuhmacher 1997, 1999; Schuhmacher et al. 2000), this experiment did not show outstanding results in Okinawa (Kudo and Yabiku 1988).

In the cases of the transplantation of whole colony and coral communities (entire reef), comprehensive survey on the depth, wave action and substrate of the transplantation site is essential, too, before selecting the new locations. Current knowledge is insufficient however. It is expected that the results of observation on the state of coral colony after transplantation will be positive. There are some report of coral colonies that were cut off together with the base rock and combined to create an artificial coral reef. In Japan, a coral reef is removed due to the necessity of port and harbor or other underwater construction, and the activities are limited to the relocation of a part of a coral reef that was cut off to a nearby site (Fukunishi et al. 1998).

The development of coral reef restoration techniques using sexual reproduction was brought up as a research theme at the Akajima Marine Science Laboratory at the beginning of the 1990's, and the supporting various fundamental biological observations and experiments were performed with collaboration from Dr. A. Heyward and Dr. P. Harrison of Australia, Dr. D. Morse and ANC. Morse of the US, and Dr. T. Sugiyama and Dr. M. Hatta of Japan. In this study, the corals were spawned in the laboratory, or embryos or larvae were collected from the slick or coral colonies on the sea that are found immediately after the simultaneous spawning, and then bred intermediately in a floating pond or water tank until the larvae start to settle on a suitable substrate, and then, they were transplanted to a suitable site. The study was performed under the sympathetic understanding and sponsorship of Nippon Foundation and others. In Australia, the above-mentioned Dr. A. Heyward and others succeeded in seeding production similar to the Akajima case (Heyward et al. 2002).

The studies so far revealed that some species of red algae such as the crustose coraline algae *Hydrolithon reinholdi* promote the settlement of larvae (Morse et al. 1996), and some species of neuro-peptide provide an effect that induces metamorphosis.

The method of intermediate rearing and that of promoting the settlement attained certain achievements. An attempt is made to raise the possibility of retention and to induce the settlement of larvae on the substrate by using ocean structures such as breakwaters with finely indented surfaces. The future research subjects include how to accelerate the growth of polyps after they settle on the substrate and to raise their survival. It is necessary to take measures for removing algae that grow on

the substrate and protecting the polyps from coral eating fish.

Table 2-1. List of coral species used for transplantation of coral fragments

Scientific name	References
Family Acroporidae	
Acropora bruggemanni	Auberson (1982)
cervicornis	Becker and Mueller (1999); Bowden-Kerby (1997); Chilcoat et al. (2000)
conferta	Chen and Xiong (1995)
corymbosa	Chen and Xiong (1995)
cytherea	Clark and Edwards (1995); Marine Parks Center of Japan (1993, 1994, 1995)
digitifera	Clark and Edwards (1995)
divaricata	Clark and Edwards (1995)
echinata	Plucer-Rosario and Randall (1987)
formosa	Okinawa General Bureau, Okinawa Development Agency (1997); Marine Parks Center of Japan (1993, 1994, 1995); Lindahl (2000); Thongtham (2000)
gemmifera	Clark and Edwards (1995); Kaly (1995); Japan Marine Science and Technology Center (1991)
humilis	Clark and Edwards (1995)
hyacinthus	Ashizuri-Uwakai National Park (personal communication); Clark and Edwards (1995); Japan Marine Science and Technology Center (1991); Okinawa General Bureau, Okinawa Development Agency (1997); Yap et al. (1992)
microphthalma	Okinawa General Bureau, Okinawa Development Agency (1997)
nasuta	Okinawa General Bureau, Okinawa Development Agency (1997)
nobilis	Japan Marine Science and Technology Center (1991); Okinawa Commemorative National Government Park Office (1999); Nishihira (1994); Okinawa General Bureau, Okinawa Development Agency (1997)
palmata	Becker and Mueller (1999); Iliff (1999); Bruckner and Bruckner (2001); Garrison (2002)
prolifera	Bowden-Kerby (1997)
prominens	Auberson (1982)
prostrata	Chen and Xiong (1995)
pulchra	Yap and Gomez (1984)

tenuis	Clark and Edwards (1995)
tumida	Awa Takegashima Marine Park (personal communication)
Montipora digitata	Marine Parks Center of Japan (1993, 1994, 1995)
foliosa	Okinawa General Bureau, Okinawa Development Agency (1997)
prolifera	Auberson (1982)
pulcherrima	Plucer-Rosario and Randall (1987)
Family Agariciidae	
Pavona cactus	Plucer-Rosario and Randall (1987); Marine Parks Center of Japan (1993, 1994, 1995)
frondifera	Yap et al. (1992)
lata	Chen and Xiong (1995)
Leptoseris gardineri	Plucer-Rosario and Randall (1987)
Pachyseris rugosa	Chen and Xiong (1995)
Family Astrocoeniidae	
Stephanocoenia bournoni	Dodge et al. (1999)
Family Faviidae	
Cyphastrea serailia	Marine Parks Center of Japan (1993, 1994, 1995)
Diploria clivosa	Dodge et al. (1999)
labyrinthiformis	Dodge et al. (1999)
Echinopora lamellose	Kaly (1995)
Favia matthai	Chen and Xiong (1995); Marine Parks Center of Japan (1993, 1994, 1995)
speciosa	Clark (1997)
stelligera	Kaly (1995)
Goniastrea aspera	Chen and Xiong (1995); Clark (1997)
reticulosa	Clark and Edwards (1995)
Leptastrea purpurea	Clark and Edwards (1995)
transversa	Clark and Edwards (1995)
Montastrea annularis	Gil-Navia (1999); Sanchez et al. (2000)
cavernosa	Dodge et al. (1999)
faveolata	Becker and Mueller (1999); Dodge et al. (1999)
Platygyra rustica	Chen and Xiong (1995)
Family Meandrinidae	
Meandrina meandrites	Dodge et al. (1999)
Family Mussidae	
Symphyllia agaricia	Chen and Xiong (1995)

Family Oculinidae	
Gallaxea fascicularis	Chen and Xiong (1995); Japan Marine Science and Technology Center (1991)
lamarcki	Chen and Xiong (1995)
Family Pocilloporidae	
Madracis decactis	Dodge et al. (1999)
Pocillopora brevicornis	Chen and Xiong (1995)
elegance	Eakin (2000)
damicornis	Chen and Xiong (1995); Clark and Edwards (1995); Harriott and Fisk (1988a); Okinawa General Bureau, Okinawa Development Agency (1997); Yap et al. (1992); Eakin (2000)
danae	Chen and Xiong (1995)
eydouxi	Marine Parks Center of Japan (1993, 1994, 1995)
verrucosa	Japan Marine Science and Technology Center (1991); Chen and Xiong (1995); Clark and Edwards (1995)
Seriatopora hystrix	Okinawa General Bureau, Okinawa Development Agency (1997)
Stylophora pistillata	Japan Marine Science and Technology Center (1991); Kaly (1995)
Family Poritidae	
Porites astreoides	Dodge et al. (1999); Gil-Navia (1999)
cylindrica	Japan Marine Science and Technology Center (1991); Marine Parks Center of Japan (1993, 1994, 1995)
lichen	Clark and Edwards (1995)
lobata	Clark and Edwards (1995); Clark (1997)
lutea	Clark and Edwards (1995); Thongtham (2000)
nigrescens	Clark and Edwards (1995)
Family Siderastreidae	
Siderastrea sidereal	Dodge et al. (1999)
Family Helioporidae	
Heliopora coerulea	Auberson (1982); Clark and Edwards (1995)
Family Milleporidae	
Millepora alcicornis	Clark and Edwards (1995)
dichotoma	Auberson (1982)
platyphylla	Auberson (1982)

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3. Restoration technology using sexual reproduction

3-1. Seed production

This chapter discusses mainly *Acropora* coral. On the coral reefs of the West Pacific Ocean including Okinawa, *Acropora* provides high species diversity and its existing amount is considerable. Since the growth rate of *Acropora* is high, it is considered to be an important coral for coral reef restoration. Since many *Acropora* corals spawn simultaneously, their initial loss is large, meaning that many of the spawn and embryos disappear before they settle. However, since it is effective for fertilizing corals to collect the eggs in the period immediately after the spawning to the next day and breed them as the seeding animals.

(1) Sampling of eggs and embryos from the sea

a. Prediction of simultaneous spawning

In Okinawa Prefecture, the corals mainly of *Acropora* show the simultaneous, mass spawning. The simultaneous spawning occurs during and around full moon, of each month mainly in May in the Yaeyama Islands (Hayashibara, personal communication), in June in the Kerama Archipelago (Hayashibara et al. 1993) and also at the Main island of Okinawa (Heyward et al. 1987) (See Fig. 3-1).

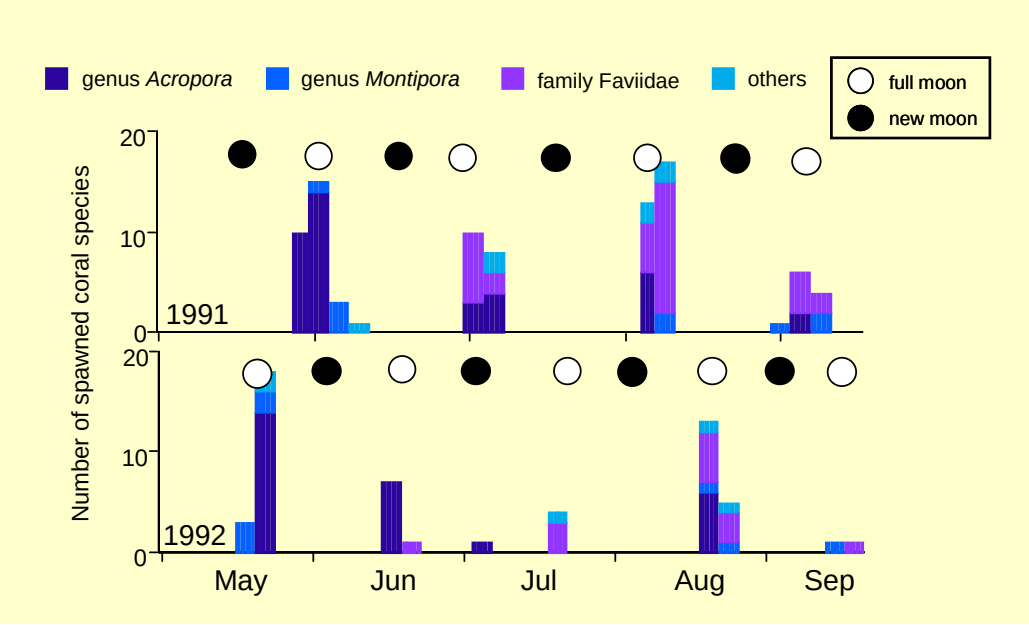


Fig. 3-1. Pattern of coral spawning at the Akajima Island, Kerama Archipelago
(Shows the number of coral species spawned in each of flood tide, middle tide and neap tide)

The period may be moved by one month or separated into two times depending on the lunar cycle and water temperature. When predicting the date, it is recommended to refer to the records of the

past spawning dates of each location. Since the shift of the date from the full moon is also affected by the water temperature, it is recommended to stand by for the spawning in the period from there days before to seven days after the full moon. The spawning date also can vary among locations in an island. To forecast the spawning date, break a branch of colony to examine the maturity of the spawn. The egg is colored as the spawning date comes nearer. The bundle appears in the mouth of polyps two or three hours before the spawning. Though the time of spawning varies a little depending on the time of sunset, the species such as *Acropora tenuis* spawns the most in the period from 19:00 hr to 19:30 hr, and for many other species, from 21:30 hr to 22:00 hr (Fukami et al. 2003). Some species spawn at around 20:00 hr or in August, however, the number of those species and population are not many.

b. Sampling eggs immediately after spawning

For *Acropora*, each polyp emits eggs and sperms as a block (called a bundle). The bundles are brought up to the sea surface by the buoyancy of the spawn. Then, the eggs and sperms dissociate from each other and meet the gametes that are emitted from other colonies to be fertilized. Therefore, by setting a funnel form bundle collecting device (bundle collector) under water and above the coral colony, the bundle can be collected easily (Kitada 2002). Schematic view of a typical bundle collector is shown in Fig. 3-2.

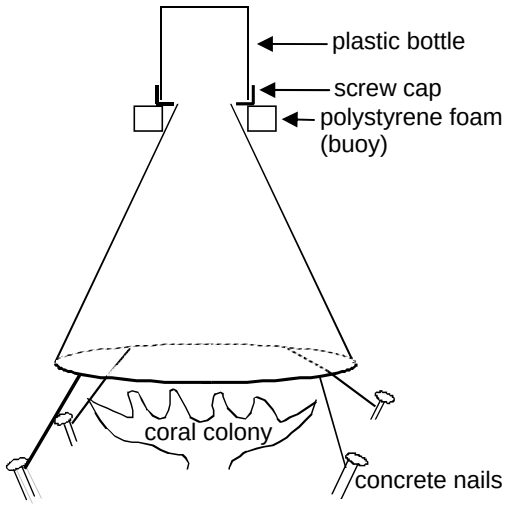


Fig. 3-2. Schematic view of bundle collector

Following are instructions for making the collecting device. Make a cone rigid with fine nylon mesh or vinyl sheet, and keep the conical form by holding the cone with stainless steel wires. Attach a plastic bottle (capacity of 500 ml to 1 liter) with a screwed cap and a foamed polystyrene float to the top of the cone. The float holds the bottle at the top of the cone. Use a bottle made of transparent

material so that the inside can be seen. Connect the bottle and the funnel together to make the bottle easy to attach / detach.

Put the bundle collecting device above the coral colony in every early evening in the predicted spawning period, and fasten it by using strings and concrete nails as shown. Watch the device during the predicted spawning time and, if no spawning is seen, set out the device again next day. Repeat this process until the spawning occurs. Leaving the device set on the site can lead to damage to the coral colony, and may cause loss of the device in case of worsening of the sea state. When the spawning occurs, remove the bottle immediately after the spawning has ended, close it with the cap, and bring it back to the laboratory. The eggs may die from lack of oxygen if it is left in the bottle in high density for long time.

Transfer the collected bundle to other container such as a bucket. Mix the bundles that are collected from 3 colonies or more per one species of coral to be fertilized. *Acropora* basically does not self-fertilize, and the fertilization of gametes from 2 colonies may cause low fertility depending on the combination of individuals. Also, it is possible that the bundle may be collected from concealed *Acropora* species that can hardly be identified based on the form. Thus, the collection of the bundles from many colonies is recommended for the purpose of making the fertilization successful.

The suitable density of gametes at insemination should be the density of sperms that makes the white turbidity visible (around 10^7/ml) and the floating eggs cover the water in the container uniformly. It is desirable that the insemination work be completed within four hours after the spawning. Mix the liquid at intervals. Leave it for one or two hours after mixing. Then, skim the spawn and transfer it to a larger rearing bowl (3～5 liter). Avoid mixing in left over sperms which can lead to deterioration of the water quality after death. The fertilized spawn (embryos) easily become abnormal or die at this time of the process.

For the corymbose type coral *Acropora tenuis*, three colonies of around 30cm diam. allow collection of approximately one million eggs (Shimomura et al. 2002).

c. Sampling eggs and embryos on the morning following simultaneous spawning

In many cases, aggregates of fertilized eggs (called “slicks”) of corals are formed on the morning following simultaneous spawning, which drift on the sea surface forming a long and narrow strip, though they are affected by the environmental factors such as the topography of the site, current and weather condition. In Okinawa where mainly fringing reefs exist, the slicks are washed on the shore in many cases because the coral reefs are located near the land. Since they drift ashore at high tide and die, it is necessary to collect them before they are washed on the shore. The development stage of *Acropora* coral in this period of time is morula, the blastomere of which is easily dissociated to death by wave impact on the pier or shore, thus selection of the sampling point(s) is also important.

The slick may drift ashore easier to some area of harbors. In this case, the slicks are preserved to around 10:00 hr in the morning (Fig. 3-3).

Fig. 3-3. Slicks drifting ashore to inside of a harbor

Fig. 3-4. Offshore slicks

The slicks may be found by searching on the sea using a boat (Fig. 3-4). The slicks that are formed offshore (where contamination materials from the land are less) are suitable for raising. It may be worth searching for slicks on other than the morning following simultaneous spawning at the investigation point, because the slicks from the simultaneous spawning at distant points may later

drift ashore at the investigation points, or the slicks may come alongside the beach on morning of the day after the next day of simultaneous spawning, which is rare. Conceivably, there will be years when the slicks cannot be found at all because of the weather conditions.

When collecting the slicks, skim them by using a dipper and put them in a bucket to carry. Since the simultaneous spawning consists mainly of *Acropora*, the embryos and larvae that are contained in the slicks are those of *Acropora*, but those of *Montipora* may be included. (The larvae of *Montipora* can be distinguished from those of *Acropora* because they are smaller than the *Acropora* larvae and contain zooxanthella.) It is necessary to verify the state of embryos by using a stereoscopic microscope. The survival may be unsuccessful if many dissociations of blastomere or unfertilized spawn are seen. When much foreign matter is mixed in, it is recommended that live embryos be selected using a pipette, which is a cumbersome work.

(2) Spawning induction

For small scale experimental research, the spawning can be conducted in the laboratory, or it is possible to induce the spawning artificially. Though there hardly exist successful cases of the artificial induction of coral spawning, apparently it can only be accomplished with some corals of *Acropora* and *Montipora* by using hydrogen peroxide. The method is as described below (Hayashibara et al. 1996; Ishigaki Tropical Station, Seikai National Fisheries Research Institute 1999).

For corymbose type *Acropora*, after putting the coral colonies collected from the sea, add 2mM of hydrogen peroxide for three hours or 5mM for two hours, and then, wait for the spawning while sustaining in the clean flowing water (Fig. 3-5). The spawning by the induction occurs in the period from 20:00 hr to 23:00 hr of the processing day under natural lighting, which is similar to the case of natural spawning in the open air. However, a period of at least 9 hours is needed from the induction to the spawning (initiation of spawning at higher probability requires 16 hours). If the estimated spawning time has come before the elapse of the 9 hour period, the spawning should not be performed that day, and is postponed to the same time of the next day.

Although it is possible to induce the spawning at a desired day and time by using a series of practices described above, this spawning induction method may give considerable damage to the coral colonies, which include secretion of mucus or emission of zooxanthella during the processing in the hydrogen peroxide, possibly leading to death. Since the degree of damage from the hydrogen peroxide varies among the state and species of the colonies, it is recommended to observe the state of colonies during the processing, and to adjust the density of the chemical and length exposure as necessary. Also, mortality can be reduced by rinsing the fragments to remove the hydrogen peroxide as soon as possible and then putting them in the clear flowing water. The spawning does not occur if the eggs of the colonies are immature. However, it was confirmed that the gametes obtained from

matured colonies are fertilized and become planula larvae, and then, metamorphose to be polyps (Hayashibara et al. 2003).

In addition to the above spawning induction method, the spawning time can be changed by adjusting the photo period. By making the environment dark by shielding the light with a camera obscura or other means after the elapse of 9 hours from the induction, the spawning can be initiated within 2 to 3.5 hours from the time (Iwao 2000).

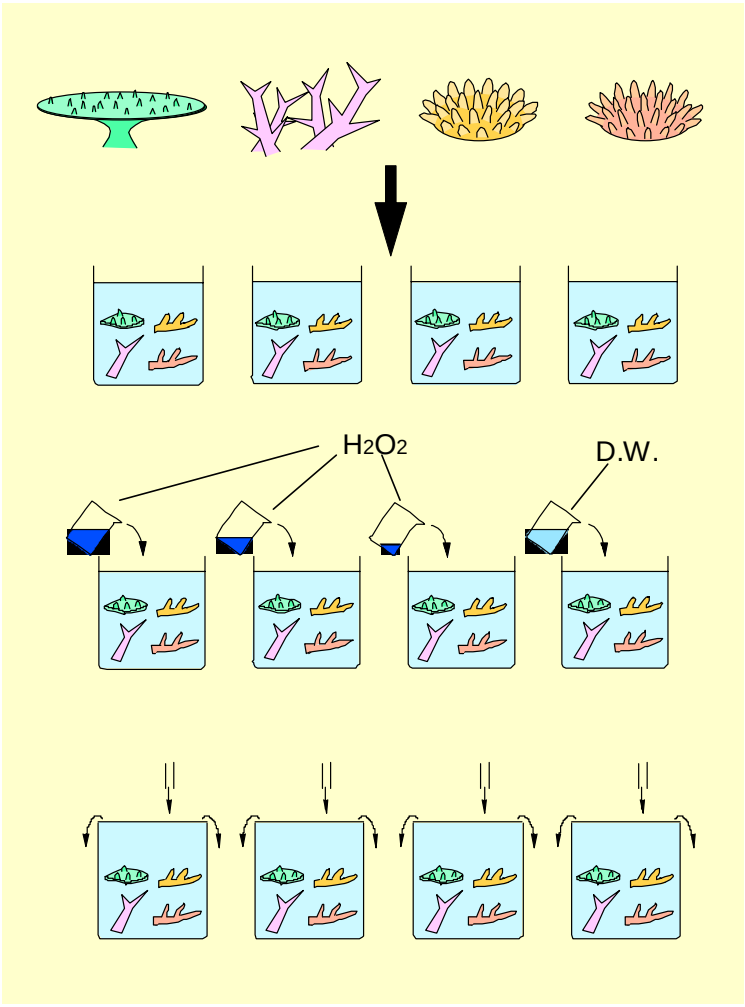


Fig. 3-5. Method of spawning induction of coral using hydrogen peroxide
(Ishigaki Tropical Station, Seikai National Fisheries Research Institute 1999)

(3) Maintenance and culture

It is possible to rear the larvae by using a bowl if they are held only for small scale experimental research. The larvae can be sustained in a good condition by transferring them to a new bowl using a pipette at a rate of once or more per day. It is desirable that the density of larvae to be reared be less than 2000 individuals per one liter of seawater. The standard period needed for commencement of settling is 6 days after spawning at a water temperature of 26ºC, or 5 days at 27ºC. The larvae of

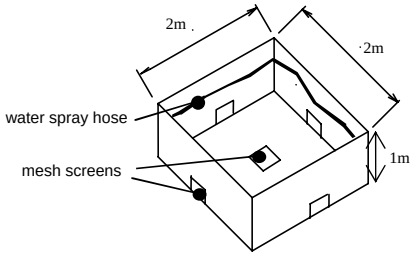
many species can sustain the settlement capability for approximately three weeks. The maturity of the larvae can be known by scattering the cells of larvae on the slide glass using maceration and counting the number of cnidae (Hayashibara et al. 2000). When performing large scale raising growth of over 100 thousand individuals, because the larvae are sustained without changing the water, the density must be lowered. The larvae that have been obtained using bundle collect could be diluted in large water tanks to raise easily, and it was possible to raise 500 thousand individuals per one ton of seawater (Shimomura et al. 2000) (Fig. 3-6). The water quality can easily be deteriorated by the foreign matter found among the larvae obtained from the slick and/or death of unfertilized spawn, and thus, growth at lower density is required. Since the embryos drift on the water surface for two days after the fertilization, the exchange of water can be made. The rise of water temperature from solar insolation is a problem when carrying out the rearing outdoors. It is necessary to keep the water temperature always less than 30ºC, and 27ºC or lower is desirable.

Fig. 3-6. Large scale rearing of larvae indoors

The measurement of density and total number of larvae raised can be ascertained by sampling approximately 0.1 liter of the seawater used for rearing and counting the number of larvae contained in the water. However, it is very difficult to obtain an accurate count, because the larvae are unevenly distributed on the water surface in the initial period of the generation and the distribution is influenced by the wind if outdoors. To prevent negative physical effects on embryos or initial larvae, avoid stirring and mixing water as much as possible within one day from the spawning. For any method, it is necessary to sample the water at least three to five times to evaluate the count in order

to obtain reliable values.

(4) Mass production of larvae



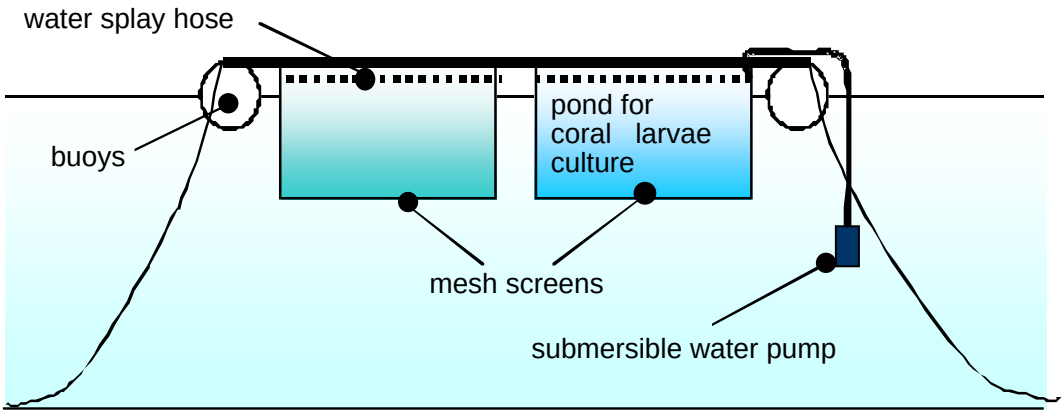


Fig. 3-7. Large-scale rearing of coral larvae using floating pond

Rearing of more larvae can be made by using floating ponds on the sea surface (Aota et al. 2002, Omori et al. 2003). The raft for segmented floating ponds on which the vinyl sheet used for eel farming is adopted as the water reserve for rearing larvae. A hose with holes made at intervals is run on the upper part of the reserve, using water taken by underwater pump from the sea around the raft. The water is sprayed against the side walls to prevent the larvae from attaching to them. The reserve is provided with water flowing through open screen on four sides and bottom to allow outflow of seawater entering from the hose. It is necessary to clean the screens (using divers) when they are clogged with micro algae or other matters. Aota et al. (2003) succeeded in rearing the larvae to over six million individuals by using eight sets of 2m square floating ponds (1.0 m deep) shown in Fig. 3-7. It is possible to breed larvae to 200－400 inds/l of seawater.

(5) Transportation

When the scale of rearing is small, the larvae can be sent to remote places using a general parcel delivery service or mailing by using a container such as a 1 liter plastic bottle. Akajima Marine Science Laboratory succeeded in sending larvae alive to the aquaria of Germany and Holland by using 50 ml tubes (Petersen and Tollrian 2001). When sending a large amount of larvae, a bucket with a lid or a plastic tank with a small mouth is used for short range transportation. A density of larvae similar to that for rearing causes no problem. For transportation that takes long time, however, lower the density, and seal the container with a little air space in it to reduce the stress from vibration during the transportation. When divers release the larvae in the water, a collapsible polyethylene container (10 liter or 20 liter) is suitable because the divers are able to discharge the water by squashing it. The recommended time for transportation is fourth day or after from the fertilization when the larvae ciliated and move actively, and have the tolerance against various stresses. In this time, the larvae at the density of 3000 ind/l or less can be transported (2-3 days) with survival rate higher than 90%.

(6) Inducement of settlement and metamorphosis

Though many of the mechanisms of settlement and metamorphosis of hermatypic corals are still unrevealed, Morse and Morse (1991) demonstrated that the settlement and metamorphosis of the coral *Agaricia humilis* larvae produced in Caribbean Sea are induced by eduction from *Hydrolithon boergenseii*, a kind of coralline algae. By further progressing the study, Morse et al. (1994) made a substrate for inducing the settlement called “flypaper” from a settlement induction fraction of coraline algae, which requires considerable labor. To induce the settlement using the coraline algae, employ the algal tissue itself or make the chips of algae and add them. The chips can be made by scraping the surface area of the algal tissue using a knife. They can be preserved for over one year if they are soaked in a beaker containing rifampicin water solution (2mg/l), washed, filtered to remove water from the chip, and then deep-frozen. The settlement occurs in 6 to 72 hours after adding the thawed chips and larvae to filtered seawater. In many cases, the larvae settle on the bottom near the chips or corners, and in some case, on the chips. It is demonstrated that the settlement of Japanese *Acropora* larvae can be induced when chips of *Hydrolithon reinboldii* are used (Morse et al. 1996). *Peyssonnelia* sp. that belongs to red algae, but is not a species of the coraline algae, can also be induced to settle at a level equivalent to the *Acropora* larvae or higher. In may cases, the settlement rate is 90% or lower (Iwao 1997). Although the settlement induction method using the coraline algae chips is relatively simple and easier, it may be difficult to obtain the algal tissue in some places, and scraping off chips from hard algal tissue may be laborious.

The use of neuro-peptide Hym-248 promotes higher and more stable settlement induction rate (Iwao et al. 2002; Hatta and Iwao 2003). Hym-248 is the isolation from freshwater hydra *Hydra*

magnipapillata (Cnidaria, Hydrozoa), which can be artificially synthesized to make the desired amount. When the larvae are added to the filtered seawater at the density of 1×10^{-6}M, it immediately reacts by contracting around body axis or rubbing the abactinal end on the substrate or container wall, and the settlement is completed within 12 hours from the addition of the peptide (Fig. 3-8). So far, Hym-248 has been effective only for *Acropora* larvae. The effect varies according to the age of the larvae, and the settlement rate is low when the larvae are too juvenile (4 days-old or less). As they get older, it is possible to achieve the settlement rate of 100%. It has been verified this possibility is sustained for two weeks or longer (Iwao 2001).

Recently, bacteria that induce the settlement of *Acropora* juvenile corals were isolated from the coraline algae (Negri et al. 2001). Though the settlement rate is around 52%, juvenile corals can possibly be produced easily if the settlement induction method that uses incubated bacteria is established.

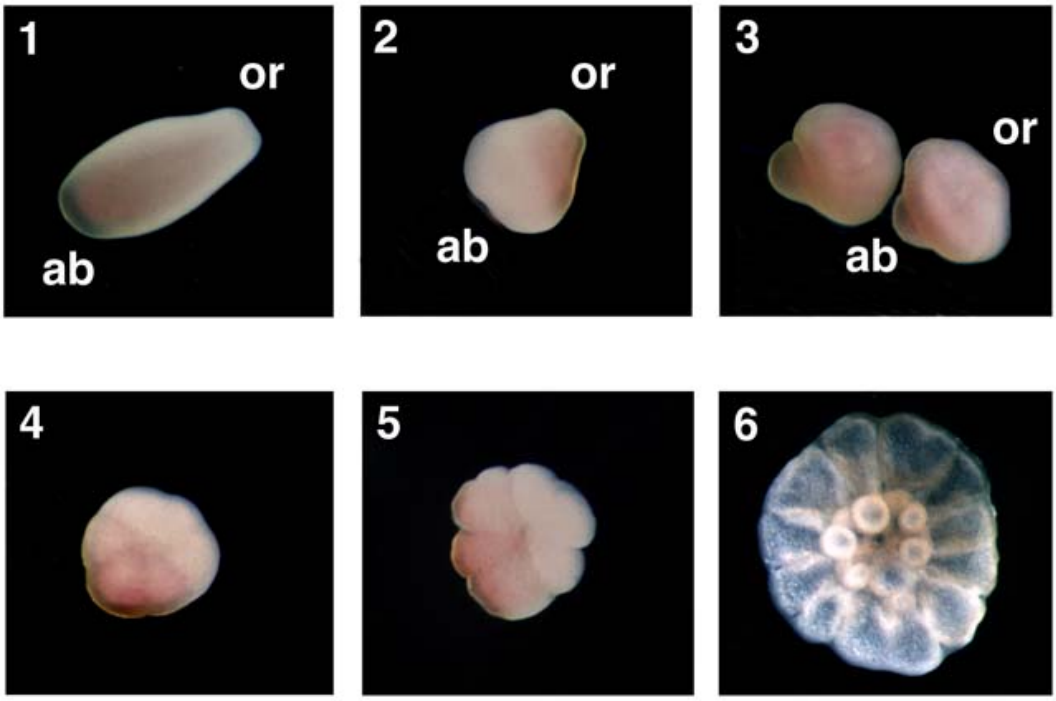


Fig. 3-8. The process of settlement metamorphosis induced with neuro-peptide

(1) and (2) The planula larvae start to contract around the body axis on the wall of a container filled with seawater, to which the neuro-peptide has been added. (3) In this stage, the adherence is weak, and thus, they sometimes come off the container wall and swim while rotating. (4) The settled larvae become flatter in the body axis direction view from the above. (5) The barrier membrane is formed. (6) Tentacles and another barrier membrane are formed so that the larvae are metamorphosed to polyps. “or” and “ab” represent an oral end and an abactinal end respectively.

(7) Substrates

As settlement substrates, materials such as concrete, unglazed tiles, shells, ceramic tiles, ceramic

works and slate plates have been utilized (various other materials are utilized by many researchers). For any material, more larvae settle when substrates are placed in the seawater beforehand, to grow some other biota on the surface than when they are settled without such pre-processing. Brush and roughly clean them immediately before using, because the marine life that grows on the substrate include large sized algae, such as brown algae, grass-like algae, and/or colonial tunicate that prevent the juvenile corals from the settling or rot during the settlement in a static water tank, resulting in adversely affects to settlement and later metamorphosis. The larvae settles in as early as one day, or in five days at the latest (Fig. 3-9).

Although it is reported that the submergence of the substrate beforehand in the sea for one year or over provide good result, coral settles well after three weeks (Taniguchi 2002).

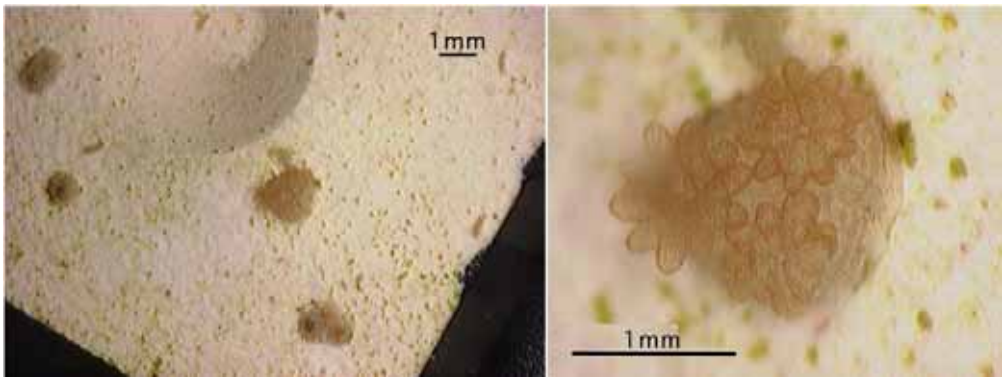


Fig. 3-9. Juvenile corals settled on the substrate, and the enlarged view (right)

Since more larvae settle inside irregular areas, the substrates with numerous declivities in structure are considered effective for the settlement. However, the most suitable form has not been clearly identified. Though many larvae also settle on the upper surface of substrates, sediments or propagation of algae can easily cause depletion, and in many cases, they can be destroyed completely.

(8) Introduction of larvae to substrate

Set up a tent (use only the lining that can pass water well) on the sea bottom, put the settlement substrate in it, and induce the larvae into it to obtain juvenile corals (polyps). With this method, the polyps can be produced on a large scale because it is not necessary to control artificially the water temperature and to be concerned about evaporation of water, both considerations when settling the larvae indoors.

Production of the polyps can be done on a larger scale. In a collaboration between Tetra Inc. and the Akajima Marine Science Laboratory, they made a nursery that had been surrounded by a vinyl cloth or by a nylon mesh enclosure (5.5 m long, 5.5 m wide, and 6.0 m high), made of a sheet in the

sea of Naha Harbor. They discharged 1.64 million individuals of juvenile corals, brought with a boat from Akajima, using divers to settle them on the blocks that had been set on the sea bottom of the nursery (Aota et al. 2003, Omori et al. 2003).

There is another method that puts the collected slick in a floating pond on the sea. It rears them; moves the floating pond to the destination; raises the water level of the floating pond by pumping seawater into it, through the water circulating opening; and leads the larvae into the tent on the sea bottom through a tube extended from the lower section, by using hydraulic pressure to induce them to the settlement substrates that are arranged on the sheet (Heyward et al. 2002) (Fig. 3-10).

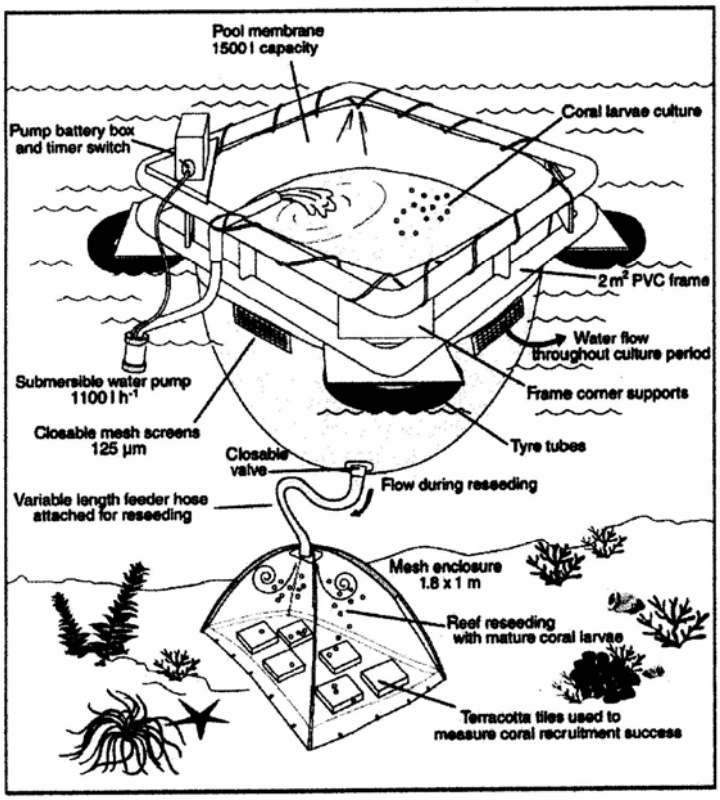


Fig. 3-10. An example of a method that induces larvae to the tent on the sea bottom from a floating pond (Heyward et al. 2002)

(9) Culture of polyps on substrate

When polyps settled on the substrate are put in a water flowing tank or on the coral reef, it has been verified that the zooxanthellae are brought into the body within one week. Since the polyps that do not symbiose with zooxanthellae die before long, it is important to start the symbiosis well before then. The newly settled polyps may be easily predated or scraped by sea-urchins and/or Gastropoda, or covered with sediment, algae, colonial tunicate and/or Bryozoa, resulting in death by blockage. Though it is desirable to breed them in an environment from which such causes of death have been

removed, no certain method of removal has been established at this time. Some researchers have raised initial polyps to large colonies (Misaki 1998, 2002), but no successful rearing of an initially large population has been reported yet. Omori (pers. comm.) attempt to culture polyps with juveniles of turban snail that remove micro-algae on substrate. When setting substrates that support polyps outdoors, they should be set so that the surface with the polyps faces downward or sideways, to avoid of them to be buried in sediments or by overgrowth of algae. To reduce the labor for setting, and to set the polyps surface sideways, it is desirable to put multiple substrates with holes onto the stainless steel rods and set them on the sea bottom. At this time, to prevent predation of polyps by fish, and the invasion of sea-urchin and/or Gastropoda place a spacer 5 to 10 mm long in between the substrates, and increase the spacer distance as the polyps grow.

Fig. 3-11 shows the settled and grown *Acropora florida* of which the planula larvae were sent from Akajima Marine Science Laboratory to the Rotterdam Aquarium, Holland.

Fig. 3-11. *Acropora florida* at the Rotterdam Aquarium. Grown from larvae sent by Akajima Marine Science Laboratory

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(Hatta M, Iwao K, Taniguchi H and Omori M)

3-2. Manufacture and placement of artificial substrates for coral settlement

This is a method that makes irregularities of approximately 1 to 3 cm on the surface of marine structures, such as breakwaters to facilitate the settlement of juvenile corals. It is expected that the turbulence of microscale flows produced by the complex shape of the substrate surface can make the larvae stay on the surface with a higher possibility of settlement when they come near. It is also estimated that the surface irregularities can reduce the danger of predation and scraping fishes and sea-urchins after the settlement. Since many of the marine structures are made of concrete, it is easy to make irregularities on the surface when manufacturing them. The irregularities can be made on the wave-dissipating blocks that are placed in front of breakwaters and foot protection blocks that are placed behind the structures (Fig. 3-12).

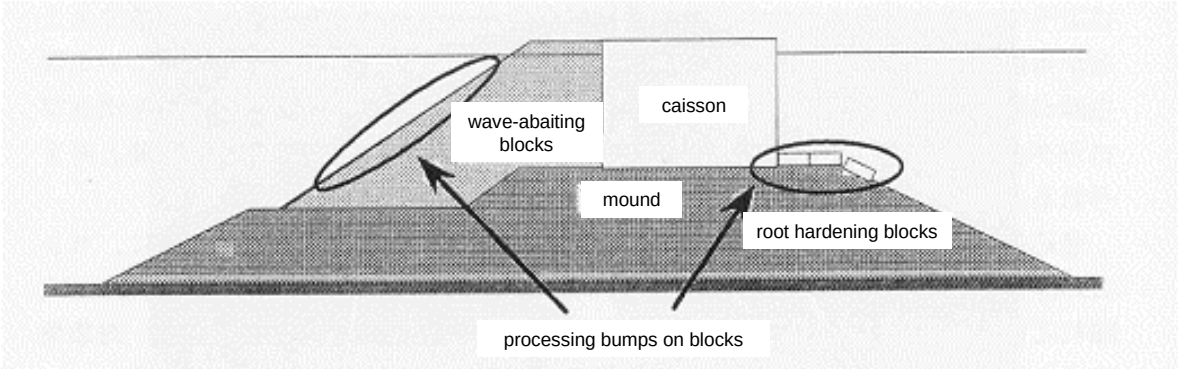

Fig. 3-12. Breakwater areas with irregularities for coral substrates
(Harbor & Marine Environment Laboratory 1999)

For making the irregularities on the blocks, the following methods have been considered (Harbor & Marine Environment Laboratory 1999):

a) Chiseled: The concrete mould of the block form is made irregular by using chiseled interior forms.
b) Drilled: Holes are made with a drill bit on the block after manufacture.
c) Streaked: Concrete is sprayed on the block surface to form the irregularities.
d) Square bar: Adhesive or bolts are used to fasten secondary products such as square bar, plates, or concrete cement on the block surface to form the irregularities.

According to research relating coral settlement success to various process to manufacture blocks with requisite irregularities (Harbor & Marine Environment Laboratory 1999), there is an indication that the use of forms which have been affixed secondary products that produce a high degree of roughness can produce more coral colonies (Fig. 3-13). As for the inclination of the substrate, there was a an indication that both the coral coverage and the number of colonies are larger on the

horizontal surface or 45 degree surface than on the vertical surface at the depth of around 2 m, but no such difference was recognized at the depth of around 10 m.

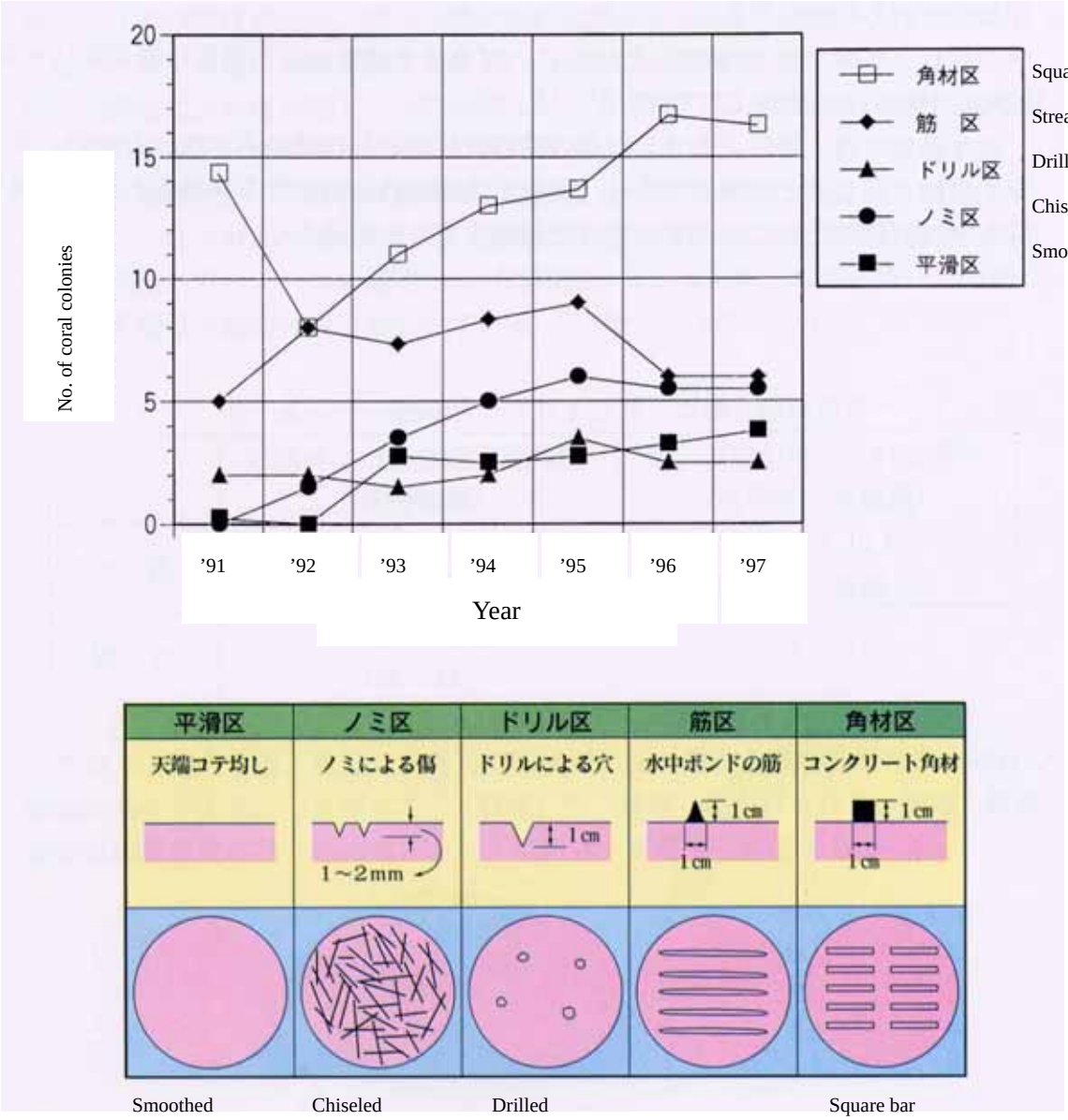

Fig. 3-13. State of settlement of corals at the processed areas of substrate
(Harbor & Marine Environment Laboratory 1999)

The only possible waters for the placing substrates are environments that the corals can readily

inhabit. Though it seems that any current coral area allows settlement corals, the settlement of larvae cannot be expected in the areas where the transparency is low and the sediments are significant. They may die out due to the luck of enough lighting or covering sediments even after the settlement. As a result of experimental settlement of *Acropora tenuis* and *Heliopora coerulea* larvae under suspended red soil or sediments, both species showed reduction of the settlement rate and secretion of mucus (Harii et al. 2002). As a red soil contamination guideline for maintaining the coral communities healthy, Omija et al. (2003) defined the annual maximum permissible value of SPSS (Content of suspendible particles in seabed sediment) as 30 kg/cm^3.

Yamamoto et al. (2002) researched the coral coverage and habitat environment on the artificial structures in Naha Harbor, and classified areas unsuitable for the growth of coral and those that allow growth. As a result, they identified suitable environmental conditions for the growth of corals, on which numerical data are shown in Table 3-1.

Table 3-1. Environmental conditions suitable for growth of coral communities in Naha Harbor (Yamamoto et al. 2002)

Environmental item	Mean value ±sd
Wave height in front of breakwater (m)	8.4±3.4
Salinity	34.7±0.1
Transparency (m)	13.7±3.5
SS(mg/l)	1.2±0.5
COD (mg/l)	0.8±0.1
TN (mg/l)	0.15±0.04
TP (mg/l)	0.012±0.005

Heeger & Sotto (2000), who studied coral transplantation, after the following set of the suitable environmental conditions for transplantation without indicating the relevant species (Table 3-2).

Table 3-2. Suitable environmental conditions for transplantation (Heeger & Sotto 2000)

Environmental item	Optimal value	Remarks
Water temperature	22℃-30℃ (25℃ is desirable)	
Salinity	32 to 36	Free from effects of rivers
Transparency	12m or more	
Tidal current	1m/s or less	
Depth	6 to 12m	Shallower areas are easily affected by drift sand.
Sediment	Sand or algae community dotted with coral communities	

Since coral eggs and larvae drift for about five days near the surface, they are affected much by the wind, which varies according to the weather conditions at the time. Therefore, the situation of coral recruitment for resettlement at a given place generally varies from year to year (Marine Parks Center of Japan 1995). But, macroscopically, there may exist some places with a repeatedly high possibility of retention of larvae, caused by middle scale turbulence and counterflow produced by the bottom topography and constant flow of current. The frequency of the recruitment may be high in such places.

The depth for this purpose can be determined by noting to the depth of water peripheral to areas that corals inhabit. The coral inhabitable depth varied depending on species, but in many cases can be from the low tide level down to 30m as far as the transparency is good (Fig. 3-14).

Manufactured structures must be placed in the sea before the coral spawning, as a matter of course, which varies according to place and from year to year. In Okinawa's coral reef areas, the spawning begins in May, in the years that water temperature is higher range, and usually in July or August in Honshu and Kyushu areas. The time of placement of structures should be determined by taking into consideration the period needed to adapt the concrete for the seawater, and in addition, strategies to prevent algae attaching to the substrate and disturbing the coral settlement, or even can covering the substrate. However, it may be acceptable that the substrates are covered with some crustose red algae because some of them encourage recruitment of coral larvae. As a result of experimental settlement of *Acropora hyacinthus* and *A. tenuis* larvae on artificial substrates (unglazed tiles) that were submerged in the sea for three weeks, six months and one year respectively, it is reported that the submergence longer the more settled larvae. Larvae settled on the coralline algae that covered the substrates, which itself had varied lengths of time to become established (Taniguchi 2002). However, if the algae other than the coralline algae and/or sea squirts, sea sponge, Bryozoa or other competing sessile animals cover the substrates, they must be removed.

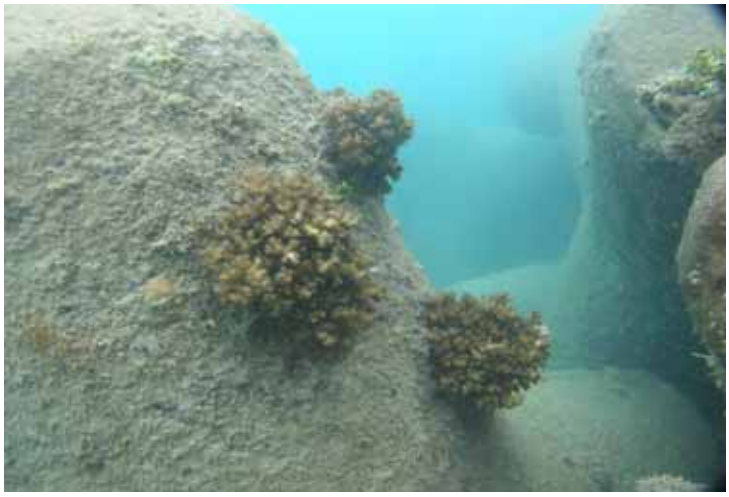

Fig. 3-14. *Pocillopora damicornis* on breakwater wave-dissipating blocks (Ishigaki Harbor)

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(Fujiwara S)

4. Restoration technology by using asexual reproduction

4-1. Transplantation of coral fragments

It is desirable that the transplantation projects are developed by taking full advantage of the experiences that have been obtained and enhancing more knowledge through experiments based on the well planned methods and long time observations in the transplantation sites, time, locations, fixing method, substrates, and other factors.

(1) Sampling of fragments from donor coral

The most important matter here is that not to many fragments should be collected from a single donor colony at a time, to minimize the effect of collection on the colony. There is little physiological knowledge about the effects of the collection of the fragments on the reproduction of donor colony (Szmant-Foelich 1985, Smith and Hughes 1999, Koji and Quinn 1985, Zakai et al 2000).

When collecting the fragments from a donor colony for transplantation, the colony and the fragments are not damaged severely if a wire cutter or underwater scissors are used for branching corals or a hammer and a chisel are used for tabular, corymbose, massive or other forms of corals, to break them at a blow. It may be possible to use the fragments broken by typhoon or other natural causes for the transplantation. Bruckner and Bruckner (2001) conducted transplantation of fragments generated by ship groundings. The result is that surviving fragments (57%) were larger than dead fragments (26%).

(2) Size of suitable fragments

Generally, it is expected that the larger the transplantation fragments are, the higher the survival rate is. However, because taking larger fragments can damage the donor colonies, the most important point in the future experimental transplantation will be to define the minimum acceptable size which allows a survival rate of 100%. To achieve this, the environmental conditions, such as the time for transplantation, which are discussed later in this document, should be taken into consideration (case examples described in Chapter 6-2).

So far, fragments of 2 to 30 cm long or in diameter, or colonies of these sizes (entire donor colonies) have been transplanted. As a result of transplantation of fragments of *Montastrea faveolata*, which forms settling type massive colonies, with diameters ranging from 2.5 to 5.1 cm, the survival rate at 9th month after the transplantation was 75%, and thus, Becker and Muller (1999) estimate that even the colonies of diameters less than 2.5 cm can be transplanted.

However, for the transplantation of *Acropora echinata* and *Pavona cactus* colonies consisting of three size groups, 5 to 10 cm, 10 to 15 cm, and 15 to 25 cm, and the fragments consisting of two size

groups, 2 to 4 cm, and 3 to 6 cm, no difference of mortality rate were seen among the three size groups of the colonies, but the mortality rate of many of the fragments groups were higher than that of the colony groups (Plucer-Rosario and Randall 1987). As a result of the experiment that compared three size groups of fragments *Acropora prolifera*, 3 to 5 cm, 8 to 12 cm, and 15 to 22 cm, smaller fragments clearly showed higher mortality rate (Bowden-Kerby 1997). According to these results, it can be said that the survival rate can be higher when the coral is transplanted in colonies than when transplanted in fragments, and the rate can be higher when the coral is transplanted in large fragment size.

(3) Transportation

Regarding the method of transportation, not much difference can be seen among the researchers (Okubo and Omori 2001). The principal methods include the use of divers, who carry fragments in containers without taking them out of the water if the destination is near (Dodge et al. 1999), or, if the destination is far, a boat is used to carry the fragments in wire mesh bag or net bags hung in the water from the boat (Dodge et al. 1999; Munoz-Chagin 1997). The fragments may be carried in water-filled buckets on deck (Bowden-Kerby 1997), however, when using this method, it is necessary to be careful that the water in the bucket is not warmed up during transportation in a hot season.

Basically, it is desirable to transport the fragments without taking them from the sea. The possibility that the transplantation fragments can survive transport in the air varies among the coral species. Fragments of *Acropora gemmifera* and *Favia stelligera* can be transported out of the water if the period is up to approximately 2 hours, but *Stylophora pistillata* or *Rumphella* sp. must be transported while submerged in water (Kaly 1995). Urabe et al. (2003) reported that the tolerance of coral communities to drying is higher for *Fungia* or massive corals and lower for branching corals, and as a general tendency, watering is effective, but mere light shielding is not effective.

(4) Fixation methods and choice of fragments

Some methods of fastening coral fragments that have been used are listed below (Fig. 4-1).

1) In many cases, epoxy waterproof cement is used for fastening fragments on the substrate. First, remove algae and other foreign matters from the substrate by using a wire brush or other means. Place the fragments on the substrate vertically or horizontally and adhere them with epoxy cement. In some past cases, regular industrial cement was used instead of the waterproof cement. Since the transplanted corals can easily come off the substrate when only this method is used, some supplemental methods are used additionally (Fig. 4-1-a).
2) Put a small plant pot in the hole made on the coral rocks used as the substrate, insert a coral fragment into each pot, and fix them by using cement that has already been mixed with

freshwater on land beforehand (Auberson 1982)(Fig. 4-1-b).

3) Put the cement with retardant in a small polyethylene bag, and then put the bag on a concrete mat. Then, insert the fragments or colonies into the cement in the bag and fix them. For the fragment of 10 cm or longer, put concrete nails in the bag and fasten the fragment to the nails (Clark and Edwards 1995) (Fig. 4-1-c).
4) Put the cement with retardant in a nylon bag, insert the fragments into the bag, attach a hook for securing the bag to the substrate, and then, keep the bag in the water tank until the cement hardens. When the cement has hardened, fasten the bag to the base rock by using the hook and a rope (Clark 1997) (Fig. 4-1-d).
5) Put nails in the substrate for transplantation, and fasten the coral fragments to the substrate with wires or cable ties (Iliff et al. 1999; Okubo and Omori 2000; Okubo et al. 2001, 2002) (Fig. 4-1-e).
6) Skewering: Drill a hole at the center of each coral fragment, and run a bamboo skewer through the hole. Make holes on the substrate for transplantation by using an underwater drilling machine, and put the bamboo skewers with the fragments in the holes (Nishihira 1994) (Fig. 4-1-f).

When fragments are attached to the nails that have been put on the substrate and fixed with cable ties (Fig. 4-1-e), the fragments of some species that do not settle on the substrate easily because of their mode of growth can increase the risk of the nails dislodging from the substrate as the years pass after the transplantation. Since the nails do not come off after the cable ties are cut, the loss of the fragments can be prevented more surely by fastening the nails to the substrate with epoxy cement. Since the chemical effects of epoxy cement on transplanted fragments have not been assessed, segments should be transplanted after the cement has hardened. Before applying the waterproof cement, it is necessary to remove the algae and other foreign matter from the substrate by abrasion, using a wire brush. A vertical orientation of fragments, fixed to the substrate is recommended to minimize the deposit of sediment (Okubo and Omori 2000; Okubo et al. 2001, and Okubo et al. 2002).

(5) Choice of place

Since the optimal growing conditions of corals vary among the species, it is necessary to investigate the location that the donor colony inhabits, which is the source of the fragments, and the location to which the fragments are transplanted, with attention to their respective physical condition (wave, tidal current, turbidity, depth, light intensity, amount of sediments, salinity etc.) prior to the transplantation. The survival rate of transplantation is higher when the environmental characteristics of the two locations are similar, and it is lower when the fragments are transplanted to an unfamiliar

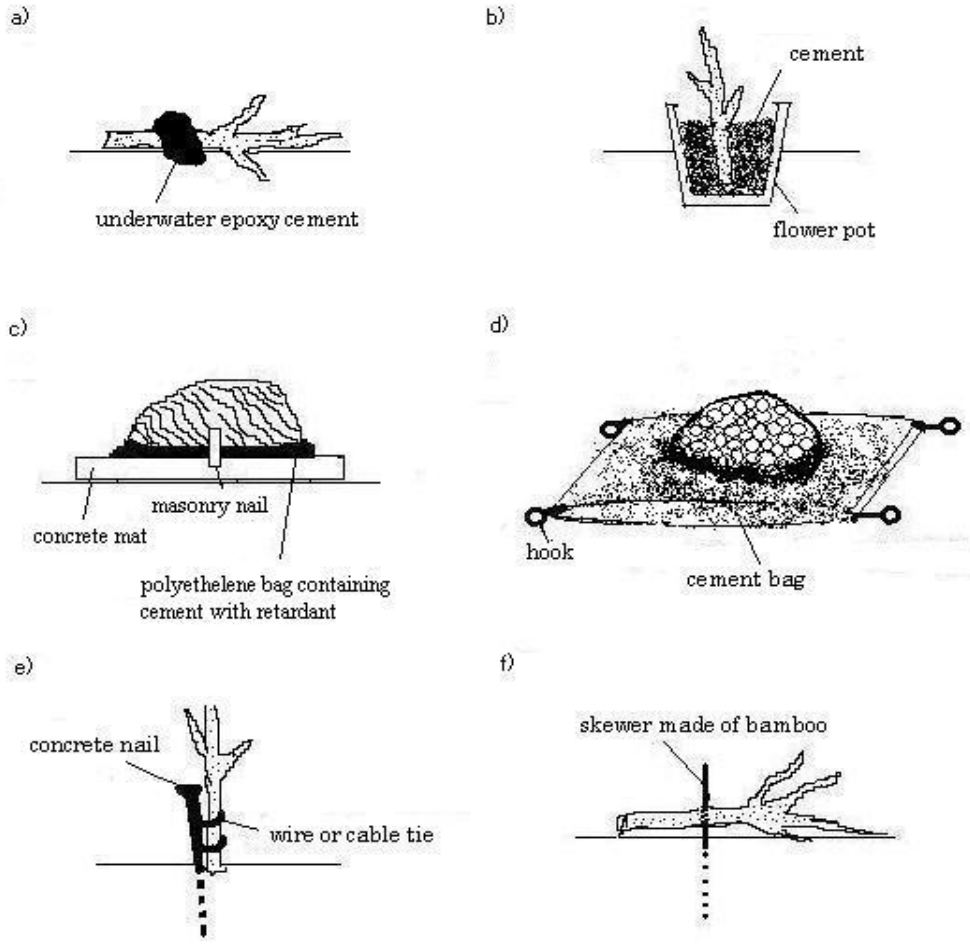

Fig. 4-1 Method of fastening coral fragments

environment (Auberson 1982; Marine Parks Center of Japan 1993, 1994, 1995).

It is also necessary to investigate the location to which the fragments are transplanted in order to discover whether marine life such as crown-of-thorns starfish or coral-eating gastropods that predate the corals are present. For the marine park zone in Ashizuri Uwakai National Park, Kochi Prefecture, Japan, the survival rate at the first year after the transplantation was 0% in the past three attempts because of mass populations of coral-eating gastropods in the location (Ashizuri Uwakai National Park, personal communication).

One experiment placed fragments collected from three forms of *Acropora*, ie. *Acropora intermedia* (branching), *A. millepora* (corymbose form) and *A. hyacinthus* (tabular), on the reef flat, reef ridge and reef slope, without fixing them (Smith and Hughes 1999). Their survival rates at the 17th month later were 37% on the reef flat, 15% on the reef ridge, and 10% on the reef slope. Rates of settlement on the substrate were 39% on the reef flat, 31% on the reef ridge, and 4% on the reef slope. The exposition of these results noted the following matters: the growth rate of the fragments placed on

the reef flat is higher than that on the reef ridge, because the chance that the fragments placed on the reef flat are shielded from the sunshine by the surrounding tabular corals is less; the fragments can easily settle on the reef flat, which consists of the coral rocks with the hard sediment; and thus, the number of fragments that are killed by the covering sediments is less.

(6) Preferable substrate

When it is necessary to transplant coral fragments on the underwater structures, it is very useful to understand what kind of substrate can permit make the corals to settle easily. There was an experiment for comparing the suitability for settlement among five types of substrates, including ferrite-containing concrete, unglazed tiles, concrete blocks and iron that are frequently used for underwater structures, and natural coral rocks (Okubo 2003). As a result of this experiment, the fragments that showed the higher settlement rates were those transplanted on the concrete and ferrite-containing concrete. Ikeda and Iwao (2001) transplanted 10cm fragments of *Acropora*. *formosa* on the substrate for transplantation that was made of concrete mixed with coal ash, which is an industrial by-product, using the method just described. As a result, the settlement rate was nearly equal to the fragments that were transplanted on regular concrete. Based on both of these results, it is estimated that the coral fragments can settle on concrete substrates easier than on other materials, for as yet reasons unknown.

(7) Preferable season

Most transplantations have been performed in warm periods when the monthly average air temperature is in the range from 24 to 28 ℃. However, since the locations, coral species, their fixing methods and other factors affecting past transplantations are different from each other, it is impossible to compare them, in order to fix the most suitable time for the transplantation. Thus, the present author have performed experiments using the same materials and methods, and only changing the transplantation time, which are discussed in Chapter 6-2.

Four experiments were carried out in the subtropical region where the range of water temperature is relatively large (the monthly average air temperature is in the range from 26.6 to 28.3 ℃) in a warm season, using the fragments of nearly the same size and the similar methods. Though the species were different, the average survival rate at the 3rd month after the transplantation was 98.5% in a total of 13 species including *Dichocoenia stoksii*, *Montastrea cavernosa* and *Porites astreoides*, etc. (Dodge et al. 1999), and the survival rate at 43rd month after the transplantation of *Acropora formosa* was 69% (Okinawa General Bureau, Okinawa Development Agency 1997). For *Acropora echinata*, the rate at an unknown time after the transplantation was 46% (Plucer-Rosario and Randall 1987). Thus, the results vary much among the species.

As a result of investigations of the relationship between the mortality and temperature an positive

correlation was shown (Yap and Gomez 1984; Yap et al. 1992). The mortality is raised in a high water temperature period by the stress from the transplantation and bleaching.

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(Okubo N)

4-2. Transplantation of juvenile corals

During the investigation of corals in the Sekisei Lagoon, the present authors found that the density of juvenile corals (at the first and second year after the settlement) per unit area varied significantly among the locations. Although this is a phenomenon that has been known generally, it can be a problem when a number of larvae of, for example, *Acropora* that becomes tabular or branching corals settle on the shallow area of a reef washed by rough waves. These juvenile corals are easily broken by the waves as they grow, and thus, they can rarely grow to maturity. So, we decided to collect such juvenile corals to transplant them to an area suitable for growth and development.

(1) Sampling

The larvae of corals settle in cracks or depressions found on coral reefs. Many of them also settle on the surface of corals that have just died and not yet covered by other marine life. It is assumed that larvae settle largely in such patterned and indented surfaces because they tend to survive better after settlement. The larvae start to grow outward from the declivities approximately one year from settlement, and become around 1 cm in size, which is clearly visible. Many of these juvenile corals can be seen in shallow waters on the reefs that are washed by rough waves, and the density frequently reaches several tens of individuals per one square meter or more. Individuals needed for transplantation can be obtained by collecting these juvenile corals together with the substrate.

Then, we attempted to collect juvenile corals for the transplantation by using a dry land pneumatic drill. They can be taken relatively easily by using a core sampling bit, with sufficiently big bore for the size of the colony (juvenile coral). This method takes not only the juvenile corals, but also the substrate coral reef as a short core 3 to 4 cm long, which is easy to transport (Figs. 4-2 and 4-3).

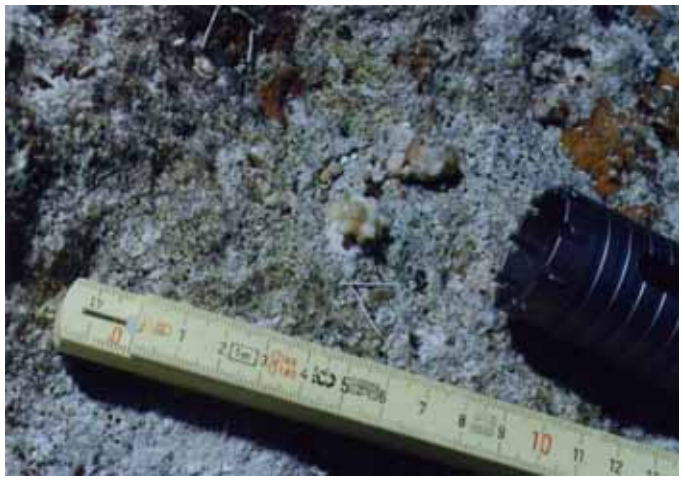


Fig. 4-2. Natural juvenile corals and core sampling bit

Fig. 4-3. Core sample of juvenile corals

Fig. 4-4. Transplanted core sample of juvenile corals (fixed with adhesive)

(2) Method of transplantation

When juvenile corals collected with substrate are kept in the sea for a long time during intermediate rearing before transplantation, it is necessary to cover the substrate with vinyl tape to prevent sessile organisms from attaching to the surface of the core base. At the destination where the coral is to be transplanted, use the core sampling (the same size as the one used for removing the core) to make a hole for fixing. Put a little underwater adhesive in the hole, and then insert the juvenile coral, applying additional adhesive to complete the fixing. In the case of an environment in which silt be easily deposited, it is desirable to make the hole shallower than the height of the core so that the juvenile corals protrude from the bottom a little (Fig. 4-4).

(3) Evaluation of the method

Conventional transplantation methods over the past few decades have used a part or entire colony, and the biggest concern has been fixing the coral securely to the substrate. The primary difficulty has

been that corals have various forms, and thus no single fastening method has been adequate for all of them. In addition, it is well known that survival after transplantation is affected by variable that can hardly be controlled, such as the risk of human error in the fixing work and the handling of corals during the work.

The core sampling drill technique described above appears to deal with these risks and variables most satisfactorily. During transit in the sea or on a boat, the juvenile corals are not normaly damaged even if a large number of cores are put vertically in container. Also, the core sampling technique allows the use of a simple and assured mechanism that drills a hole in the substrate for the removal and transplantation. It fastens the core using adhesive. Therefore, the removal and neimplantation phases can be better standardized and the risks during travel reduced.

As discussed above, it is technically possible to collect the naturally settled and grown juvenile corals to utilize them for the transplantation. Corals can be severely damaged by the bleaching events and the predation of the Crown-of-Thorns starfish, so sometimes it is advisable to perform mass collection of the juvenile corals, which is a key to regenerating corals. But this should be done under substantial regulation. It is a fact that the places where the juvenile corals settle naturally are not always suitable for growth and development. If they are to be moved, then the decision must be made by experts of this field.

In conclusion, an attempt to collect juvenile corals to utilize them for the transplantation can be implemented under the lead of the experts if it is performed on a small scale (several thousand juvenile corals). Several tens of thousand or more may have to be collected, for large scale restoration project, but it is harder to find sufficient numbers in Okiawa for movement to new sites.

(Okamoto M & Nojima S)

5. Transplantation of whole coral colonies and coral reefs

5-1. Transplantation of whole coral colonies

Depending on species and size, there are several methods to recover and transplant coral communities, if coral communities are threatened by a development work on the shore (and there is no other choice but to save them than transplantation). Chapter 6-3 introduces the example case of *Porites lutea* colonies. Massive *Porites* exceed 1m in diameter, which is unsuitable for the usual transplantation method that uses fragments of colonies. Therefore, whole colonies have to be transplanted together. The example is a case of colonies which range from 20 to 30 cm in diameter, which are relatively easy to handle.

As the first step of *Porites* transplantation, the colony base is separated from the bottom using a chisel, taking care not to damage to the corals, and the base is put into a container under water, transported to the destination, and then adhered to the substrate using waterproof cement. As the settlement substrate, two types, an artificial substrate made of concrete plate and a natural base rock were used. For the case of the natural base rock, a method that drives anchor bolts into the colony base to fix them was used, which was demonstrated to be effective.

As a result of a follow-up survey four years following the transplantation, both the survival and growth of the corals on the natural base rock were better than on the artificial substrate. For the massive *Porites*, which is usually distributed in reef moats, it is recommended that they be transplanted to moats. However, moats generally provide no suitable substrates for settlement, because they frequently have sandy or gravel bottom areas. Nevertheless, if there is no other choice than transplanting on the sandy or gravel bottom, use the iron stakes driven in the bottom by a pneumatic hammer as the settlement surface, after finely adjusting their positions. For this example case of transplantation, colonies in the range from 20 to 30 cm in diameter were moved. However, it is still difficult to transplant colonies larger than this size if only human power is used. The relocation method discussed in the next section should be used for massive removals.

5-2. Replant of coral reefs

Fukunishi et al. (1998) and Waterfront Vitalization & Environment Research Center (1999) introduced techniques for relocation of coral communities, as one of the methods for planning a harbor construction project that harmonizes with the coral reef. They describe this technique that moves the coral communities together with the base rock if there is a threat that the coral reef will be eliminated by reclamation or construction of a breakwater.

Utilizing this technique, a new experimental project has been implemented continuously since 1998 in Hirara Harbor, Miyako Island, Okinawa, for carrying forward the harbor construction

project harmonized with coral community. Project achievements were partly reported by Ishii et al. (2000, 2001) and introduced by the booklet issued by the Hirara Harbor Construction Office, Okinawa General Bureau, Okinawa Development Agency (2002).

The process of the coral community relocation is shown in Fig. 5-1, and the process and experimental results of the coral community relocation in Hirara Harbor are outlined below.

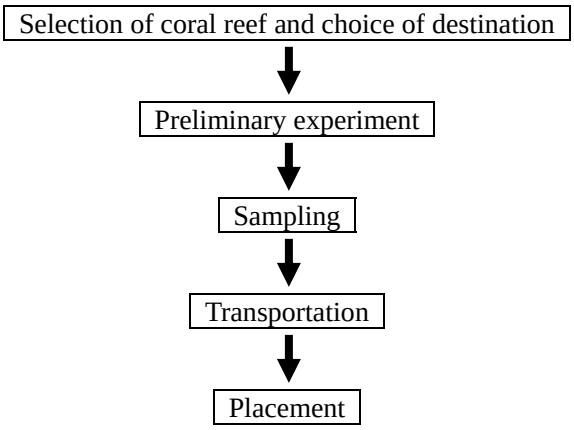


Fig. 5-1 Process of coral community relocation
(Waterfront Vitalization & Environment Research Center 1999)

(1) Selection of coral reef and choice of destination

Prior to the beginning of the coral community relocation project, examine the areas that are affected by it directly and indirectly, survey the actual state of the coral communities that exist in the areas, and select the coral communities that require the relocation.

Since it is desirable that the destination of the coral community relocation is environmentally similar to the source, confirm beforehand the selection of the suitable site by performing a survey of the destination, selecting a concrete candidate site and then performing a preliminary experiment.

(2) Sampling

It is necessary to break the base rock to collect the base rock fragments on which coral communities settle. When breaking the base rock, it is necessary to examine the fracture method and the size of base rocks to be relocated so as to minimize the damage to the coral communities. When relocating the coral communities, because there were no previous cases of relocation, an underwater backhoe was used in FY1998 and 1999, and water jet, air lift and other means in FY2000 to break out the settlement substrates (Fig. 5-2).

Fig. 5-2. Collection of base rock fragments on which the coral communities are settled (Hirara Harbor Construction Office 2002)

(3) Transportation

The transport of the base rocks in Hirara Harbor was performed by using a method that prevents the corals from contacting the air, which stress them, as shown in Fig. 5-3. This method is effective for the relocation to places near the source, but the safety and efficiency of the transport lowers the farther is the destination from the source. So, transportation aboard a boat should be considered because some components of the species of the coral communities can be tolerant to a certain level of drying. Doing that requires measures for minimizing damage to the corals, such as spraying sea water on the coral communities continuously and/or covering with a light shielding sheet. It is desirable to confirm the effect of the measures by performing the drying tolerance experiment beforehand.

Fig. 5-3. Transport of base rock collected (Hirara Harbor Construction Office 2002)

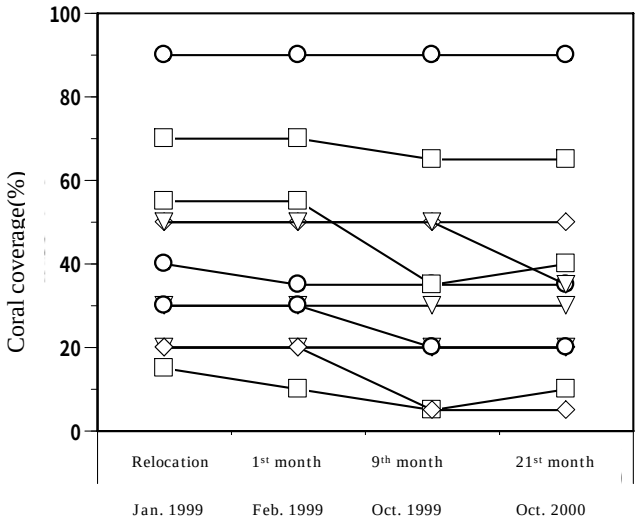
(4) Placement

When setting the base rock, it is desirable to make it stable by taking into consideration the size, wave conditions of the location, and other factors. At Hirara Harbor, an underwater backhoe was used in FY1998 and 1999, and a water jet was used in FY2000 to dig out the settlement substrate. To

avoid giving stress to the corals, the use of a pneumatic lift allowed the transportation of big coral communities, keeping them below the water surface to relocate them to the periphery of the mound in the harbor.

(5) Conclusion

The results of the experimental but massive coral community relocation in Hirara Harbor are as described below. Figure 5-4 shows the temporal change of coverage of the coral communities that were relocated to 12 points in January, 1999 (Ishii et al. 2001). The relocated coral communities mainly included Poritidae, Siderastreidae and Faviidae. For the coral communities relocated in January, 1999, the mass of rock was small, and some of them were washed away by the ocean waves from a typhoon. However, most of the coral communities on the new stable substrate have survived. Also, no affects of the settlement due to the environmental conditions of the relocation destination could be seen, and we observed that the most of them survived the stress of relocation and environmental change before and after the relocation. At the survey in October, 2000 (21st month after the relocation), some points showed new growth of the coral.



	Inside of the harbor	Outside of the harbor
Upper layer	□	○
Lower layer	◇	▽

Fig. 5-4. Change of coverage of corals relocated at Hirara harbor in FY1998 (Ishii et al. 2001)

In 1998, a coral bleaching event occurred worldwide due to high water temperature, and the coral communities relocated to Hirara Harbor were also affected by the bleaching event. However, as shown in Fig. 5-5, they were confirmed to have been recovered at the investigation in October, 1999 (9th month after the bleaching event), and no effect of the bleaching event was recognized in October, 2000. These results may show that the stress the coral communities received from the relocation was small.

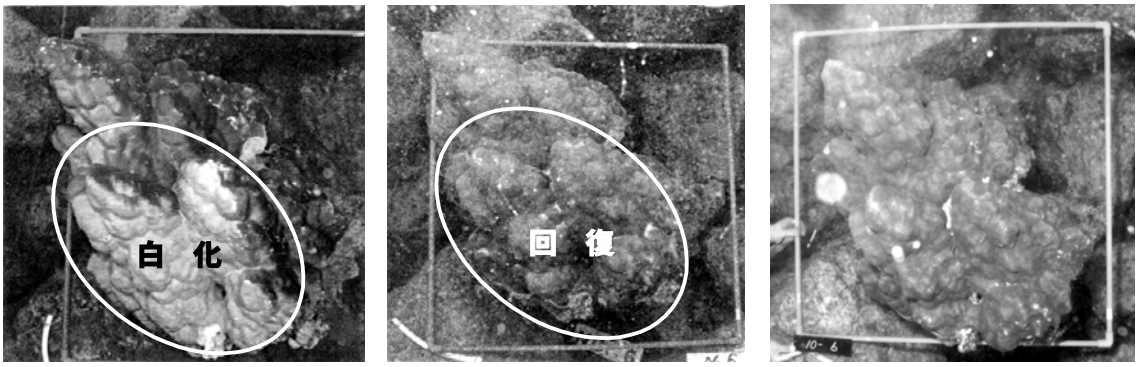


Fig. 5-5. State of recovery from the 1998 bleaching event (Ishii et al. 2001)
Left: Immediately after relocation (Jan. 1999), Middle: 9th month after relocation (Oct. 1999), Right: 1 year and 9th month after relocation (Oct. 2000)

Table 5-1 shows the state of appearance of reef animals larger than corals observed on the mass of rock supporting the coral communities immediately after the relocation in November 2000. The number of species of large size benthic animals ranges from 0 to 11, and the number of individuals ranges from 0 to 25. The sponges, colonial sea-squirts and other species of which individuals cannot be counted also settled. These large size benthic animals were the same types that had lived on the rock before the relocation and had been carried together with the base rock by the relocation. It is worth noting that the coral community relocation technique allows conservation not only of the coral, but also other animals and plants that adhere to them.

The principal advantages of the coral community relocation technique as compared with other coral transplantation techniques, are that it allows conservation of whole coral communities relocated and that handling of massive corals such as *Porites* is feasible. A patent is being applied for this technique.

Table 5-1. State of appearance of large animals observed on the mass of rock, including the coral communities (Ishii et al. 2001)

	Mean	Standard deviation	Minimum	Maximum
No. of species /m^2	5.0	2.7	0	11
No. of individuals/m^2	7.3	8.5	0	25

In Hirara Harbor, the construction of a “environmental breakwater” that uses components such as “water pass caissons”, “wave-abating blocks with slots” and “root hardening blocks with various

structures” is underway in addition to the “coral community relocation” (Fig. 5-6).

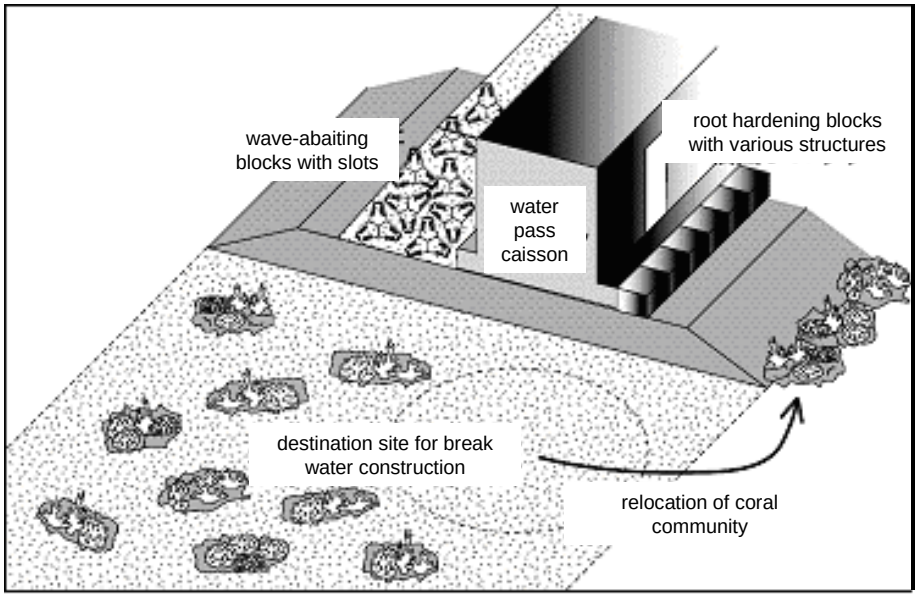


Fig. 5-6. Schematic view of environmental breakwater at Hirara Harbor (Ishii et al. 2001)

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(Fujiwara S & Omori M)

6. Development of underwater techniques for coral reef restoration

6-1. Experiment 1. Restoration using sexual reproduction

(1) Introduction

In the period from FY1998 to FY2000, Japan Marine Science and Technology Center developed various investigations and studies for the whole area of Sekisei Lagoon (the waters between Ishigaki Island and Iriomote Island, Okinawa) in its research project (Okamoto 1998). Unfortunately, a worldwide bleaching event occurred in 1998, when the project was initiated, which cataclysmically changed the state of Sekisei Lagoon's corals (Okamoto et al. 2000; Yamazato 1999). Moreover, in 2001, Sekisei Lagoon's corals were damaged by a bleaching event again, causing growing concerns about the survival of the corals.

The present authors commenced basic studies in Sekisei Lagoon from FY2001 as an effort to regenerate the coral reefs. This is an activity derived from our recognition that "Sekisei Lagoon does require the actions that strengthen the regeneration capability of the corals, and the lagoon is water having such potentiality", based on: the ecological knowledge of the corals that were obtained through conventional investigations and researches; and the fact that the corals of the wide Sekisei Lagoon are on the road to death from the repeated bleaching events. Underwater working techniques mainly include a diving.

The attempted coral reef regeneration that is discussed in this paper was planned and implemented as one that can deal with "the fact that the corals are dying out due to the effect of the warming of the earth", based on the conventional coral transplantation method. We would like to introduce the new technique as one underway for the coral reef restoration, asking the reader to understand that final results cannot be shown because the activity is still in process.

We have performed the experiment by using settlement tiles (10 × 10 × 1.3 cm, with a hole in the center for securing) made of Amakusa porcelain. This tile substrate tests the settlement potential of the juvenile corals that were generated at the simultaneous spawning. The settlement tiles were placed on architectural blocks. On each block, three pieces of the tiles were arranged at even intervals in two layers, 6 pieces in total. The setting of the settlement tiles was performed from about one week before the date of simultaneous spawning to the spawning date. This way of setting the tiles prevents the surface from being covered by competitors of the coral larvae at the time of the settlement or by substances that can disturb the settlement.

Investigating the settlement potential of the larvae on the tiles, we collected the settlement boards at the 3rd month after the simultaneous spawning, brought them back to laboratory after drying them, and counted the settled corals. For some of the tiles, we investigated the depletion rate of the settled corals by collecting them at the first year after the spawning and counting them.

The corals that settled on the settlement tiles concentrated near the outer edge of the lower side of

the tiles when they were collected at the 3rd month after the settlement. When the tiles are left in the sea as they are, the corals grow outward, and then gradually appear on the upper side. The corals, once appearing on the upper side, grow faster to a size that limits injury to them from sea urchins and fish (Fig. 6-1). The juvenile corals that have grown on the settlement tiles can be utilized for the transplantation as they are.

Fig. 6-1. Juvenile corals that appear on the side of the settlement tile after they settled on the other side

(2) Conception of methodology

The method utilizes an indefinitely large number of coral larvae generated by simultaneous spawning to breed the juvenile corals for transplantation. It requires a longer period of preparation compared with the conventional fragment transplantation. Simultaneous spawning is a fundamental natural strategy for the survival of corals. The largest advantage of this approach is that it does not disadvantage any of transported species, and thus, juvenile coral communities consisting of various species present in the spawning areas can be obtained. That is, we take advantage of population patterns to aid the coral reef regeneration while maintaining the coral community, not the mere regeneration of the singular species of corals.

The fundamental idea of this regeneration includes the steps described below.

a) Putting the larvae settlement apparatus on the suitable larval settlement site immediately before the simultaneous spawning of the corals, and letting the larvae generated by simultaneous spawning settle on their natural site;
b) Moving the settlement apparatus to the rearing water just after larvae have settled, because conditions at the settlement site are not as conductive to growth always the best destination site;
c) Removing predators and cleaning the apparatus occasionally to raise the survival rate until the

larvae grow to juvenile corals;

d) Utilizing the grown juvenile corals for the transplantation because they have survived through the stage of mass depletion.

(3) Problem of use of substrate

The conventional tiles that have been used for the study of larvae settlement potential offered the following problems:

a) The tiles were larger than necessary.

Though many corals settled on the under side of the tile, the settlement is limited to the area near the outer edge where they could easily spread to the top side as they grow. After the transplantation, only one coral individual may be able to grow per one piece of tile as a result of their competion for survival.

b) The work of setting out many boards was hard.

Since the coral larvae attached to the under side of the tile, they had to be arranged at even intervals horizontally in layers. One tile that we employed weighs approximately 300g in the air and, thus, arranging many tiles required a specially made frame. Also, when taking into consideration the packing and transportation in the sea, it was desirable to keep the total weight including the settlement tiles and frame at around several tens of kilograms. Thus, only about 50 tiles could be arranged on one frame.

c) The work of fixing the tiles at the destination place was complex.

The juvenile corals which have reached the transplantable stage often had grown on both sides of the tile. To prevent the corals from being damaged when fixing tile on the bottom, therefore it was necessary to use a variety of fastening methods and materials such as nails with a spacer put below the tile.

(4) Development of new artificial substrate

When using the slabs (tiles) for the settlement, rearing and transplantation of the juvenile corals, it was necessary to improve this artificial substrate surfaces to solve the fundamental problems described above. In the initial stage of the examination, we attempted to use slabs and stone blocks as the settlement surfaces after processing them into various forms. As we progressed, we performed a series of experiments with regard to the underwater fastening methods, the settlement state of the corals, the transplantation methods etc.

With regard to the materials for settlement surfaces, we examined materials such as Amakusa porcelain clay and Ryukyu limestone, together with materials that naturally grown juvenile corals are attached. As a result, we determined that factors such as the roughness of the surface of the material and minute pores are crucial to make the settlement successful.

Then, we determined that it is necessary to abandon the use of flat slabs for corals settlement, and, then, to design and develop settlement devices with new design characteristics, taking into consideration all requirements ranging from the settlement of corals to the transplantation works. For the forms of the settlement devices, we took account of their relative easiness of use during underwater works and ecological affects based on the mechanisms of settlement, growth and survival of the corals.

With regard to the easiness of use in the sea, the following three points were noted.

a) Suitable for the transplantation work (especially, the method of fastening on the substrate)

b) Easiness of handling many settlement devices in the sea (small and light)

c) Lower manufacturing cost (suited to mass-production)

From the ecological viewpoint, the following three points were considered important.

a) Allowing easy settlement of coral larvae

b) Protecting corals from predation at their initial stage of settlement

c) No disturbing the growth of juvenile corals

When manufacturing the settlement devices that meet the above requirements, we noted that the use of existing materials after processing them is technically difficult, and may cost significantly even if they can be used. Then, we considered the utilization of ceramics, and came to conclusion that the settlement devices could be mass-produced by utilizing the ceramic work techniques found in Seto, Aichi Prefecture, Japan. Settlement device No.1 (ONF-1) was manufactured in April, 2002, by molding the porcelain clay with pressure casting, utilizing a plaster former and then baking in oxidizing fire at 1,250 ℃. Since the ceramic devices have sufficient strength and minute pores allow water into approximately 5% of the volume, we expected that this feature would be advantageous for being saturated with seawater in a short period after they are set in the sea. As the raw material, regular porcelain clay was used here, however, the use of this baking method also allows manufacture of settlement devices with similar characteristics using the coal ash that is produced by thermal power plants (Fig. 6-2).

The settlement devices are given a form that allows easy implantation of juvenile corals and are made small and light to make the transplantation simpler. By laying the settlement devices inside each other and arranging them on the frame, many settlement devices can be easily transported and then individually set in the sea. Also we made a consideration to prevent the invasion of the predator after the settlement of the larvae by adjusting the spaces between stacks of units.

(5) Design and construction of artificial substrate

The settlement devices are given a conical form similar to a toy wooden top. The unit consists of a

Fig. 6-2. Coral settlement devices

settlement section, spacer and connecting-inserting section. The settlement section is given a wide settlement area on the bottom side and provided with stacking holes on the top side, based on the form of Amakusa porcelain clay plates that have been used previously. Multiple grooves are arranged on the bottom side as a feature for increasing the settlement area. The spacer section is given a role of keeping a certain gap between the settlement sections when stacking the settlement devices on each other after implantation. The connecting-inserting section functions as a guide that makes it easier to lay many settlement devices on each other (Figs. 6-3 and 6-4).

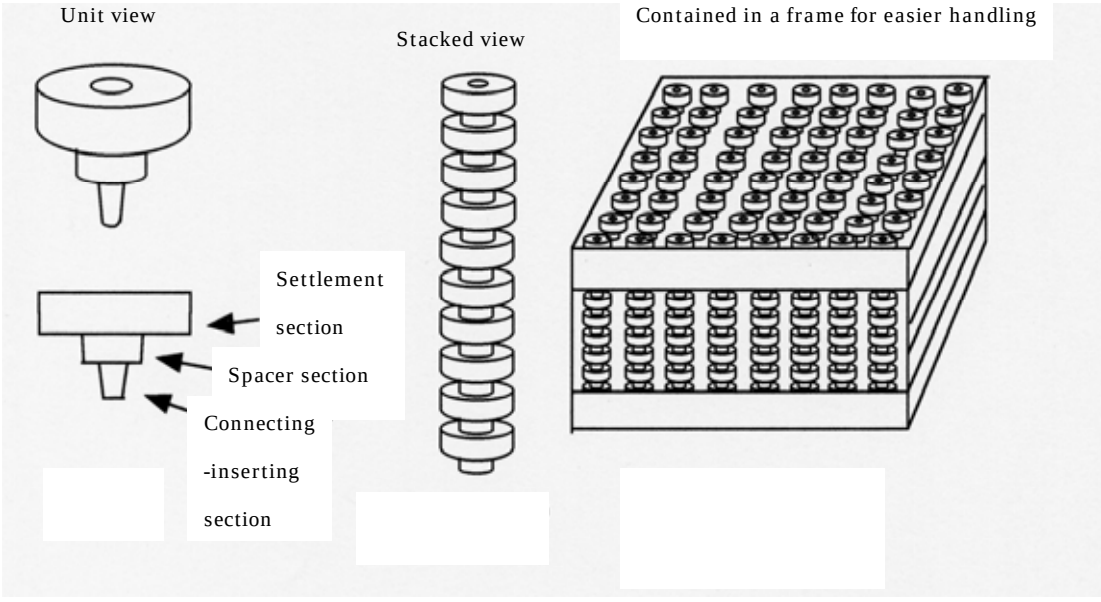

Fig. 6-3. Coral settlement device and assembling method

Fig. 6-4. Settlement devices assembled in a frame

Multiple settlement devices are stacked and arranged in a frame at close intervals. The gaps between the settlement devices formed by the spacer section protects the settled corals from being affected by the predation of invading sea-urchins. The stacks of settlement devices should be arranged in the frame at closer intervals to make it difficult for shells and fish to enter the gaps, preventing the predation from these life. Of course, it is assumed that some of the stacks of the settlement devices are thinned out to keep suitable intervals as the corals and sessile organisms grow.

After the juvenile corals have grown so that they appear on the side of the devices, up from the bottom side of the settlement section, make small holes on the bottom, put some adhesive in the holes, and them press the connecting settlement section of the device down into the adhesive. The connecting-inserting section is in the hole made on the upper surface of the settlement device, so that any life can hardly grow on it. The spacer section functions as a feature that prevents the juvenile corals from the bottom side of the settlement device from being damaged at the transplantation (Fig. 6-5).

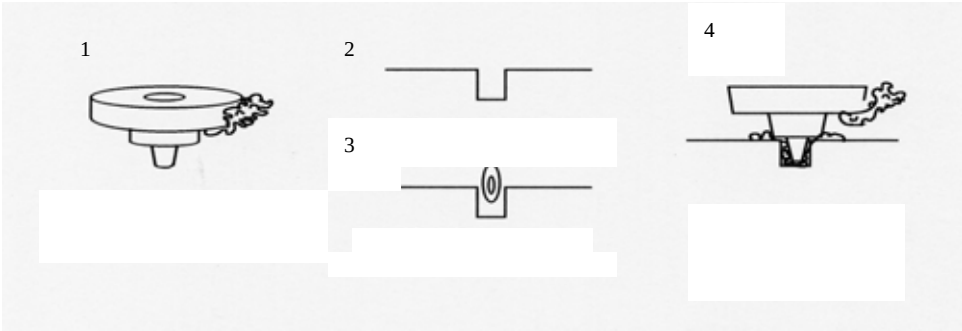

Fig. 6-5. Settlement device inserting and adhering procedure

1. Juvenile coral at 1st year after settlement. 2. Make hole on the substrate. 3. Fill the hole with adhesive. 4. Put the device into the hole to fix.

(6) Larval settlement and replant of substrate

In the natural world, many of the corals settle in relatively shallow waters, where it is hard for the corals to form the colonies because they are affected by strong waves (Fig. 6-6). When setting the settlement devices contained in a frame in such areas, it is safer to move them to calmer waters in early time, after the settlement. Such calm waters also provide advantages for maintenance workers such as; removal of predators, investigation of the state of growth and development of the corals; reduction of settlement devices performed as corals grow (to adjust the intervals between settlement device stacks); and easy access to such working areas. In the case of the Sekisei Lagoon, there are wide and calm moats to the south of Taketomi-jima, where suitable sites for rearing can be found in various places with depth of water ranging from 7 to 10m.

Fig. 6-6. Settlement devices contained in frames on the bottom

The settlement of larvae is completed generally in 10 days from the simultaneous spawning. Therefore, the settlement devices are moved to calmer rearing waters after about one month, after the larvae have surely settled and become stable. The movement to a nearby place can be made in the water with divers, but when moving to remote areas, an air lifter (a balloon-like bag which hangs an object under the water by using lifter buoyancy) can be used to float the corals easily under the sea surface (Fig. 6-7). In the future, when several hundred larvae are moved as a project base, the utilization of towing type table for underwater movement or enclosed floating pond for transportation can be expected.

Larvae settlement using this system has been performed experimentally, but has not reached the stage of routine transplantation work yet. However, the transplantation techniques are very simple, consisting of two steps: putting adhesive in the holes made on the substrate to which the larvae are transplanted, and inserting settlement devices in the holes and pushing them. As described in Chapter

4-2 “Transplantation of juvenile corals”, the steps can use conventional underwater working methods. We expect that the transplantation can be done by volunteer divers, not necessarily by professional divers, after these essential techniques are practiced.

Fig. 6-7. Moving the frames containing settlement devices to rearing water by suspension from air lifters

(7) Evaluation of methodology and perspective

Let us assume that juvenile corals are transplanted, to reefs located in the south of Sekisei Lagoon. This area is considered a good source of supply of coral larvae in Sekisei Lagoon because of relatively rich corals and high openness, and their high flow circulation environment. We consider that it is important for Sekisei Lagoon to maintain its coral regeneration capacity, and in the long term we view Sekisei Lagoon as the most important water for preventing Japanese corals found in the Kuroshio areas from extinction. As to the causes of extinction of transplanted corals in this water, two main factors are supposed: the rise of water temperature and predation by the crown-of-thorns starfish. Even with these factors assumed, the conditions for the transplantation in Sekisei Lagoon are far better than those of the Okinawa main island and Ishigaki Island where the corals near the shore are exposed to terrestrial run-off and other marine pollution.

This experiment has been performed to verify that a series of the coral regeneration project utilizing sexual reproduction can be implemented in waters where corals currently grow. That is, the verification is for the methodology for “strengthening the recovery capacity by performing a little manipulation of natural life without giving any damage to the coral community of a body of water” and “performing every intervention in the sea that the corals inhabit”. The undersea experiments are performed generally based on the results of many land experiments. The reason why we performed the sea area experiments hastily was that the number of the naturally settled corals is few, even in the

present situation, and we expected that comprehensive verification of this idea in the sea become more difficult in the future. Of source, this experiment was commenced based on the assumption that it can be implemented completely. However, the results of measurements of the number of settled larvae by using the settlement tile were, even in the southern sea area of Sekisei lagoon, much smaller than the values obtained in the periphery of Sesoko Island before 1998. Also, estimate that the number of the juvenile corals that can be obtained by the new settlement devices set in 2002 may be few due to the effect of the bleaching event in 2001.

We expect that the method can be applied practically and relatively easily by adopting easier and surer methods in the process of verification of the general idea of the coral reef regeneration through this experiment, if the verification is successful. For example, even if the number of settled corals in a body of water reduces significantly in the future, surer settlement may be achieved by setting many settlement devices in the land water tank or in the sea enclosed by a partition net, and rearing many larvae that are generated by the simultaneous spawning in it. The studies of such ideas has already being carried out (see Chap. 3-1). Figures 6-8 and 6-9 shows these ideas.

The advantage of this method is that the coral regeneration can be made in various places by boring into the existing substrate and then inserting juvenile coral-bearing devices into the holes. In the natural environment, the chance that the corals can settle on the artificial substrates or reefs is limited significantly with regard to the season. For example, possibility of natural settlement is low if the surface of the substrate has not been washed cleanly by waves in spawning time. Utilization of the settlement devices to which the juvenile corals are attached may compensate such inadequate condition.

It is inevitable that some corals die as the time elapses even if they are the naturally settled ones or those transplanted using the settlement devices. In such case, continuous transplantation of corals that are bred with the settlement devices can achieve coral reef regeneration surer (Table 6-1).

(8) Conclusion

This section has introduced an attempt to obtain juvenile corals for transplantation in the sea by utilizing the simultaneous spawning of corals. For the settlement and rearing of juvenile corals in the sea, it is necessary to understand the phenomenon of scattering of fertilized eggs that are generated by the simultaneous spawning and to collect valid information about the waters where the larvae settle. The southern reach of the wide Sekisei Lagoon has coral communities which are generally healthy and wide enough for the fertilized eggs to stay until they grow to larvae. Therefore, the area is considered one of the most suitable waters for rearing the juvenile corals. It can be said that Sekisei Lagoon has been able to sustain the wide coral reef area up to now due to such good natural conditions.

Table 6-1. Envisioned coral transplantation waters

1. Waters that are deemed supply sources of coral larvae (strengthening regeneration capacity) Open waters south of Sekisei Lagoon and Kerama Archipelago, etc. 2. Locations where the corals are damaged by a transient cause (regeneration) Damages from bleaching event (areas where seawater is exchanged well); Destruction of corals by predation of crown-of-thorns starfish, typhoon, and stranding of ships, etc. 3. Creation of coral reef using artificial substrates (garden making) Utilizing existing installations (reutilization); Utilizing new installations of structures (mitigation)

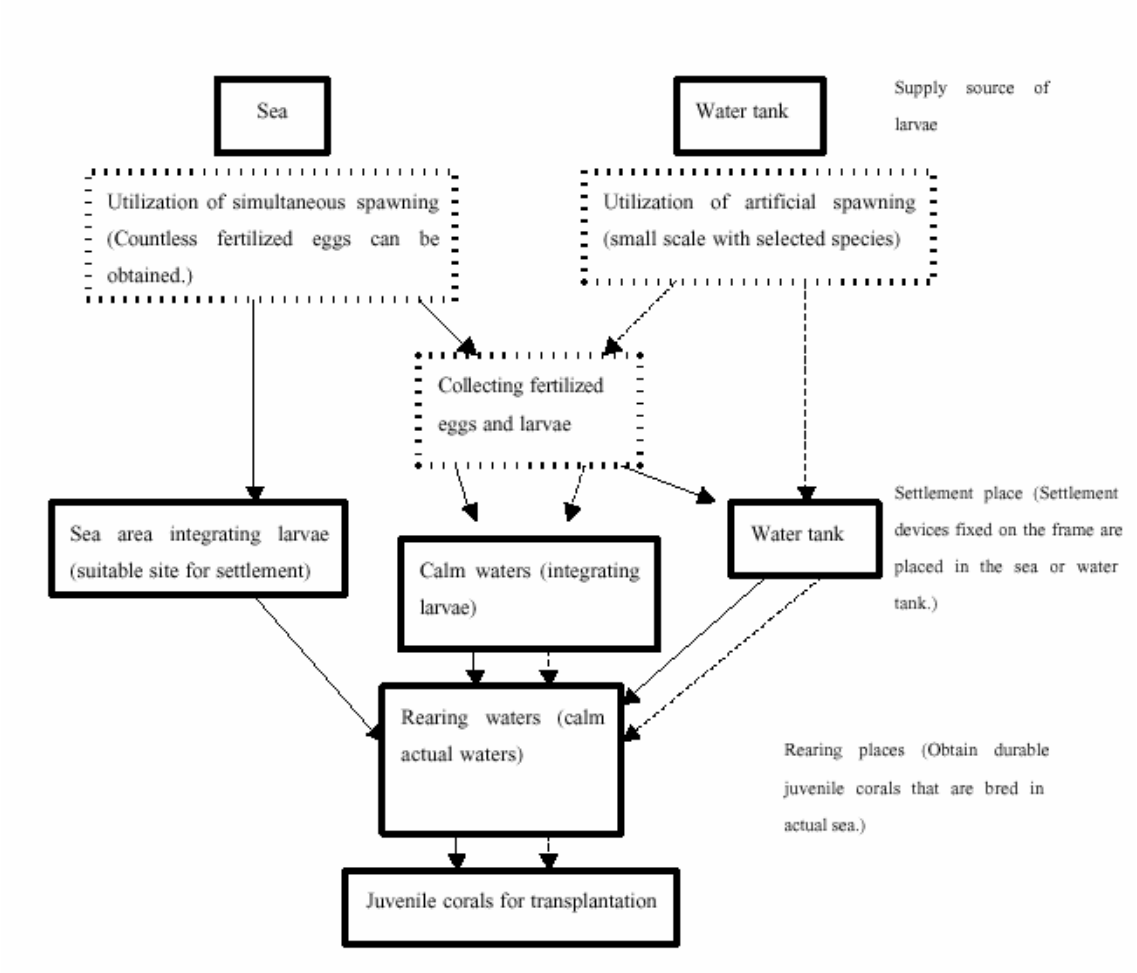

Fig. 6-8. How to obtain juvenile corals for transplantation

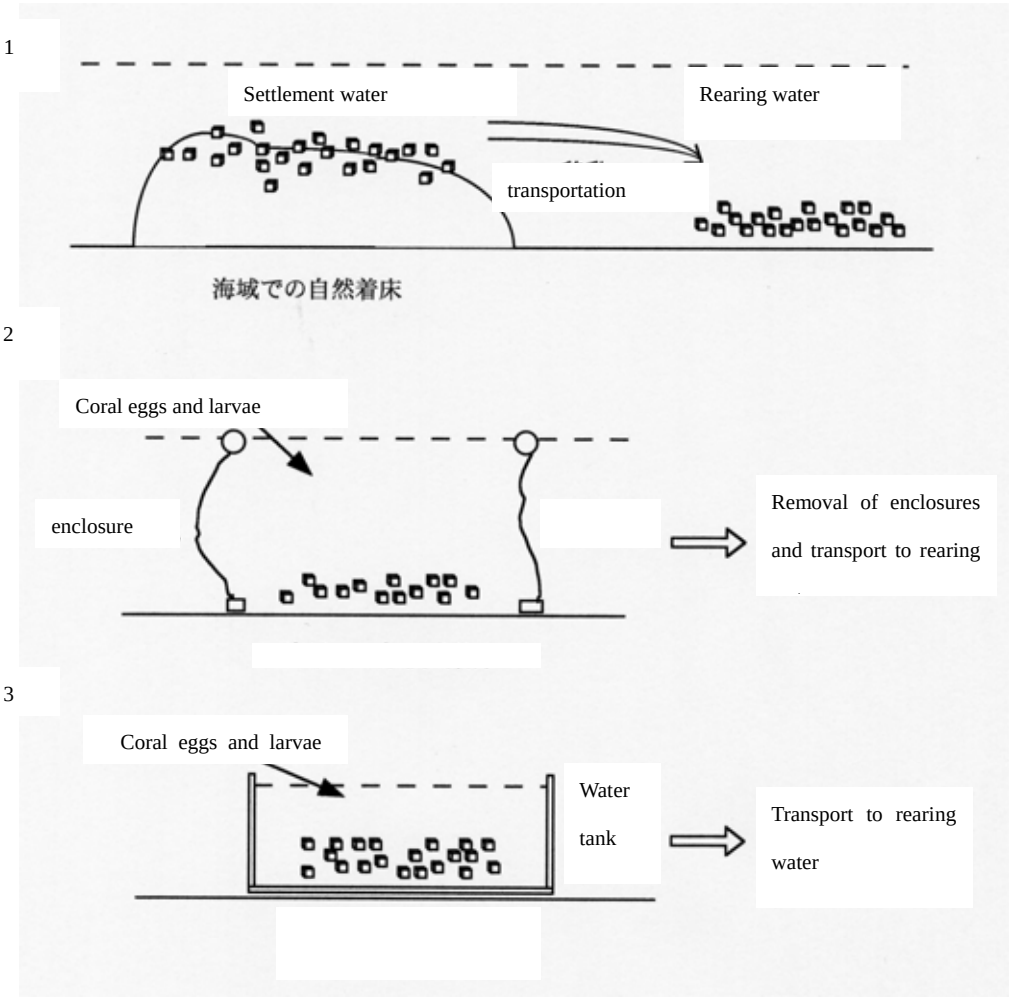

Fig. 6-9. Schematic view of process from settlement to rearing

1. Natural setting in the sea, 2. Integrated setting in the sea, 3. Integrated settlement in water tank

The settlement devices that were used for the experiment have features, such as strength suited to the transplantation that prevents damage from predators from implantation to the rearing by stacking the units. The use of ceramics allows flexibility for the forming, and utilization of the coal ash as the raw material instead of regular clay allows mass-production at lower cost. Needless to say, it is necessary to advance the practical application of this system through various elementary experiments and development of techniques, including not only the experiments in the sea but also those on the land.

Acknowledgment

The attempt to breed juvenile corals through sexual reproduction was implemented from 2001 based on the knowledge that was obtained when we engaged in a study of coral reefs conducted by Japan Marine Science and Technology Center. This experiment was made possible because it was

based on the many insights and basic understandings obtained through the intensive studies of the coral reefs conducted by the Center. We express our hearty thanks to the relevant persons of Japan Marine Science and Technology Center, who kept a close eye on our studies of the coral reefs and those who participated in and provided cooperation in the experiment in the sea and development of the techniques since FY2001

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(Okamoto M & Nojima S)

6-2. Experiment 2. Transplantation of coral fragments: *Acropora formosa*

(1) Material and methods

The location of the transplantation was Majanohama, Akajima, Kerama Archipelago, Okinawa. Because of its indented coast line, the current is slow except in typhoon. The average speed of the current in 2001 was 4.08 cm/sec (Taniguchi 2001). The amount of sediments is relatively large, and some turbid water flows out from the overhanging areas around the island. The water temperature in the period from 1999 to 2002 was in the range from 21.3 to 30.3℃ (Iwao 2001, Taniguchi 2002), and the monthly mean light intensity in the period from 1974 to 1990 ranged from 8.4 MJ/m^2 (January) to 20.5 MJ/m^2 (July) (Scientific Data Year Book 2000). The water for the transplantation is inhabited preferentially by *Acropora* on the reef flat, and the coral coverage by branching *Acropora* is 44%, the one by corymbose type *Acropora* is 21.7%, the one by tabular type *Acropora* is 7.0%, the one by sub-massive *Acropora* is 6.0%, and the one by other than Family Acroporidae is 21.4% (Taniguchi et al. 1999).

The species used for this experiment was branching *Acropora formosa*. In Okinawa, they begin to make oocyte in the period from the end of September to October, and spawn in May and June of the next year. Transplantation was performed on November 7, 1999, March 15, 2000, and August 11, 2000.

Fragments in three sizes, 5cm, 10cm and 20cm long, were taken from large donor colonies (approximately 1 m high, and surface area of approximately 4.5m^2), with a total of 470 pieces, by means of a wire cutter, and the lengths were measured using a vernier calipers having a measurement accuracy of 0.1mm. The fragments collected were put in a basket, and carried in the water to the destination for the transplantation 10 to 20 m from the donor colonies. The depth of the place where the fragments were collected and transplanted ranges from 2 to 3 m.

Concrete nails 8 cm long was driven into the substrate (coral rocks) approximately 3 cm, and then, the fragments were fixed vertically to the nail with cable ties (Fig. 6-10). The fragments of 5cm long and 10cm long were fixed to each nail with one piece of cable tie, and 20cm fragments were fixed using two pieces. The species normally grows vertically but for the experimental transplantation in November half of the fragments of each size were fixed horizontally to compare the results between that and the normal direction. After the transplantation, the survival rate of the transplanted fragments was measured every two months by SCUBA divers.

(2) Results

Among the fragments which were fastened using the cable ties and nails, no loss was found at the first year after the transplantation. Regarding most of the fragments that were fixed vertically, new polyps budded from the top of the branch. Many of them were tossed by the waves so as to wobble

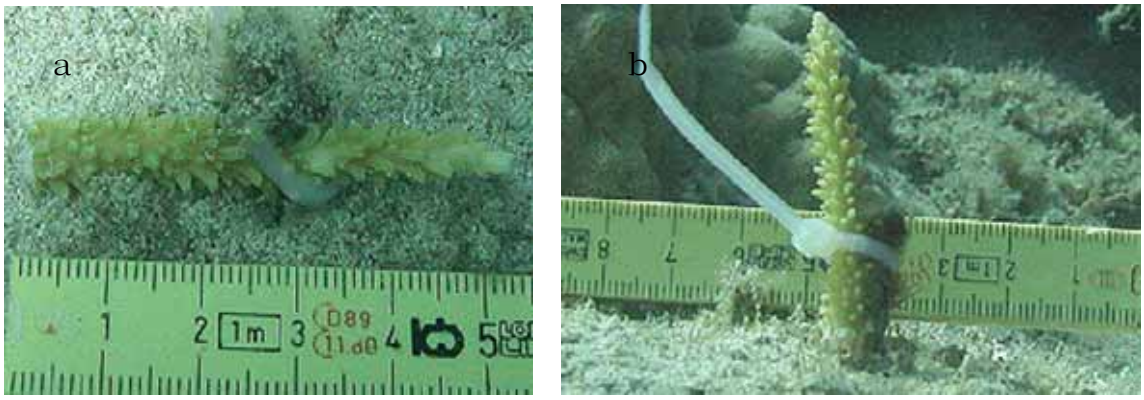

Fig. 6-10. Fragments of *Acropora formosa* immediately after the transplantation
a: 5cm fragment, horizontal b: 5cm fragment, vertical

even 1.5 years after transplantation, but no sediments were seen. The fragments that were fixed horizontally changed their orientation to vertical as the time elapsed after the transplantation. Among the fragments that were fixed horizontally, many of their surface areas contacted the substrate of the coral rock, and thus, they were stable and hardly tossed by the waves, but they also did not show the settlement to the substrate, and the surface of the fragments was covered with sediments consisting of fine particles. The bottom side of the fragments that did not get the sun died and were covered with algae. As a result of comparison between the vertically fixed fragments and horizontal ones that were transplanted in November, those vertically fixed showed higher survival rate for all sizes. For the fragments that were fixed horizontally, 90% of the large size ones survived, but 10cm and 5cm ones showed low survival rates and died one after the other after the observation. 5 cm fragments showed the survival rate as low as 10%.

The survival rates are summarized as follows. The vertically fixed fragments showed higher survival rates without regard to the time for transplantation. The 20 cm fragments survived nearly 100%. Among the fragments that were transplanted in August, the 10 cm ones also survived nearly 100% (Figs. 6-11 and 6-12). The effect of timing on the transplantation survival rates is clear for smaller fragments. For the 5 cm fragments that were transplanted in November and March, about 15% of them died within three months from the transplantation. In contrast, all of the 5 cm fragments that were transplanted in August died. The fragments that were transplanted in March did not die after that, but those transplanted in November had a reduced their survival rate, to as low as 28% within 1 year and a half after transplantation.

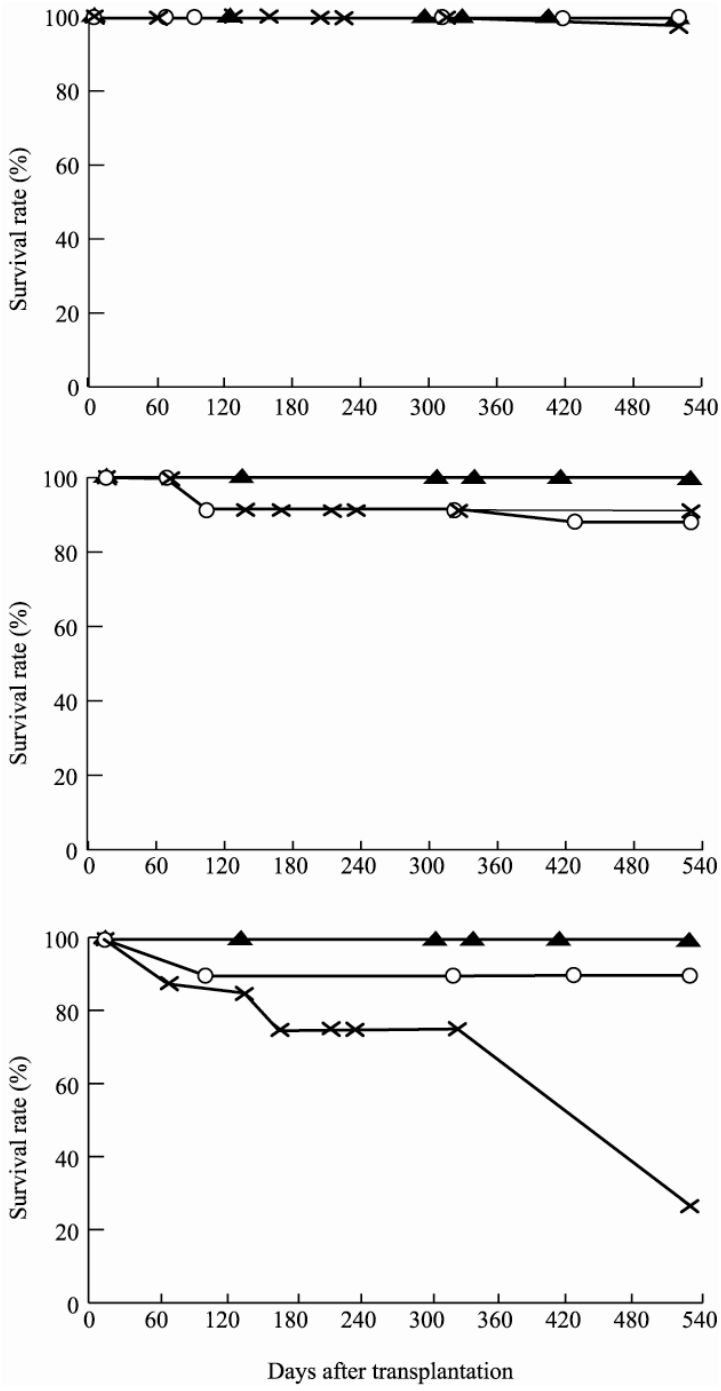

Fig. 6-11. Survival rate of vertically fixed *A. formosa* vs number of days after transplantation
a: 20cm fragments, b: 10cm fragments, c: 5cm fragments. ▲ : August transplantation; ○ : March transplantation; ×: November transplantation

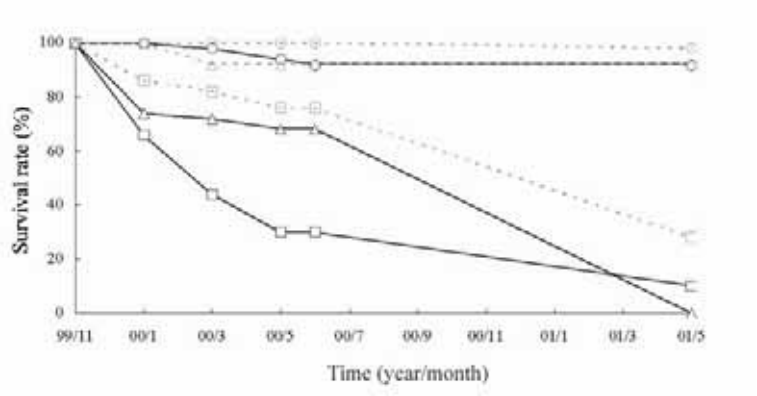

Fig. 6-12. Survival rates of horizontal *A. formosa* fragments transplanted in November (solid line)
The rates of vertically fixed fragments are shown with dotted line for comparison.
○: 20cm fragments, △: 10cm fragments, □: 5cm fragments

(3) Discussion

a. Fastening methods

Acropora formosa showed a higher survival rate when they were attached vertically than horizontally, mainly because of greater sedimentation. Since the surface of horizontal fragments is near the substrate, the sediments easily deposit and accumulate; the low profile of the fragments receive little water flow, preventing the sediments on them from being removed by the flow. Also, the predation of the polyps is disturbed, and photosynthesis does not occur to support zooxanthellae because of luck of sunshine. Algae can easily invade. Therefore, with all species, the vertically fixed fragments show higher survival rates.

b. Timing of transplantation

For the larger fragments, the survival rates are higher regardless of the season. But, when the survival rates of 5cm fragments transplanted in November and those in March are compared with each other, though the water temperature is higher in November than March, the survival rate of the 5cm fragments transplanted in November was 29%, which is low, but the rate of the 5cm fragments which were transplanted in March, where the water temperature is lower, is 87%. The values of the light intensity in November and March were nearly equal. Because the water temperature and light intensity rises from March, but these are reduced from November, we are able to conclude that transplantation can be successful even in water of 20℃ if the water temperature and light intensity rise within 2 months after transplantation, when they easily survive the stress due to the transplantation. With regard to the best season for transplantation, the fragments that were transplanted in August showed the highest survival rate. But, in the summer of the next year, many corals died from the bleaching event in the experimental region because of the continuous high water

temperature over 30℃ (Taniguchi 2002). Yap and Gomez (1984) who transplanted branching *Acropora pulchra* reported a reduction of the survival rate of corals that were transplanted in water of 30℃ or higher. Kajiwara et al. (1995) reported that the total amount of photosynthesis increased linearly in the range of from 20 to 30℃, and the calcification was maximized at 28℃ but reduced at 30℃. Since there is no doubt that the fragments are sensitive to the external stress in the period immediately after the transplantation, it is estimated that the best season for the transplantation is spring, when the amount of photosynthesis increases and there is no danger of a bleaching event.

c. Size of fragments and their survival rate

The survival rate of the 20cm fragments was nearly 100%. The larger the size the higher the survival rate is, and this relationship became clearer as the degree of stress from external causes (fastening method and the time of transplantation) became greater.

Based on the result that the survival rate lowers as the size of the fragment become smaller, there is an opinion that transplantation of an entire colony is more effective that fragmentation (Plucer-Rosario and Randall 1987). But, since the mortality rate does not exceed 3% for the fragments larger than a certain size (Connell 1973), taking a small number of fragments of sufficient size for the transplantation may produce a satisfactory survival rate. Nevertheless, it is important to know the size of fragments that can achieve the survival rate of 100%.

Also, since fragments of *Acropora* are fixed after the transplantation, and thus, are not turned over to damage the texture, the survival rate and growth rate of the transplanted fragments are higher than naturally made fragments. It is reported that the survival rate of branching *Acropora intermedia* fragments scattered loose on the reef flat at the 17th month after the transplantation was 31% for those of 6 to 9 cm long, and 81% for those of 13 to 16 cm long (Smith and Hughes 1999). Therefore, to do successful restoration of corals without damaging the donor colonies, it is considered that the best way is to fasten down the coral fragments produced by storm immediately after the event.

Since the 20 cm fragments that are transplanted and vertically fixed at the suitable time survived well, the size of fragments of *A. formosa* that is the most suitable for the transplantation wil be about 20cm.

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(Okubo N)

6-3. Experiment 3. Transplantation of a whole coral colony: *Porites lutea*

In recent years, occasional attempts to transplant whole coral colonies have been made in order to prevent possible extinction of coral communities due to events such coastal development works. In one example case, corals threatened by the installation of a bridge were transplanted from the bridge supports.

This section discuss the transplantation at Kouri-ohashi Bridge, that was built in the northern part of main island of Okinawa by the Okinawa Prefecture (Dept. of Civil Engineering and Construction, Okinawa Prefecture and Metocean Environment Inc. 2002).

(1) Method

On the bridge support section in shallow water located on the main island side of Kouri-ohashi Bridge, massive *Porites* of 5 to 80 cm diam. were distributed preferentially (Fig. 6-13). Of those corals, the communities of *Porites lutea* ranging from 20 to 30 cm in diameter were mainly collected and transplanted in December 1998 in sandy bottom near the eastern part of the bridge. The depth of the artificial substrate was approximately 2m below the reference water level, and that of the natural base rock was approximately 1m below the reference water level.

In all, 25 colonies were transplanted to five pieces of concrete substrates, and 5 colonies to the natural base rock, and the follow-up investigations were performed for four years, five times a year. The investigation items were the states of corals mainly including: survival and death, growth, activity, and deposition of soil.

Each concrete substrate consisted of a concrete block plate (1.2m×1.2m×0.2m) with five holes (10 cm diam. × 5 cm deep) in the surface for transplantation (Figs. 6-14 and 6-15).

The collection of *P. lutea* from the colony base was performed by using a chisel, taking care not to damage the corals. The corals removed from the bottom were put into a container in the water and transported to the place where they were transplanted. To adhere the corals to the substrate, waterproof adhesive was used. To adhere them to the natural base rock, holes for the transplantation (similar to those made on the artificial substrate) were made on the rock beforehand by using a chisel and a hammer. Later, a method to fasten the corals to anchor bolts driven into the colony base substrate was used; the colonies grew to wrap around the bolts, demonstrating that the method was effective for the settlement.

(2) Results

a. State of survival and death

One of the colonies transplanted to the concrete substrate was dead within 7 months, but other ones on both concrete and natural base rock survived for 45 months after the transplantation,

Fig. 6-13. Massive *Porites* for transplantation

Fig. 6-14. Artificial substrate placed on the sea bottom

especially the colonies transplanted to the natural base rock which scarcely showed even partial death of the polyps. Regarding the colonies transplanted to the artificial substrates, while there was partial death due high water temperature in summer, did not lead to overall extinction.

b. Growth

When the annual average growth (width and height) of the colonies transplanted on the concrete substrate and the ones on the natural base rock are compared, the growth of corals on the concrete in 1999 was higher for both height and width. But, for those 1st and 2nd years after transplantation the growth on the natural base rock was higher than on the concrete substrate, with the difference ranging from 3 to 4 mm/year in width, and 2 to 5 mm/year in height. The growth of colonies which had many polyps killed on the concrete might have been affected by that event (Fig. 6-16).

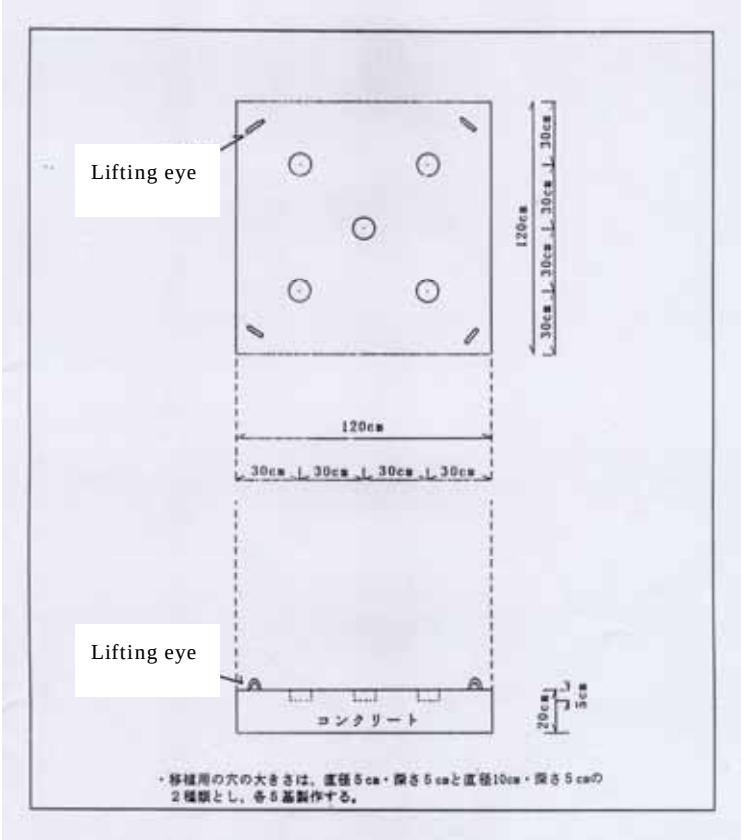

Fig. 6-15. Dimensions of concrete substrate

Two types of the concrete substrates, one with holes for transplantation (5cm diam. × 5cm deep) and another with holes (5 cm diam. × 10 cm deep), are prepared.

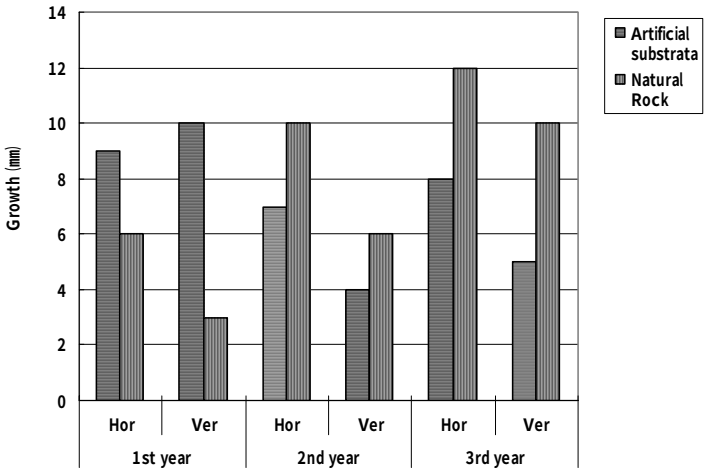

Fig. 6-16. Annual average growth after transplantation in December 1998.

Hor-Horizontally fastened, Ver-Vertically fastened

c. Activity

The data on the annual variation of healthy coral colonies (not those bleached and those with thin color) show that the colonies on the natural base rock tend to have higher health rate for each year than the ones on the concrete substrates. In the 3rd year after transplantation, many coral colonies were bleached due to the high water temperature in summer 2001, resulting in reduction of healthy ones (Fig. 6-17).

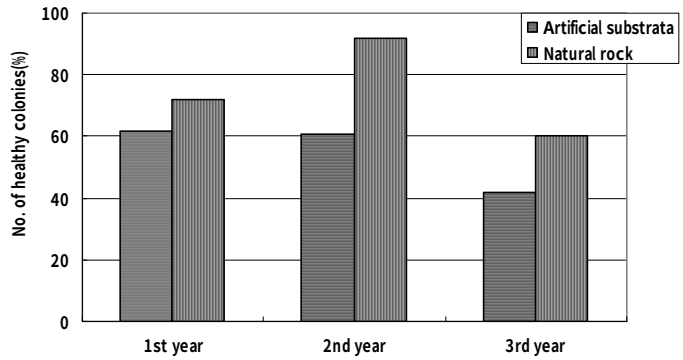

Fig. 6-17. Percentage of healthy colonies to the whole after transplantation in December 1998

d. Number of mucus secreting colonies

The results of observation of the soil sedimentation show that the concrete substrate tends to have more sediment than the natural base rock, and thus, the number of colonies that secrete mucus as an action to eliminate soil sediment is greater on the concrete substrate for each year. In 2001, the number of colonies that secrete mucus increased, possibly due to the high water temperature in summer (Fig. 6-18). The phenomenon of colonies covered with the sediment and secretion frequently to eliminate the soil was observed, and it is estimated that this activity may require much energy.

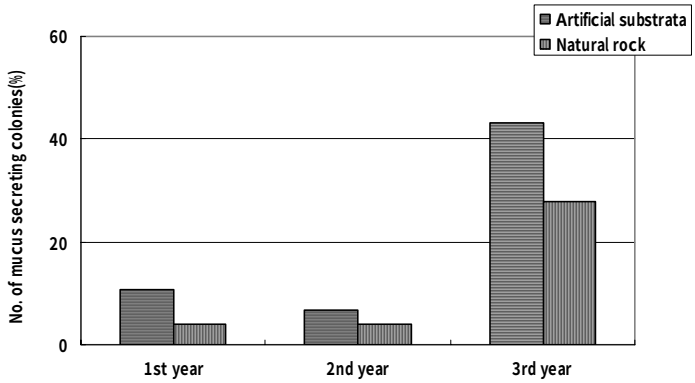

Fig. 6-18. Percentage of mucus secreting colonies to the whole after transplantation in December 1998

According to conclusions drawn from observation of generative cells of *P. lutea* on the eastern beach of Ishigaki Island, the percentage of colonies that have generative cells was less on the area with much red soil sediment than on the area with less red soil sediment, giving an indication that the formulation of generative cells diminishes when the corals have to consume energy to eliminated sediment (Harii and Nadaoka 2002). This fact indicates that *Porites lutea* should be transplanted to natural base rock to prevent the effect of the sediment on their regeneration.

(3) Conclusion

Since *P. lutea* is found much in the moats, it is recommended that they be transplanted to moats. According to the results of investigation of massive *Porites* distribution in the moats of Miyara Bay, Ishigaki Island, colony density is higher in the areas with deeper bottoms, where many of the colonies are distributed on the bottom at 2 to 3m deep (Sato et al. 2000). Based on these observations, it is desirable to transplant colonies in the deepest possible bottom in the moats for stable settlement. However, because the bottom of moats is mainly covered with sand, it may be difficult to find firm natural substrate, and often sort of artificial substrate is needed. Since the use of anchor bolts has provided acceptable results, as described before (Fig. 6-19), installation of reinforcing steel stakes that can be used on sand bottom may be adequate. According to the results of experimental use of reinforcing steel stakes on the sand bottom, stakes of 12 mm in diameter and 1m long are easy to handle. A pneumatic hammer can drive the stakes easily, and it is recommended that stakes be sharpened so that they can crush or push aside gravels that may be encountered during the driving. Since the reinforcing steel stakes are relatively soft, their forms can be finely adjusted after the installation by bending.

As for the size of the colonies to be transplanted, it is estimated that those of 20 to 30 cm in diameter are easy to handle by divers. However, massive *Porites* exceeding 1 m in diameter or length is not uncommon, and when there is a need to transplant the colonies of such scale, it is necessary to examine the colony size and the bottom topography of each location, so that they can be firmly fixed on bottom.

Fig. 6-19. Coral wrapping around an anchor bolt

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(Hosoya S)

7. Maintenance of polyps on substrate, transplanted fragments and colony, and replaced reef

(1) Removal of algae and predators

Since algae can easily attach to the settling part of transplanted coral fragments, and the algae may grow to cover the corals leading to possible death, it is necessary to monitor and remove the algae frequently (Fig. 7-1). The monitoring is also necessary when organisms eating corals, such as crown of thorns starfish or coral-eating gastropods are distributed around the transplantation area. Especially immediately after the transplantation, the corals frequently secrete mucus because they were cut, which can attract crown-of-thorns starfish. They should be eliminated from the periphery before the transplantation.

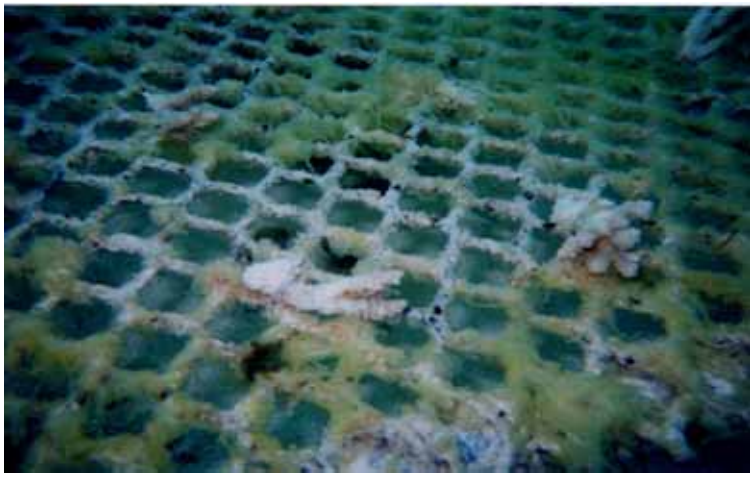

Fig. 7-1. *Acropora tenuis* on the substrate for transplantation covered with algae (March, 2002)

(2) Announcement and enlightenment

When the location of the transplantation is near a beach, surface-cast fishhooks can snag on the transplanted corals. So, it is necessary to obtain cooperation from sportfishermen by setting a notice board calling on them to avoid the fishing near the location. The most serious cause of damage to the transplanted corals is the anchoring by boats. So, it is necessary to set a buoy to indicate that corals are transplanted in the area, and to set a mooring buoy in the areas where boats may otherwise anchor frequently. It is also desirable to involve parties concerned with the transplantation by obtaining the cooperation from fishermen and diving services. For the fishermen and diving services, the coral reefs are places of their business, and thus, the coral reef regeneration is their prime concern. So their positive participation can be expected. The transplantation may also lead to deepening of the public awareness of the coral reefs, possibly promising an environment-educational effect. However, for successful transplantation, it is necessary that sufficient technical instructions are made, and Natural Parks Law and ordinances related to fisheries regulations are observed.

In Sekisei Lagoon, organizations that are related to the coral reefs such as International Coral Reef

Research and Monitoring Center of Ministry of the Environment, Okinawa Prefecture, Municipality of Taketomi-cho, Yaeyama Marine Park Research Station, and Union of Diving Services set up Yaeyama Coral Reef Conservation Conference for regeneration of the coral reefs of Iriomote National Park. They performed transplantation of coral fragments as part of its project, which continued for several years with support from various funds. The project contributed to the regeneration of the coral reefs, and led to other positive activities of the Conference (Fig. 7-2). Activities like these are also performed around Akajima, Kerama Archipelago, where the local fishermen's Cooperative and Aka-Geruma Diving Association participate in activities such as reef checking, removal of crown-of-thorns starfish, and transplantation of corals with the cooperation of the Akajima Marine Science Laboratory.

Fig. 7-2. Volunteer diver performing transplantation of corals

(3) Monitoring

It is important to monitor how the transplanted or relocated corals survive over time for evaluation of the techniques and results of transplantation or the relocation. For the details of the monitoring, the following items should be noted.

a. State of survival and death of corals

Corals can be classified as follows according to the state of extinction of the transplanted coral colonies;

Healthy corals: no extinction at all,

Partly dead corals: extinction less than 30%,

Partly alive corals: more than than 30% alive

Very small number of corals alive: extinction equal to 70% or over and less than 100%,

Extinct：No corals alive,

Disappeared: coral colonies disappear.

b. State of activity of corals

Coral colonies can be classified as follows according to the state of transplantation;

Good: Color of polyps and coenosarc is normal, no abnormality in extension of tentacles, and not covered with much mucous membrane.

Bad: polyps and coenosarc are bleached, the surface is covered with much mucous membrane, or dead area is expanded by the abnormal growth of algae and sea sponge.

Basically monitoring of transplanted corals is performed in each season, and it is necessary to add a monitoring at the first month immediately after transplantation, where corals are unstable. When juvenile corals made from settled larvae is on the bottom, it is assumed that colonies may take approximately four years until they present a scene close to the natural state. Therefore, it is necessary to continue monitoring five years including the follow up investigation.

For scientific assessment of a transplantation, the transplanted communities should be compared with natural communities. However, there may be no normal colonies near by. Sometimes it is impossible to find a comparison in an area previously cleared of healthy coral by bleaching or other natural or human causes.

c. Monitoring of habitat environment

When monitoring corals, it is necessary to monitor their habitat environment at the same time to understand it as one of several factors affecting coral survival. The important environmental factors that can affect the survival of corals are water temperature, water movement, sedimentation and predators. Long term rise of water temperature kills corals, which is why coral bleaching due to high water temperature is a worldwide problem. By monitoring water temperature continuously for long time, it is possible to discover water temperature trends in locations that affect survival of the corals. Recently, compact and low cost self-recording water temperature thermometers have come on the market. Some are now can measure the temperature hourly for six months used for temperature measurement in the coral reef waters (Nature Conservation Bureau, Ministry of the Environment 2001).

Water movement is also a factor that can affect the survival of corals. Many species have great need for the transplantation to locations with more water movement (Marine Parks Center of Japan 1995). As a simple and easy method for discovering the movement characteristics of the transplantation destination, there is a measuring method that uses calcium sulfate balls (Nabeshima and Kida 1990; Komatsu 1992; Komatsu and Kawai 1992; Furushima et al. 2000, 2001). Since this method provides the state of the cumulative tidal flow in time series, it is suitable for knowing the average characteristics of the site. This method allows repetition of measurement at low cost.

On some coral reefs, terrestrial run-off of soils occurs frequently, which affect mainly the

environment of moats. It has a serious impact on the survival of corals. Among the methods for discovering the state of the sedimentation are visual observation to see the state qualitatively, the use of sediment traps, and the SPSS method that measures the optical penetration of object to be examined (Ohmija and Mitsumoto 2001). These methods are easiest in the following order: visual, SPSS and trap. It is recommended to pick one of them for scale of monitoring because any of them can provide useful data for any purpose.

There are some other important factors such as light photon and nutrient salts, which should be selected as necessary because they require much cost for long term measurement.

(4) Reproduction

The speed of growth and survival rate should be thought of as more important than mere regeneration with regard to the transplantation of coral fragments, but if the transplanted corals can spawn, that should contribute more to the recovery of total coral reefs because the coral supply source is increased. Since the possibility of the spawning is increased as the transplanted colonies grow, verify the spawning at the time of spawning. Though the time of spawning varies from year to year and among the various waters, it is known that many species usually spawn around the full moon in early summer night (see Chapter 3-1). Therefore, it is desirable to verify the spawning by continuously monitoring the polyps (Fig. 7-3).

Fig. 7-3. Genital gland of matured *Acropora* (photo by K Shimoike)

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(Fujiwara S)

8. Concluding observations

(1) Techniques of reef rehabilitation: development and comparison

The very first case of coral transplantation in Japan was possibly the one for the restoration of undersea scenery around the undersea observatory that was constructed in 1970 in Kushimoto, Wakayama Prefecture by the Marine Park Center. This water is a typical area of hermatypic coral distribution in Honshu which is known for its wide community of tabular type *Acropora hyacinthus*. As a result of experimental transplantation (fragment) using the similar form corals, it is reported that the species transplanted at this time grew to a size that shows the feature of colonies within 1 to 2 years (Tatsuki 1977). Worldwide, the transplantation of coral fragments has been implemented as a technique for recovery of coral reefs that suffered a large scale coral predation from an explosive infestation of crown-of-thorns starfish from 1960's to 1970's. Then efforts made little progress in the recovery. Nowadays, several organizations (such as Reef Ball Development Group, Ltd: www.artificialreefs.org), that promote restoration of coral reefs deteriorated by human activities, are taking an active part. Also an attempt to increase employment opportunities of fishermen in developing countries has been made through the restoration activities and production of ornamental coral (for example, Heeger et al. 1999). If a full-scale coral farming (seed production and culture or transplantation of coral fragments) can be set underway by Japanese research laboratories that have the highest developed techniques in the world for fishery farming, they will be able to support development of restoration techniques based on their actual performance and transfer them to developing countries in sub-tropical and tropical regions, resulting in better conservation of the world's marine environment.

If the coral reefs suffer widely from the predation by crown-of-thorns starfish, resulting in extinction of most of the coral communities, the recovery takes very long time. Their case was an extinction of almost all corals from the severe infestation in Sekisei Lagoon of Okinawa in 1981. It took dozens years until the recovery could be fully recognized (Mori 1995). The progress of the recovery in this period has not been simple. Recovery is usually retarded in the moats, that have closed features, as compared with reef edges which are open. It is common that different and the places, even if they are closed, generally show a difference in the degree of the recovery linked to the differences in their habitat environments. The effects of the physical environment of waters, such as uncertainly of larvae recruitment and impact of wave motion or influx of water on the growth and development of corals and formation of the reefs have scarcely been studied. So now is the time to advance studies of selection of suitable sites for the release of larvae or transplantation.

The sedimentation from terrestrial run-off became prominent from the 1980's in Okinawa, which disturbed the settlement of juvenile corals and prevented their growth, making the process of the recovery complex. The variation of the degree of the recovery of coral reefs among locations is

greatly affected not only by the possibility of supporting juvenile corals and their survival rate, but by the deterioration of habitat environment. Under such circumstances, it is vital to attempt positive restoration (not simply to wait for natural recovery).

The techniques for restoration of coral reefs was initiated by transplantation of fragments. They have been evolved to the larval production techniques that utilize sexual reproduction, and then to the larvae settlement induction techniques. This process seems to show that the techniques are advancing toward measures that do not affect the existing coral communities. Since it is expected that coral reef restoration techniques will be evolved more through further studies in the future, this document has made clear the status of past and present research and has collected them systematically. Table 8-1 summarizes the restoration techniques for comparison.

The technique that can be implemented immediately is the transplantation of coral fragments if one has a certain level of ecological knowledge and transplantation technique. Since it can be implemented if the nails and wires are available, this is the method that was adopted earlier than the other techniques. After that, the adhesive that is effective in seawater became available, which has become widely used as a typical method of the transplantation. The method that attempts to increase the coral by utilizing the sexual reproduction while minimizing the effect on the donor colonies mainly include production of larvae in the laboratory, collection of eggs in the field, and induction of the settlement. Since each of the methods has specific features, it is desirable to select the most suitable method based on the situation. The seed production is carried by using an indoor water tank or outdoor floating ponds, thus involving higher material than the other methods, but it allows relatively stable collection of the seed and allows selection of the species. The larvae-releasing method can settle the seeding in wide range. The labor and material cost of the larvae settlement induction method is lower than the other methods. The larvae settlement induction method, in which the reach of larvae is affected by the weather and oceanographic condition, can restore the coral more assuredly if the movement of the drifting larvae can be predicted more assuredly and a method that can detain the slick near the transplantation structure is developed.

For the growth of corals, Yamamoto et al. (2002) shows an approximate formula for obtaining the growth at the depth of water of 1m and inclination angle of 10° based on the results of continuous observation for six years in Naha Harbor.

Coral coverage $y = 0.0227e^{1.25t}$ ($R^2 = 0.69$, $n = 9$ and t: year)

The approximate formula for obtaining the growth of corals at the depth of water of 0.5 m based on the report of investigation of natural coral communities on Heron island of the Great Barrier Reef is as shown below.

Coral coverage $y = 0.0261e^{1.10t}$ ($R^2 = 0.95$, $n = 4$, and t: year)

According to the results of calculations using these formulas, the coral coverage increases rapidly at 3rd to 5th year and enters the stable growing period at 6th year. This is why the “Time to

Table 8-1. Comparison of coral reef restoration techniques

	Fragment transplantation	Juvenile coral transplantation	Seedling production	Larvae releasing	Induction of larvae settlement
Outline of the method	Colony fragmentation and fixation on substrate	Collection of juvenile colonies and transplantation	Collection of larvae from tank or field, nurture larvae, induce settlement on plate, and transplant plate to the field	collection and nurturing larvae is same as left. Transport larvae and releasing (establish sheet on destination)	Induce larvae settlement on settlement tool or surface processed structure
Time required	ca. three years (fix in spring)	ca. three years (can fix year-round)	Five years after transplantation (collect larvae in summer)	ca. five years after settlement (settle in summer)	ca. five years after settlement (settle in summer)
Equipments and materials	Wire, nail, cable tie, and adhesive	Adhesive	Nurturing devices, settlement plate, and adhesive	Nurturing devices and surface processed structure	Surface processed structure
Required effort*	100 fragments fixation/worker/day	same as left	ca. one week, every-day care for larvae nurturing, and 100 plates fixation/worker/day	ca. one week, every-day care for larvae nurturing	Transplantation and collection of settlement tools, and structure processing
Scale of restoration	Number of settlement plate will be restricted by number of divers	same as left	Amount of seedling and fixation will be restricted by nurturing facility and number of divers, respectively	Relatively broad scale, although amout of larvae will be restricted by scale of nurturing facility	Number of settlement tool and processed structure will be restricted by number of divers and project's scale

*Required efforts after fixing on to the field substrate is common, thus will be abbreviated.

restoration" shown in Table 8-1 is "approximately 5 years". Chapter 7-3 describes the required monitoring period as 5 years.

In addition to the techniques shown in Table 8-1, there is a method that transplants all colonies of *Porites lutea* discussed in Chapter 6-3. This technique is a little different from the restoration of coral reefs by transplantation, which is a kind of emergency evacuation of the communities that may otherwise be killed off. That is why it was introduced only as an example case and not listed in the Table 8-1. The relocation of an entire coral reef is also not listed in the table for the same reason.

(2) Future subjects to study

It is expected that the mainstream of the future coral reef restoration techniques will be improvements in the handling of larvae. In contrast with relatively closed waters where the movement of larvae can be predicted relatively easily, the open sea areas, it is essential to predict the movement of larvae slicks because they are transferred long distance in a short period by the wind currents. However, the current significant lack of oceanographic information makes it impossible to predict the movement of the drifting larvae and thus, it is crucial to develop the research in this field.

When planning the transplantation or release of corals, selection of the suitable destination is a very important matter. Needless to say, successful restoration requires the favorable growth of corals introduced into a new environment, and expansion of their habitat through satisfactory generation of the progenies. To achieve that, it is necessary to make clear the relationship between each stage of the overall life of corals and field factors, including biological causes, such as the effects of interspecies competitions and predation from coral feeding fish such as damselfish. As discussed previously, our knowledge for microscopic or mesoscopic physical and topographic factors such as flow of water and inclination in the coral reef areas are scarce. These also are important research subjects in the future.

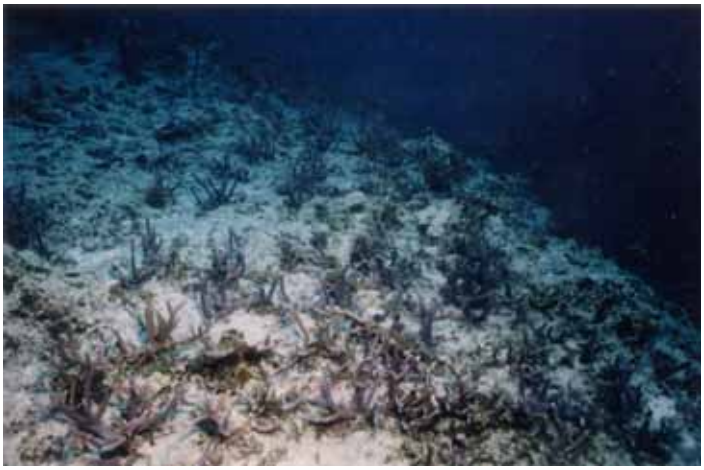

Fig. 8-1. Transplanted branching *Acropora*

Finally, we would like to discuss the effects of genetic disturbances by hybridization on the coral population and the deterioration of genetic diversity of the coral communities that can be caused by the restoration projects. The transplantation of corals may cause no problem if the destination is within the range of natural diffusion of the spawn. But the long distance transportation of corals may disturb the characteristics of the local genetic constitution.Since it is well known that hermatypic corals have high genetic diversity and easily form hybrids (Hatta et al. 1999; Willis 1997), it is necessary to obtain the eggs and coral fragments from many colonies that are distributed as wide as possible, regardless whether they are used for the seed production or for the fragment transplantation. If the number of donor colonies is limited or the diversity of their genetic pool is lacking, coral communities that are very susceptible to environmental change or disease agents may be made. It is known that *Polites compressa*, that is found in Hawaiian Islands, shows high genetic diversity in the places with relatively large environmental disturbance and low genetic diversity in the places with relatively small environmental disturbance (Hunter 1993). Molecular biology, that is making considerable progress today, should be applied to the estimation of the size of genetic diversity of current coral communities. The results could be used as a reference for larvae production in laboratory and in the transplantation of coral fragments from the source sites.

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(Fujiwara S & Omori M)

Profile of Authors

Makoto Omori
Director
Akajima Marine Science Laboratory
Aka, Zamami-son, Okinawa-ken 901-3311
makomori@amsl.or.jp

Shuichi Fujiwara
Senior Researcher
Okinawa branch, Metocean Environment Inc.
2-6-19 Aja, Naha-shi, Okinawa-ken 900-0003
fshuichi@notes.metocean.co.jp

Masayuki Hatta
Associate Professor
College of Science
Ochanomizu University
2-1-1 Otsuka, Bunkyo-ku, Tokyo 112-8610
mhatta@cc.ocha.ac.jp

Seiichi Hosoya
Director
Okinawa branch, Metocean Environment Inc.
2-6-19 Aja, Naha-shi, Okinawa-ken 900-0003
seiiti@notes.metocean.co.jp

Kenji Iwao
Researcher
Akajima Marine Science Laboratory
Aka, Zamami-son, Okinawa-ken 901-3311
iwao@amsl.or.jp

Satoshi Nojima
Associate Professor
Amakusa Marine Biological Laboratory
Kyushu University
Tomioka, Reihoku-cho, Kumamoto-ken 863-2507
satoshi@mbox.ambl-unet.ocn.ne.jp

Mineo Okamoto
Associate Professor
College of Marine Science
Tokyo University of Marine Science and Technology
4-5-7 Konan, Minato-ku, Tokyo 108-8477
okamotom@s.kaiyodai.ac.jp

Nami Okubo
Graduate School
Department of Life Science
Tokyo Institute of Technology
2-12-1 Okayama, Meguro-ku, Tokyo 152-8551
namiokubo@nifty.com

Hiroki Taniguchi
Researcher
Akajima Marine Science Laboratory
Aka, Zamami-son, Okinawa-ken 901-3311
taniguchi@amsl.or.jp