REPRODUCTION OF A SPONGE, *PETROSIA* SP. (PORIFERA : DEMOSPONGIAE : HAPLOSCLERIDA : PETROSIIDAE) IN CORAL COMMUNITIES OF THE INNER GULF OF THAILAND

SAIPRATEEP ASA

A THESIS PRESENTED TO RAMKHAMHAENG UNIVERSITY
IN PARTIAL FULFILMENT OF THE REQUIREMENTS
FOR THE DEGREE OF MASTER OF SCIENCE
(BIOLOGY)
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การสืบพันธุ์ของฟองน้ำ *Petrosia* sp. (Porifera: Demospongiae: Haplosclerida: Petrosiidae) บริเวณกลุ่มปะการังอ่าวไทยตอนใน

สายประทีป อาษา

วิทยานิพนธ์เสนอต่อมหาวิทยาลัยรามคำแหง
เป็นส่วนหนึ่งของการศึกษาตามหลักสูตรปริญญา
วิทยาศาสตรมหาบัณฑิตสาขาชีววิทยา
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Thesis Title	Reproduction of a Sponge, *Petrosia* sp. (Porifera: Demospongiae: Haplosclerida: Petrosiidae) in Coral Communities of the Inner Gulf of Thailand	
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ABSTRACT

Thesis title : Reproduction of a Sponge, *Petrosia* sp. (Porifera: Demospongiae: Haplosclerida: Petrosiidae) in Coral Communities of the Inner Gulf of Thailand

Student's Name : Mr. Saiprateep Asa

Degree Sought : Master of Science

Major : Biology

Academic Year : 2000

Advisory Committee :

1. Dr. Thamasak Yeemin Chairperson
2. Asst. Prof. Dr. Sutat Subinprasert

Sexual reproduction and ecology of a marine sponge, *Petrosia* sp., (new species) were studied at Khang Khao Island and Nok Island, the inner Gulf of Thailand during December 1997 – April 2001. The sponge grew mostly on a dominant massive coral, *Porites lutea*, and obviously showed clumped dispersion. Reproductive biology of *Petrosia* sp. was examined by monthly sampling of marked individuals for histological analysis in the laboratory. *Petrosia* sp. was a hermaphroditic brooder. Its gamete development from the two study sites showed the same pattern. Spermatic cysts, oocytes, embryos and larvae were found during December 1997 – April 1998. The severe coral bleaching phenomenon in the Gulf of Thailand during

April - May 1998 resulted in disappearance of gametes and larvae in all samples collected during May - October 1998. The fecundity was higher in the sponge from Nok Island. Releasing of parenchymella larvae was observed in the morning in aquaria. Most larvae settled on live colonies of *Porites lutea*. Average growth rate of *Petrosia* sp. at Nok Island (0.7 cm^3/day) was significantly higher than that at Khang Khao Island (0.6 cm^3/day). Average mortality rate of *Petrosia* sp. at Nok Island (0.9 cm^3/day) was higher than that at Khang Khao Island (0.8 cm^3/day). Results from the present study progress debates concerning life history strategies and taxonomy in marine sponges.

BRT 541015

บทคัดย่อ

ชื่อเรื่องวิทยานิพนธ์ : การสืบพันธุ์ของฟองน้ำ *Petrosia* sp. (Porifera: Demospongiae: Haplosclerida: Petrosiidae) บริเวณกลุ่มปะการังอ่าวไทยตอนใน
ชื่อผู้เขียน : นายสายประทีป อายา
ชื่อปริญญา : วิทยาศาสตรมหาบัณฑิต
สาขาวิชา : ชีววิทยา
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ศึกษาการสืบพันธุ์แบบอาศัยเพศและนิเวศวิทยาของฟองน้ำทะเล *Petrosia* sp. ที่เกาะค้างคาวและเกาะนกบริเวณอ่าวไทยตอนใน ตั้งแต่เดือนธันวาคม 2540 – เมษายน 2544 พบว่าฟองน้ำส่วนใหญ่เจริญบนปะการังก้อน *Porites lutea* และมีรูปแบบการแพร่กระจายเป็นหย่อม จากการศึกษาชีววิทยาการสืบพันธุ์ของฟองน้ำ *Petrosia* sp. โดยเก็บตัวอย่างฟองน้ำที่ทำเครื่องหมายไว้ทุกเดือนแล้วนำมาวิเคราะห์เนื้อเยื่อด้วยวิธีทางไมโครเทคนิคในห้องปฏิบัติการ พบว่าฟองน้ำชนิดนี้เป็นกะเทยและออกลูกเป็นตัว ฟองน้ำจากทั้งสองบริเวณมีรูปแบบการพัฒนาเซลล์สืบพันธุ์คล้ายกันโดยพบ spermatic cysts, oocytes, embryos และ ตัวอ่อน ในช่วงเดือนธันวาคม 2540 - เมษายน 2541 การเกิดปรากฏการณ์ปะการังฟอกขาวที่รุนแรงในอ่าวไทย ช่วงเดือนเมษายน - พฤษภาคม 2541 มีผลทำให้ไม่พบเซลล์สืบพันธุ์และตัวอ่อนในทุกตัวอย่างที่ศึกษาในช่วงเดือนพฤษภาคม - ตุลาคม 2541 ฟองน้ำจากบริเวณเกาะนกมีความดกของตัวอ่อนมากกว่าฟองน้ำจากเกาะค้างคาว จากการสังเกตในบ่อเพาะเลี้ยงพบว่าตัวอ่อน parenchymella จะถูกปล่อยในช่วงเวลาเช้า โดยตัวอ่อนส่วนใหญ่ลงเกาะบนก้อนปะการัง *Porites lutea* ที่มีชีวิต อัตราการเจริญเติบโตเฉลี่ยของฟองน้ำ *Petrosia* sp. ที่เกาะนก (0.7 ลูกบาศก์เซนติเมตรต่อวัน)

มากกว่าที่เกาะค้างคาว (0.6 ลูกบาศก์เซนติเมตรต่อวัน) อย่างมีนัยสำคัญทางสถิติ อัตราการตายเฉลี่ยของฟองน้ำ *Petrosia* sp. ที่เกาะนก (0.9 ลูกบาศก์เซนติเมตรต่อวัน) มากกว่าที่เกาะค้างคาว (0.8 ลูกบาศก์เซนติเมตรต่อวัน) อย่างมีนัยสำคัญทางสถิติ ผลการศึกษานี้นำไปสู่ความก้าวหน้าขององค์ความรู้เกี่ยวกับการดำรงชีวิตและการจัดจำแนกชนิดของฟองน้ำทะเล

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Saiprateep Asa

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CHAPTER 1

INTRODUCTION

Sponges can be found in all aquatic habitats and throughout all the world's ocean categorized into four main classes, i.e., Hexactinellida, Calcarea, Demospongiae and Sclerospongiae. Demospongiae have colonized from upper intertidal area to hadal zone. The ecological dominance of Demospongiae is the results of their diversity in form, structure, reproductive capabilities and physiological adaptation. However, biology of coral reef sponges in Southeast Asian countries is poorly known. Although reproductive processes are important criteria for the higher systematics of class Demospongiae, the reproductive biology of only few species have been studied in details (e.g. Reiswig 1983, 1; Fell 1983, 1; Ilan and Loya 1988, 745; Fromont and Bergquist 1994, 119; Tsurumi and Reiswig 1997, 1). Among various marine sponges studied there are several examples of gonochorism, successive hermaphrodism and contemporaneous hermaphrodism (Fell 1974, 67). Surprisingly, reproductive biology of *Petrosia* has been reported only once (Sara 1992, 6). This lack of basic knowledge on sponge reproduction occurs despite the fact that *Petrosia* are a common member of the benthos in coral reefs (Allen and Steene 1999, 38). Marine sponges contain various groups of microorganisms and provide many novel bioactive compounds, such as cytotoxic substances and antimicrobial activity. Chemicals isolated from sponges have been proved to be produced by symbiotic microorganisms (Lee et al. 1997, 103; Wattanadilok et al. 2000, 516). Some marine sponges were extracted bioactive compounds, such as cytotoxic activity against human tumor cell lines, anti HIV-1, antimicrobial activity and antibiofouling activity (Suwanborirux and Menasveta 1996, 35; Hattori et al. 1997, 104). However, natural products of sponges were used very quantity for extraction. The culture of marine sponges is a choice for producing natural products. Aquaculture of sponges, with the aim of producing metabolites, a farming method is required that promotes sponge growth and survival, and produces high yields of target metabolites (Duckworth et al. 1998, 155).

The present thesis provides data on reproductive ecology of *Petrosia* sp. on different substrates at Nok Island and Khang Khao Island in the Inner Gulf of Thailand. I concentrate on the studies of population density, distribution pattern, size versus maturity, reproductive pattern, fecundity, larval settlement, growth and mortality rates of this coral reef sponge.

Objectives

The major objectives of this study are as the following:

1. To investigate gamete development of a sponge, *Petrosia* sp. on coral communities in the Inner Gulf of Thailand.
2. To study onset and releasing time of *Petrosia* sp.
3. To apply the finding as fundamental data for management of living resources in coral communities.

Hypothesis

A sponge, *Petrosia* sp. is a viviparous gonochoric. Gametes and parenchymella larvae are developed in a brood chamber.

Scope of Research

This research concentrates on reproductive biology of a sponge, *Petrosia* sp. on coral communities at Nok Island and Khang Khao Island, in the Inner Gulf of Thailand. In addition, distribution patterns and ecology of *Petrosia* sp. are also studied in details in order to provide basic biological data for future aquaculture of sponge.

CHAPTER 2

LITERATURE REVIEW

Morphology

The sponges in submarine caves are differences in their morphological, pigmentary and spicules. Adaptations to darkness and precarious trophic condition have been studied in the Balearic Island (Bibiloni et al. 1989, 317). *Sycon* sp. morphogenesis and types of metamorphosis of *Halichondria panicea* have been studied in the Red Sea and Barentz Sea (Ilan et al. 1996, 133; Ivanova 1999, 262).

Physiology

The sponge can crawl up 160 µm/hr. on glass, plastic and rubber substrata by component cells (pinacocytes, mesohyl cells and choanocytes) as be studied in an aquarium (Bond 1988, 271; 1992,284). The marine sponge *Suberites domuncula* is induced of DNA damage and expression of heat shock protein HSP70 by polychlorinated biphenyls (Schröder et al. 1999, 285).

Reproduction

Marine sponges are recognized that there are several reproduction modes, i.e., gonochorism, successive hermaphrodism, and contemporaneous hermaphrodism. However, in *Oscarella lobularis* appears to be a contemporaeous hermaphrodism (Fell 1974, 67-68). A sponge, *Neofibularia nolitangere* is a gonochoristic oviparous as reported from Curacao (Hoppe 1988, 45). Four Caribbean commercial sponge species, *Hippospongia lachne, Spongia barbara, S. cheiris* and *S. graminea* are gonochorism (Kaye and Reiswig 1991, 3). *Erylus discophorus, Geodia cydonium, Stelletta grubii, Ancorina alata, Tetilla serica, Tetilla japonica, Tethya crypta, T. aurantium, T. citrina, Chondrosia reniformis, Polymastia hirsuta, Aaptos aaptos, Axinella damicornis, A. verrucosa, Agelas oroides* and *Hippospongia communis* are gonochorism (Sara 1992, 5-7). However, *Hymeniacidon sanguinea, H. heliophila*, *Polymastia mammillaris, Suberites massa* and *Octavella galangaui* are successive hermaphrodism (Sara 1992, 5-6). Both sponge species, *Spongilla lacustris* and *Ephydatia fluviatilis* are contemporaneous hermaphrodism (Leveaux 1941 cited by Sara 1992, 7). The sponge *Halisarca dujardini* was a viviparous as studied in the White Sea (Ereskovsky and Gonobobleva 1999, 60). *Tethya citrina* and *Tethya aurantium* are gonochoric oviparous while *Mycale contarenii* is a viviparous, according to the studies in the Mediterranean coastal area (Corriero et al. 1996, 175; Corriero et al. 1998, 319). Sex phenotypes of most species in Petrosida from the Great Barrier Reef are oviparity. However, many species of Haplosclerida are viviparity (Fromont 1994, 119; 1994, 307; 1994, 127; 1999, 185). Spermatogonia are probably derived from choanocytes. Oogonia has been reported to be derived from archeocytes and choanocytes. Fertilization has been reported in many cases. Internal fertilization is viviparity while external fertilization is oviparity (Harrison and De Vos 1991, 63-73). Selection for settlement on suitable habitats may be advantageous as it may result in low mortality rates after settlement. Larvae may be induced to settle by phototaxis (Uriz et al. 1998, 138). Larval settlement of *Halichondria* sp. was maximum at the time 50% of sponges contained large female reproductive elements (Fell et al. 1984, 127). Larvae of *Sigmadocia caerulea* settled mostly between 20 and 28 hours after releasing (Maldonado and Young 1999, 9). Gametes of calcareous sponges were developed from choanocytes while gametes of acalcareous sponges were developed from archaeocytes. However, spermatocyte was developed from choanocytes. Asexual reproduction of sponges were fragmentation, budding and gemmulation (Fell 1974, 71; 1983,7; 1993,2-5; Franzen 1996, 19). Sperms and *oocytes of Leucosolenia complicata were fertilized within the* choanoderm as were studied in the Barents Sea (Anakina 1999, 44), The hexactinellid sponge, *Oopsacas minuta* reproduced sexually all year round as reported in the Mediterranean Cave (Boury-Esnault and Vacelet 1994, 407). Free-swimming larvae of calcareous sponges exhibited metamorphosis according to the studies from Northern Japan (Amano and Hori 1992, 81; 1996, 161). Larvae of calcareous sponge such as *Clathrina* sp., *Sycon* sp. and *Leucosolenia* sp., are amphiblastula. However, larvae of demosponge, such as

Petrosia sp., *Haliclona amboinensis, Halisarca* sp. and *Tethya aurantium* are parenchymella (Bergquist 1987, 118; Barnes 1987, 88; Boury-Esnault and Rü tzler 1997, 16). Genetic structure of dictyoceratid and *Halisarca laxus* were clearly different among populations according to the studies by using allozyme techniques. At least, there were six polymorphic loci (Benzie et al. 1994, 335; Davis et al. 1996, 27). The organization of the eggs of *Tethya tenuisclera* and *Tethya seychellensis* were studied in two atolls of the Maldive Island (Gaino 1994, 99). *Niphates* sp. from tropical Indo-Pacific is a hermaphroditic brooder (Ilan and Loya 1988, 745). Larval settlement of a Red Sea sponge *Negombata magnifica* was reported (Ilan 1999, 248). Radiate accretive growth of new layers in two sponges *Haliclona oculata* and *H. simulans* were studies by iterative geometric construction method (Kaandorp 1994, 235). Sexual reproduction, reproductive cycles, spermatogenesis, oogenesis, larval behavior, settlement and metamorphosis in four sponges, i.e., *Hippospongia lachne, Spongia barbara, S. cheiris* and *S. graninea* in the Caribbean were reported (Kaye and Reiswig 1991, 1; 1991, 25; Kaye 1991, 13). The sponge *Microciona prolifera* showed reproductive activity in the explants appeared to be normal at 20% salinity as reported at Long Island Sound in Connecticut (Knight and Fell 1987, 253). Larval types of sponges, *Halisarca dujardini, H. metschnikova* and *Tethya aurantium* were also the parenchymella (Levi 1957, 180). Larval activities and settlements of *Halisarca tubifera* and *Halichondria magniconulosa* were observed that their swimming speeds decreased in low temperature (Maldonado and Young 1996, 169). Most larvae of *Crambe crambe* and *Scopalina lophyropoda* settled preferentially in the microrefuges of the grooved plates (Maldonado and Uriz 1998, 141). Larval type of *Sycon* sp. was an amphiblastula (Meglitsh et al. 1991, 64). The sponge *Mycale fistulifera* reproductively provided gametes throughout the year. Sponge larvae are capable of fast settlement and metamorphosis in minutes to 30 hours after releasing. Sexual maturity occurred at an early age (Meroz and Ilan 1995, 443). A sponge *Suberites domuncula*'s primary cells were cultured (Müller et al. 1999, 205). Sponges, *Verongia gigantea*, *Mycale* sp. and *Tethya crypta* clearly exhibited the relationship between onset of gametes and water temperature (Reiswig 1970, 538; 1973, 208). Spermatozoa of *Sycon calcaravis* was transformed to spermiocysts in a choanocyte before fertilization, as detected by using EM (Nakamura et al. 1998, 179). A sponge *Aiolochroia crassa* released spermatozoa into seawater at 26 m. in depth on the coral reef at Discovery Bay, Jamaica (Reiswig 1983, 15). The modes of reproduction of two marine sponges, *Tethya aurantium* and *T. citrina* were effected to genetic variability that were studied in the Mediterranean Sea (Sara et al. 1989, 293) The sponges in family Petrosiidae were oviparity (Sara 1992, 7; 1992, 52). A sponge *Halichondria panicea* was gonochorism. Oogenesis started in late summer/early autumn (Witte and Barthel 1994, 297). Parenchymellae of *Haliclona tubifera* were strongly photonegative on releasing and remained throughout the duration of larval existence (Woollacott 1993, 301) Asexual reproduction by fragmentation of massive sponges, *Jaspis stellifera* and *Rhopaloeides odorabile* showed high rates of success in transplantation in the Great Barrier Reef (Wilkinson and Thompson 1997, 1417). The sponge

Xestospongia cf. *testudinaria* was synchronised spawning gametes from osculum at 09.00 hours on 29 August 1989 in the Banda Sea, Indonesia (Sarano 1991, 166). Larvae of *Spongilla lacustris* were able for regeneration (Seminov et al. 1999, 568). A sponge *Leucosolenia* sp. had a parenchymella larva (Sherman et al. 1976, 53). A sponge *Adocia varia* was a viviparous gonochoristic at Adriatic Sea, Italy (Simpson 1984). A sponge *Halichondria panicea* had maximum specific growth rate of 4% d^{-1}. The net growth efficiency was about 30% (Thomassen and Riigård 1995, 239). A sponge *Aplysina cauliformis* was oviparity at Barbados (Tsurumi and Reiswig 1997, 1). A sponge *Crambe crambe* reproduced in summer and larvae released during July-August as reported from the Coast of Spain (Uriz et al. 1995, 159; Turon et al. 1999, 617). Stages in spermatogenesis and oogenesis of sponge *Alectona wallichii* and *A. mesatlantica* sp. nov. were presented at the same time as embryos in various stages of development at Oahu, Hawii (Vacelet 1999, 627). Two carrier cells of *Sycon calcaravis* were pushed the spermiocyst to the surface of the oocytes (Wanatabe and Okada 1998, 197). *Tedania ignis* had a parenchymella larva composed of two types of cells, peripheral and flagellated epithelium-like (Weyrer et al. 1999, 660). Recruitment of sponge varied seasonally in the Carribean Sea (Zea 1993, 1). Boring sponges bored into a coral *Porites furecata* in the Caribbean Sea (Wulff 1984, 160). The sponges, *Iotrochota birotulata, Amphimedon rubens* and *A. fulva* were asexual reproduced by fragmentation (Wulff 1991, 227; 1995, 55; 1997, 1377). The influence of temperature on the kinetics of allograft reactions in sponge was obviously recognized (Johnton et al. 1981, 280). Secretion of new spicules in demosponge *Pellina semitubulosa* may be connected with the process of remodeling occurring after gamete and larval releasing, according to the reports from the Mediterranean area Coastal (Mercurio et al. 2000, 87).

Taxonomy

New discovery of the genus, *Strepsichordaia* and two new species, *Strepsichordaia lendenfeldi Carteriospongia contorta* are clearly known. Descriptions of species in the Petrosida and Haplosclerida have been illustrated in the Great Barrier Reef (Bergquist et al. 1988, 291; Fromont 1991 73-96; 1993, 7-40; 1995, 149-180; Fromont et al. 1994, 735; De Laubenfels 1954, 52-98; Hooper 1997, 101-106). Genus Petrosia, Xestospongia and Strongylophora are the sponges which be grouped into the family Petrosiidae (Kelly-Borges and Pomponi 1992, 29; Hooper 1994, 73). Sponges, *Plakina monolopha, P. dilopha* and *P. trilopha* were identified by allozyme electrophoresis in the Western Mediterranean (Murricy et al. 1996, 181). Phenotyppic variation of *Aplysina fistularis* and *A. cauliformis* were tested by a histocompatibility bioassay by grafting (Neigel and Schmahl 1984, 413). A sponge *Microciona prolifera* was categorized into the order Demospongiae (Pierce and Maugel 1987, 44). The microscleres of *Acanthochaetetes wellsi* is bigger than those of *A. seunesi* (Reitner and Engeser 1987, 13). Three morphotypes of *Suberites ficus* were isolated by isozyme loci assay (Solé-

Cava and Thorpe 1986, 247). A sponge *Petrosia muta* is a synnonym with *Xestospongia muta* (Wiedenmayer 1977, 115).

Ecology

In the Mediterranean Coastal Lagoon, distribution and density of sponges varied on substrata, water movement, sedimentation and food (Corriero et al. 1989, 303). Salinity was a factor which influenced dormancy of a sponge *Microciona prolifera* in the laboratory culture (Fell et al. 1989, 195). Retention efficiencies, pumping and respiration rates in *Mycale acerata* and *Isodictya kerguelensis* were reported at King George Island, Antarctic (Kowalke 2000, 85). Ecological significance of microhabitat-scale movement in an encrusting sponge *Scopalina lophyropoda* was clearly known (Maldonado and Uriz 1999, 239). Distribution of sponges, *Suberites carnosus, S. ficus* and *Thenea muricata* was a good example for general conclusion (Pansini and Musso 1991, 317). Most populations of the sponges, *Cliona aprica* and *C. lampa* grew on a coral *Acropora cervicornis* in Jamaica (Pang 1973, 227). Two sponges, *Mycale laevis* and *Niphates erecta*, exhibited chemical defense products in the Caribbean (Pawlik 1997, 1257). Biodiversity of sponge varied according to depth of the habitats (Reed and Pomponi 1997, 1387). Insufficient photosynthetic radiation limited the growth of the sponge population below 30 m. depth as many of the species were phototrophic with a dependence on cyanobacterial symbionts for nutrition which were studied in the Great Barrier Reef (Wilkinson and Evans 1989, 1; Wilkinson and

Cheshire 1989, 127; 1990, 285). Marine sponges, *Tethya microstella, T. orphei, T. robusta* and *T. seychellensis* grew on the rubble of dead corals. There were also several studies in a Maldivian coral reef lagoon and all around the world (Sara et al. 1993, 341; Sara 1997, 1384). The giant barrel sponge, *Xestospongia muta*, restored from damaging at Florida Keys (Schmahl 1999, 532). Rates for photosynthesis and respiration of *Cymbastela concentrica* decreased from shallow to deep at Davis Reef, Great Barrier Reef (Seddon et al. 1992, 847). A brittle star, *Ophiothrix fragilis*, recruited on the sponge *Crambe crambe, Scopalina lophyropoda* and *Dysidea avara* in Mediterranean (Turon et al. 2000, 201). The sponges distributed along a light gradient at Balearic Island, Mediterranean Sea (Uriz et al. 1992, 106). The influence of temperature on the kinetics of allograft reactions in a sponge was reported (Johnton et al. 1981, 280).

Symbiosis

Symbiosis between sponges and other organisms are obviously recognized. The calcareous sponge *Clathrina cerebrum*/ gram-negative elongate bacteria is an example. The locations of symbionts are in choanoderm. The sponge *Halichondria panicea* has it symbiont bacteria *Rhodobacter* sp. The sponge *Aplysina gerardogreeni* was symbiosis with cyanobacteria as studied in Lingurian Sea, Adriatic Sea, Eastern Tropical Pacific and Coastal Florida (Burlando et al. 1988, 35; Althoff 1998, 529; Diaz 1997, 1399; Hill 1996, 650). A marine sponge, *Petrosia* sp. were found

Propionibacterium, *Aeromonas* sp. and *Bacillus*-like as its symbionts (Lee et al. 1997, 103). Two sponges, *Ircinia strobilina* and *I. felix* had bacteria counterparts (Pile 1997, 1403). Populations of the symbiotic association between the sponge, *Haliclona cymiformis* and the red macroalga, *Ceratodictyon spongiosum* were studied in One Tree Lagoon, Great Barrier Reef (Trautman 1999, 602; Trautman et al. 2000, 87).

Interactions

There are many cases of sponge and organism interactions. The sponge *Desmapsamma anchorata* frequently overgrowth the coral *Diploria strigosa*. Peripheral contact of the sponge, *Rhaphidophlus venosus*, with the coral, *Montastrea cavernosa*, tissue contact of the sponge, *Niphates erecta*, with the coral, *M. cavernosa*. Successful overgrowth of corals by sponges depends on coral cover and sponge species composition as have been studied in Santa Marta Coastal and Caribbean Reefs (Aerts et al. 1997, 127; Aerts 1998, 241). *Callyspongia diffusa* allograft reactions with a coral, *Montipora verucosa* tests in aquaria in Hawaii were reported (Johnston et al. 1981, 280). An epizoic zoanthidean grew on the surface of sponge *Calcifibrospongia actinostromarioides* in Caribbean Sea (Willenz and Hartman 1994, 279). A sponge, *Stelletinopsis dominicana*, were fed by Hawksbill turtles at Mona Island, Puerto Rico (Van Dam and Diez 1997, 1421). A sponge *Xestospongia muta* was fed by hermit crab in the Caribbean Sea (Waddell and Pawlik 2000, 125; 2000, 133).

Bioerosion

The skeleton of sponges, *Agelas clathrodes, Ectyoplasia ferox* and *Xestospongia muta* did not provide a defense against predatory reef fish. However, *Xestospongia muta* can protect itself by the chemical defense. (Chanas and Pawlik 1996, 225; 1997, 1363). Clionid sponges were the principal bioeroders of coral reefs as reported in the west coast of Barbados (Holmes 1997, 1411). A bioeroding sponge *Cliona lampa* bore in living corals, *Porites astreoides* and *Montastrea cavernosa* at Burmuda (Mckenna 1997, 1369).

Natural Products

Agelasine compound from the marine sponge *Agelas mauritiana* was extracted as an antifouling substance against macroalgae (Hattori 1997, 411). A marine sponge *Halichondria okadai* were isolated Halichondrin B compound for preclinical candidates (Schmitz 1994, 486). A marine sponge *Reniera sarai* provided 3-alkylpiridinium oligomers (Sepcic et al. 1997, 991). A marine sponge *Polyfibrospongeia australis* also provided polyfibrospongols A and B (Shen and Hsieh 1997, 93).

Aquaculture

Marine sponges, *Latrunculia brevis* and *Polymastia croceus* were successfully with aquaculture in the good farming system as shown in temperate water of New Zealand (Duckworth et al. 1999, 155). Cultivation of marine sponges, *Spongilla* sp. was successful *in situ*. However, *Cliona celata* and *Pseudosuberites andrewsi* were cultivated *in vitro* (Osinga et al. 1998, 133; Osinga et al. 1999, 419).

CHAPTER 3

MATERIALS AND METHODS

Study Site

One of the study areas was located at Nok Island, situated geographically east of the Pattaya shoreline in the Gulf of Thailand (13° 01 10"N, 100° 49' 25"E; Figure 1). The corals around the island are mostly composed of species of *Porites, Acropora, Pavona, Styllocoeniella, Turbinaria, Favia, Fungia, Lobophyllia, Pocillopora, Goniopora* and *Millepora*. The northern, western and southern sides of the island are characterized by rocky shores and are directly exposed to the southwest monsoon. The protected eastern side contains a small bay with a rubble and sand beach (Chou et al. 1991, 296-301). The other study area was located in the Khang Khao Island, inner part of the Gulf of Thailand (13° 06' 45"N, 100° 48' 52"E; Figure 2). Khang Khao Island does not have a developed reef flat while scleractinian corals mostly occur in the shallow waters, 3 m. below LLW. The corals around the island are mostly composed of species of *Porites, Pavona, Pocillopora, Acropora, Favia, Favites, Platygyra, Fungia, Montipora and Galaxea. Porites lutea* was the most abundant species (Sakai et al. 1986, 28-31; Yamazato and Yeemin 1986, 166; Yeemin 1999, 8). All sides of Khang Khao Island are influenced by the differentiation of monsoons. The northern side of the island which is directly effected by the Northeast

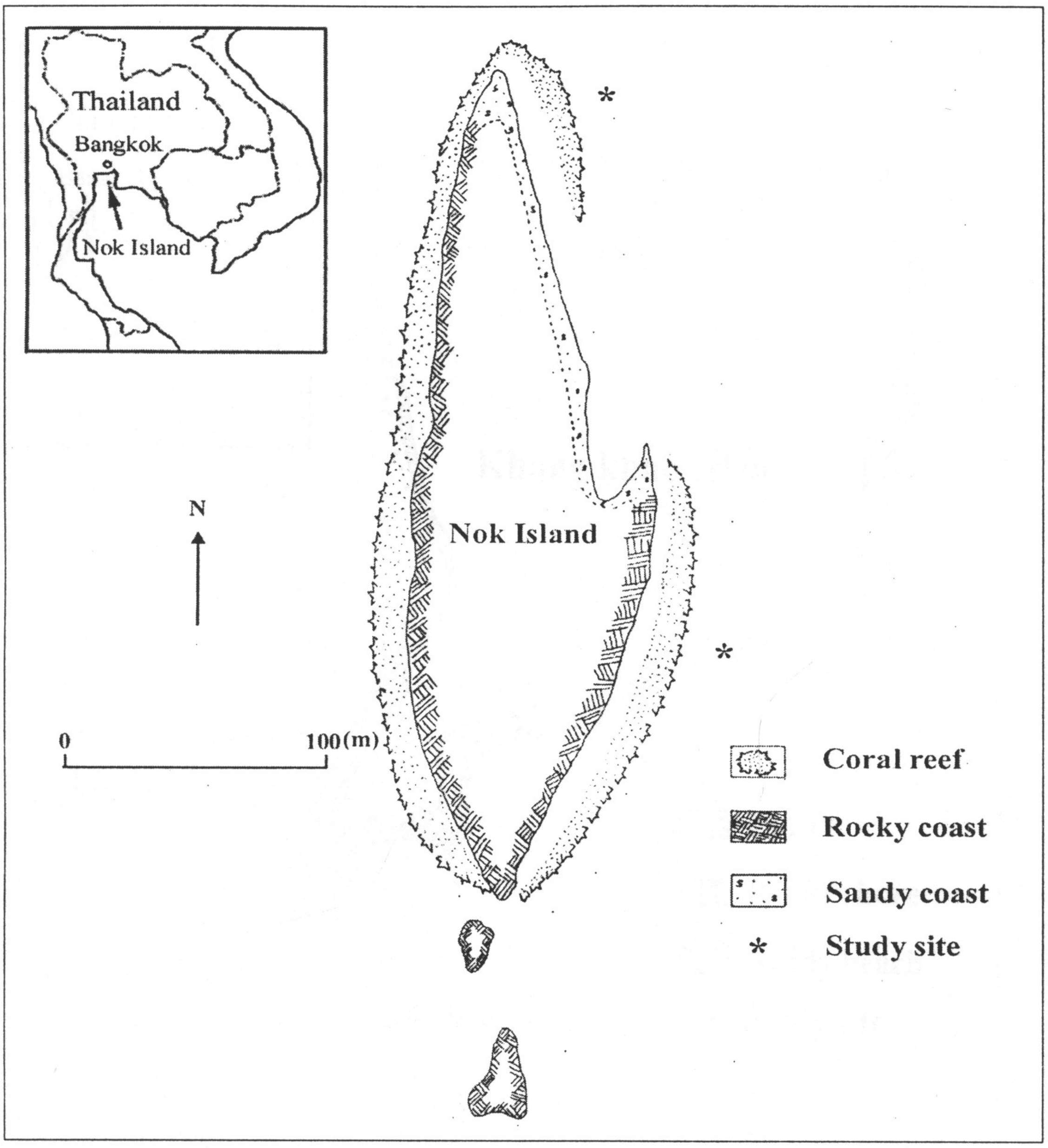

Figure 1 Map of Nok Island Showing the Study Sites

Source: Chao, L. M., S. Sudara, V. Manthachitra, R. Moredee, A. Snidvongs, and T. Yeemin. 1991. Temporal variation in a coral reef community at Pattaya Bay, Gulf of Thailand. **Environmental Monitoring and Assessment** 19: 295-307.

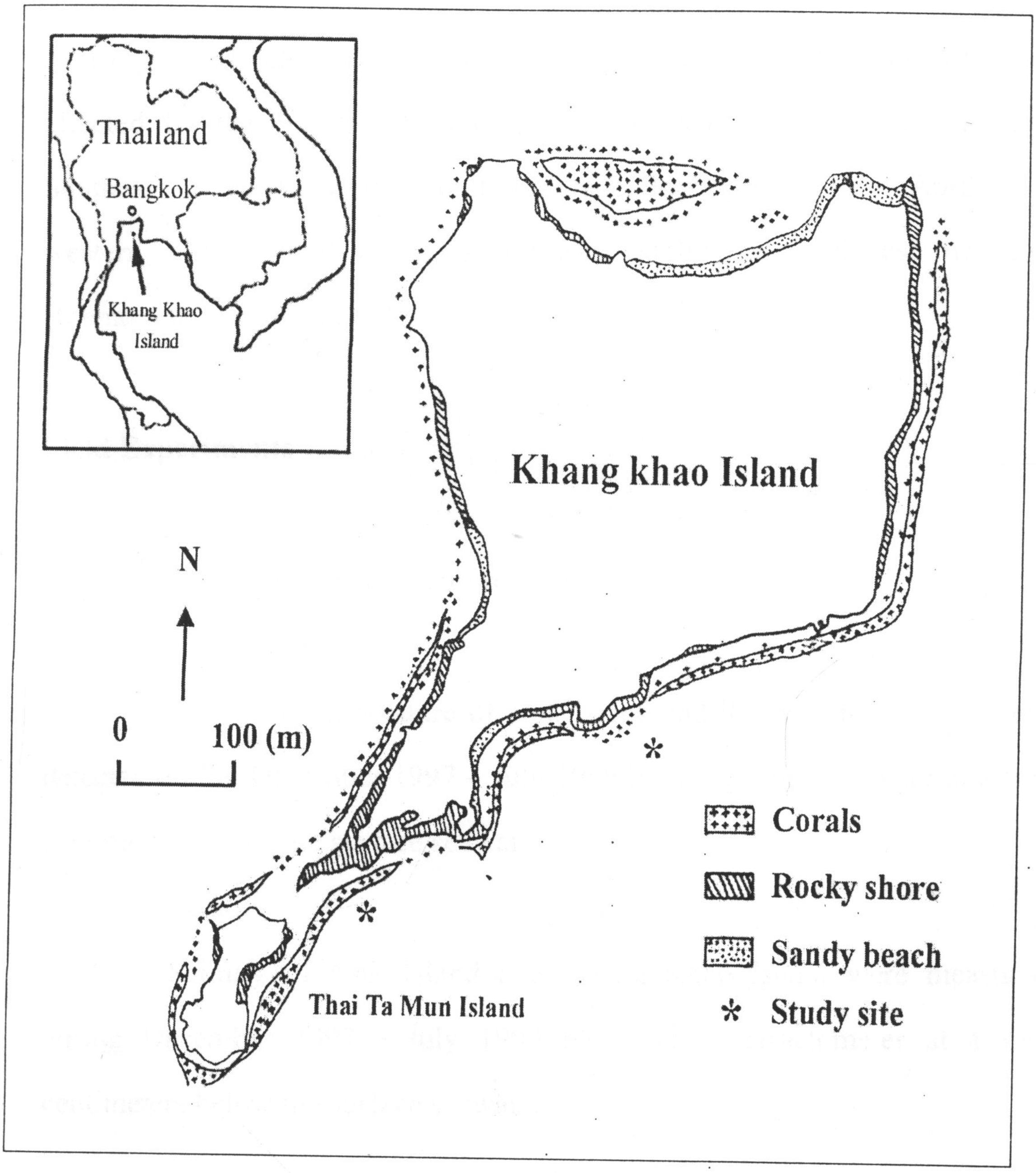

Figure 2 Map of Khang Khao Island Showing the Study Sites

Source: Yamazato, K., and T. Yeemin. 1986. Preliminary study on the inter- and intra-specific interaction among corals of Khang Khao Island, the Sichang Island, Gulf of Thailand. **Galaxea** 5: 163-174.

monsoon during October - February. The eastern side of the island which is effected by the Southwest monsoon during May – September and the Southeast monsoon or the Southeast trade during February – April. The western of the island which is also effected by the Southwest monsoon (Menasveta et al. 1986, 8-9).

Field Experiments

Measurement of Environmental Factors

1. Temperature

Seawater temperature of Nok Island and Khang Khao Island were detected during December 1997 – July 1999 by using a thermometer at a few centimeters below the surface seawater.

2. Salinity

Salinity of Nok Island and Khang Khao Island were measured during December 1997 - July 1999 by using a refractometer at a few centimeters below the surface seawater.

Population Density and Distribution Pattern

Population densities of *Petrosia* sp. on coral communities of Nok Island and Khang Khao Island were determined in August, 1998. Thirty random 1x1 m quadrats were placed in coral zone of the two islands and the number of *Petrosia* sp. in these quadrats were recorded (Figure 3-4). The finding was used to analyze mean population densities and distribution

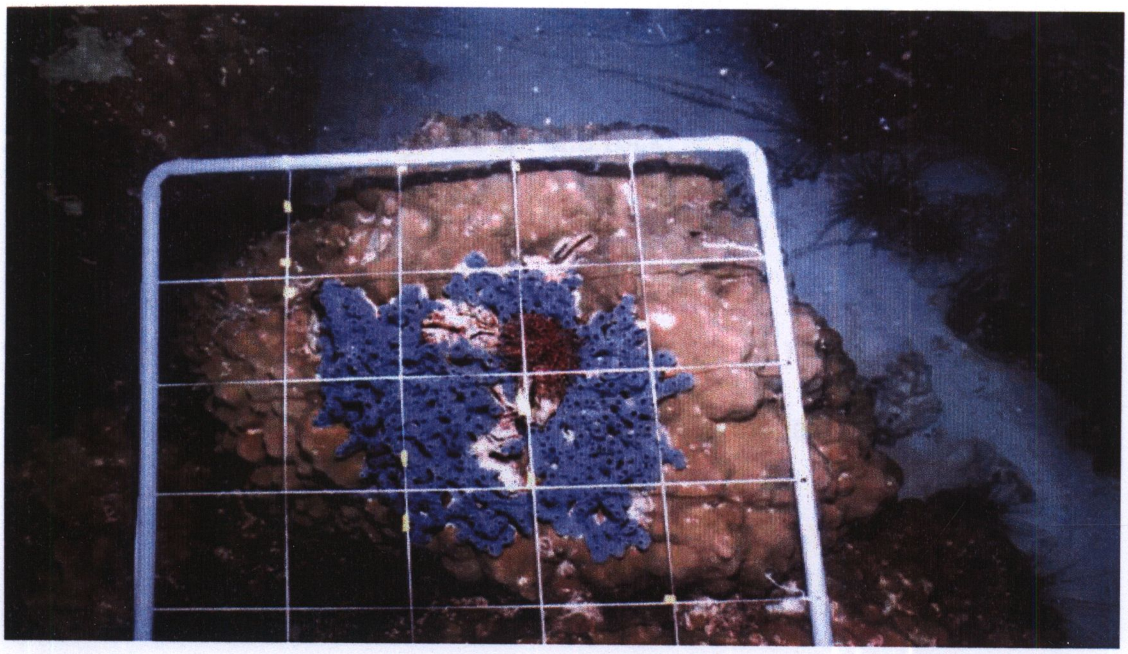

Figure 3 Random Quadrat was Used to Examine Population Density and Distribution Pattern of *Petrosia* sp. at Nok Island.

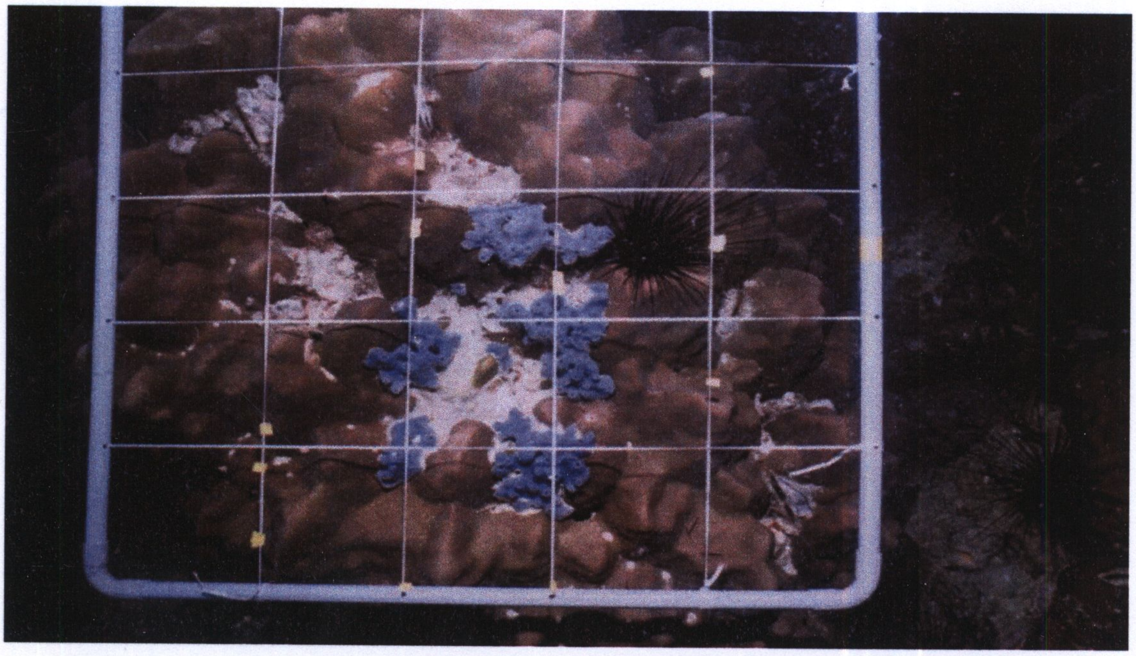

Figure 4 Random Quadrat was Used to Examine Population Density and Distribution Pattern of *Petrosia* sp. at Khang Khao Island.

patterns of *Petrosia* sp. on three types of substrate, i.e., a massive coral *Porites lutea,* a zoanthid *Palythoa* sp. and rock.

Size versus Maturity

Sizes of *Petrosia* sp. on coral communities of Nok Island and Khang Khao Island were determined in the parenchymella larval peak during December 1998-January 1999. Sizes of *Petrosia* sp. were measured each individuals and observed larval present (Figure 5-6).

Gamete Development

Gamete development from December 1997 to March 1999 was studied. Reproductive activities of *Petrosia* sp. were monitored by repetitive sampling of the marked individuals from the coral zones of Nok Island and Khang Khao Island. Each month pieces of 1-2 oscules were cut from the sponge individuals with a sharp dissection knife. The samples were then fixed in 10% formalin-sea water and transferred to 70% ethanol (Figure 7). The samples were embedded in paraffin, sectioned at 8-10 µm thickness and stained with haematoxylin-eosin (Yeemin 1999, 5; Figure 8-10). In addition, presence of parenchymella larvae in brood chambers was also examined directly in the field during December 1997- June 2000. The finding was used to analyze seasonal sexual reproduction of *Petrosia* sp.

Figure 5 Size of *Petrosia* sp. was Measured Directly Underwater.

Figure 6 Parenchymella Larvae Contained in an Individual of *Petrosia* sp.

Figure 7 Preserved Samples in 70% Ethanol Which were Used for Histological Studies.

Figure 8 Samples of *Petrosia* sp. were Embedded in the Paraffin.

Figure 9 Samples of *Petrosia* sp. were Sectioned by a Microtome.

Figure 10 Tissue of *Petrosia* sp. on the Slides were Stained with Haematoxylin and Eosin.

Fecundity of Parenchymella Larvae

Fecundities of parenchymella larvae in *Petrosia* sp. on three types of substrate, i.e., *Porites lutea, Palythoa* sp. and rock were monitored by sampling during a peak of the individuals containing parenchymella larvae from coral communities at Nok Island and Khang Khao Island. The samples of *Petrosia* sp. were analyzed in the laboratory.

Larval Releasing and Settlement

Releasing and Settlement of parenchymella larvae of *Petrosia* sp. were tested on six types of substrate, i.e., *Porites lutea*, dead coral, *Palythoa* sp., rock, gypsum plate and gypsum plate+sediment in a aquaria.

Vegetative Growth

From November 1998 to May 1999 growth rates of *Petrosia* sp. on three types of substrate, i.e., *Porites lutea, Palythoa* sp. and rock were monitored by repetitive measurement of the marked individuals from the coral zones of Nok Island and Khang Khao Island (Figure 11).

Mortality

From November 1998 to May 1999 mortality rates of *Petrosia* sp. on three types of substrate, i.e., *Porites lutea, Palythoa* sp. and rock were monitored by repetitive measurement of the marked individuals from the coral zones of Nok Island and Khang Khao Island (Figure 12).

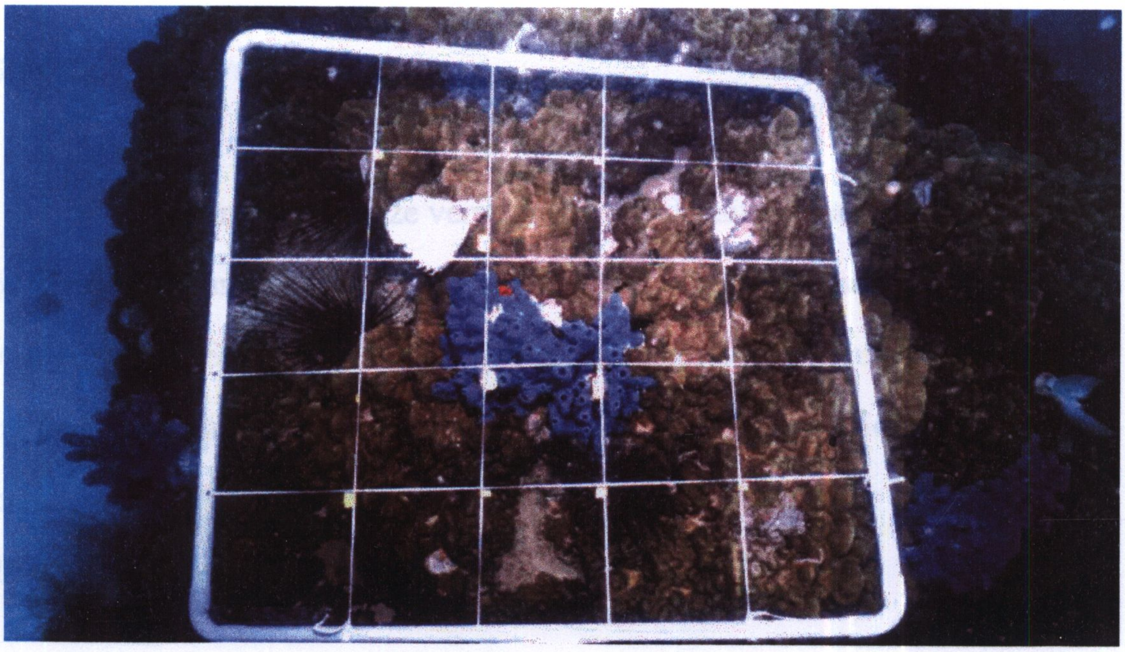

Figure 11 A Marked Individual of *Petrosia* sp. for Growth Rate Studies at Nok Island

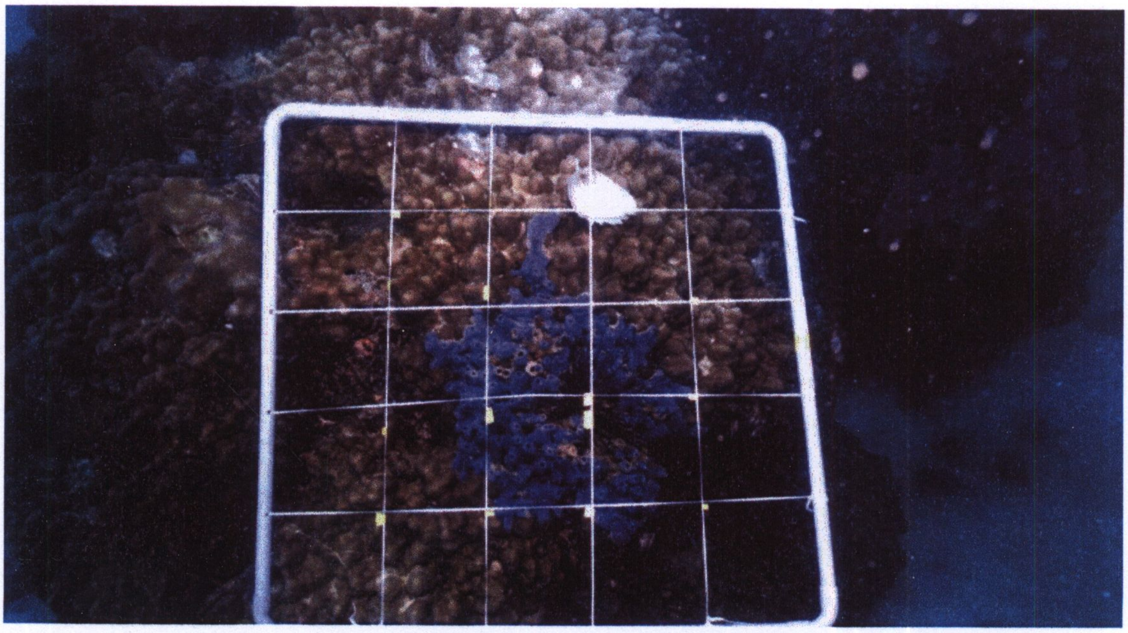

Figure 12 A Marked Individual of *Petrosia* sp. was Used to Examine Partial Mortality Rate at Nok Island.

Statistical Analysis

Population density, size versus maturity, density of gametes, diameter of gametes, fecundity, growth rate and mortality rate were analyzed by two way ANOVA (Sokal and Rohlf 1987, 186). Dispersion pattern was analyzed by variance/mean ratio (Ludwig and Reynolds 1988, 26-28). Settlement of parenchymella larvae were analyzed by one way ANOVA (Zar 1984, 163-164).

CHAPTER 4

RESULTS

Environmental Factors

Temperature

The seawater temperatures of Nok Island and Khang Khao Island during May – June, 1998 (32 - 32.5 °C) were higher than those during May – June, 1999 (Figure 13). Moreover, the first severe event of occurrence of coral bleaching in the Gulf of Thailand in May – June, 1998 was undoubtably resulted from abnormal increasing of seawater temperature during May – June 1998. These results are supported by the data of the NOAA (National Oceanic and Atmospheric Administration) (Figure 15-16). The sponge, *Petrosia* sp. was also effected by the bleaching phenomenon (Figure 17-18).

Salinity

The results in Figure 14 showed that the average salinity of Nok Island (32.8 ± 0.7 ppt.) was slightly higher than that of Khang Khao Island (32.3 ± 0.7 ppt.).

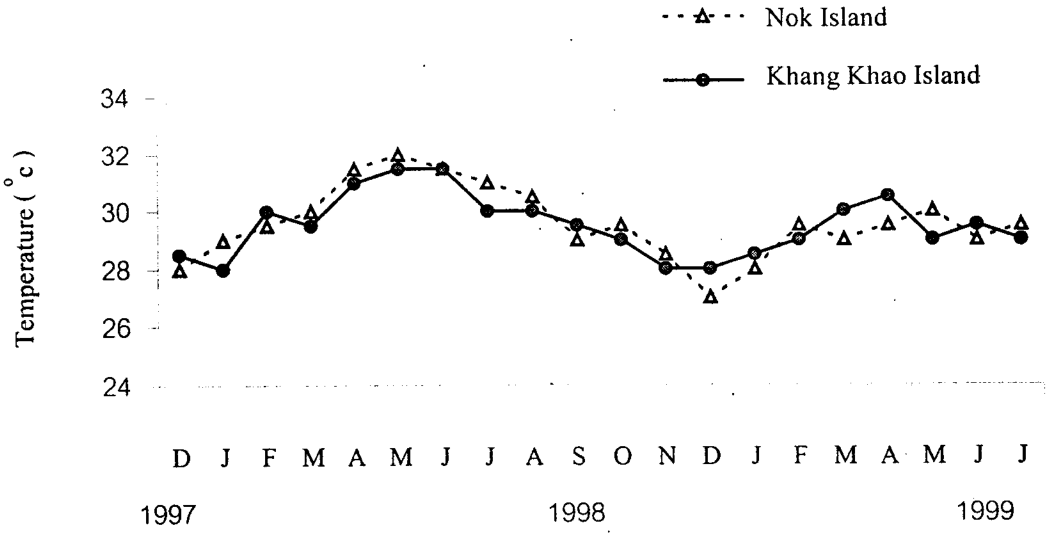

Figure 13 Seawater Temperature at Nok Island and Khang Khao Island during December 1997 - July 1999

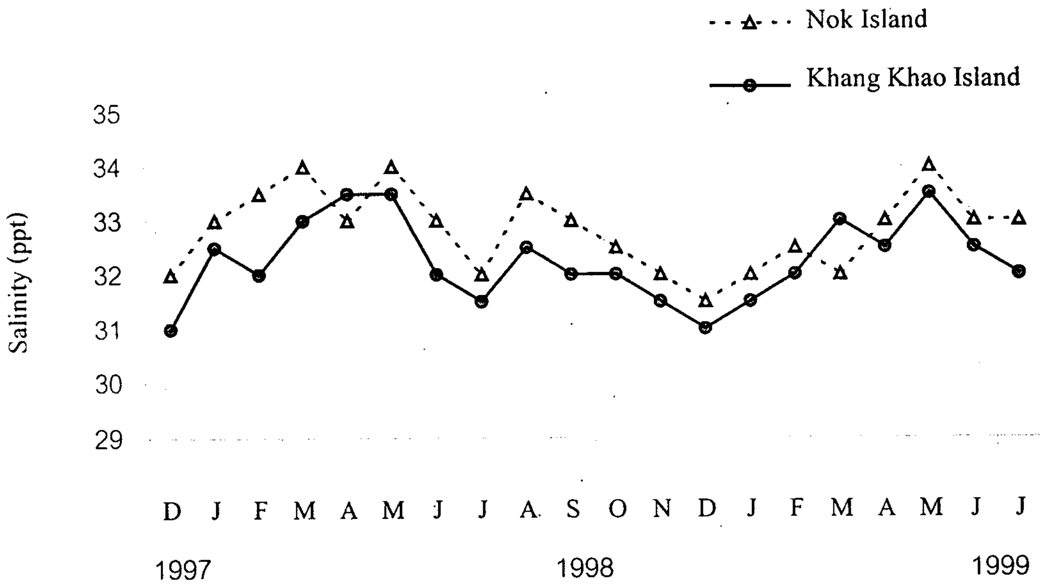

Figure 14 Salinity of Seawater at Nok Island and Khang Khao Island during December 1997 - July 1999

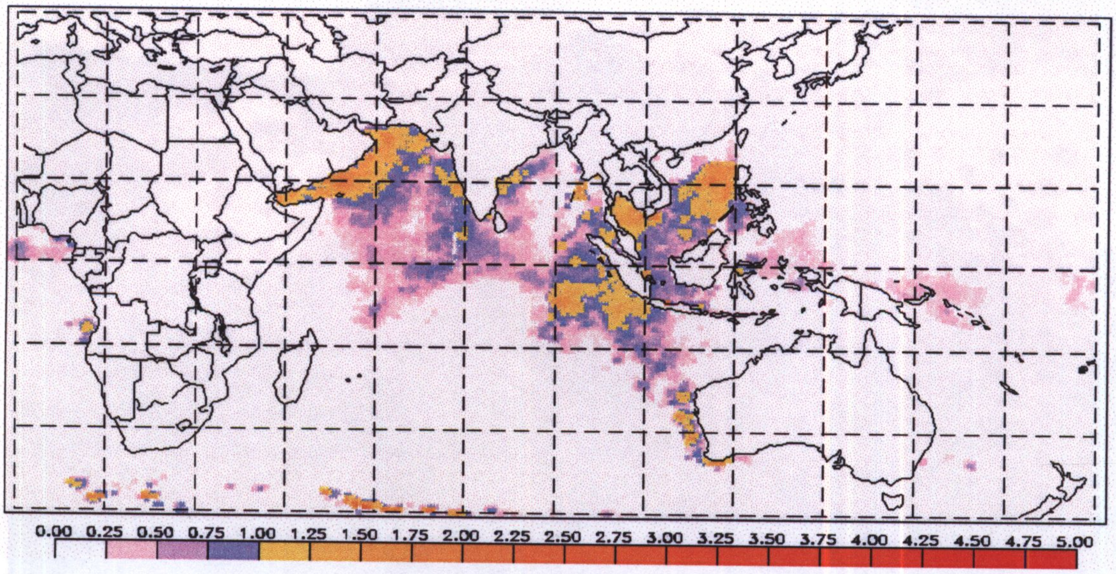

Figure 15 Report of Sea Surface Temperature in May 1998 from NOAA (A Color Table is the Degree of Temperature (°C) that Exceed the Maximum Monthly Climatology in Each Region.)

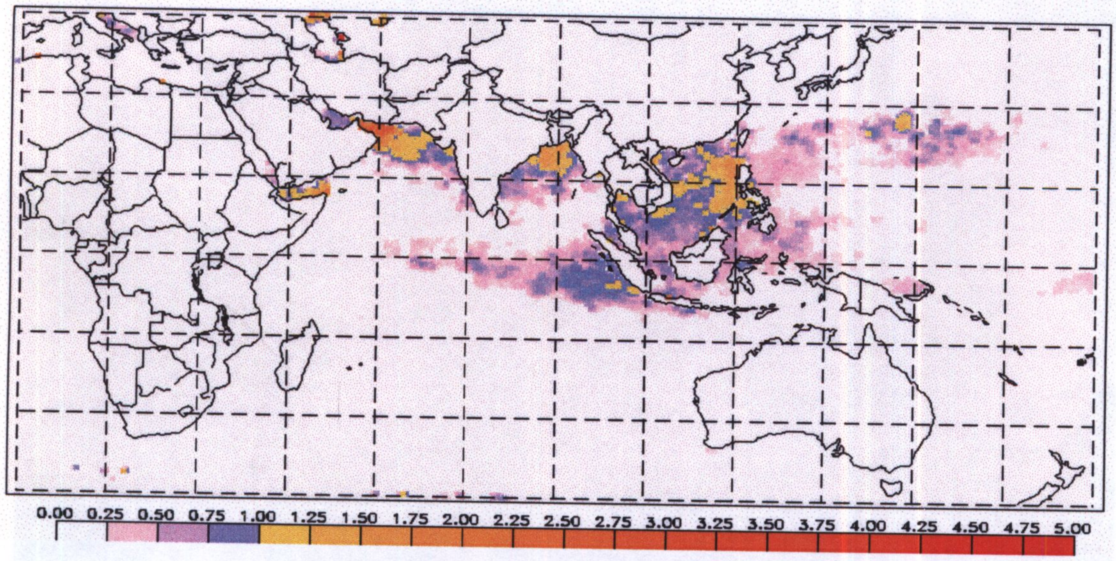

Figure 16 Report of Sea Surface Temperature in June 1998 from NOAA (A Color Table is the Degree of Temperature (°C) that Exceed the Maximum Monthly Climatology in Each Region.)

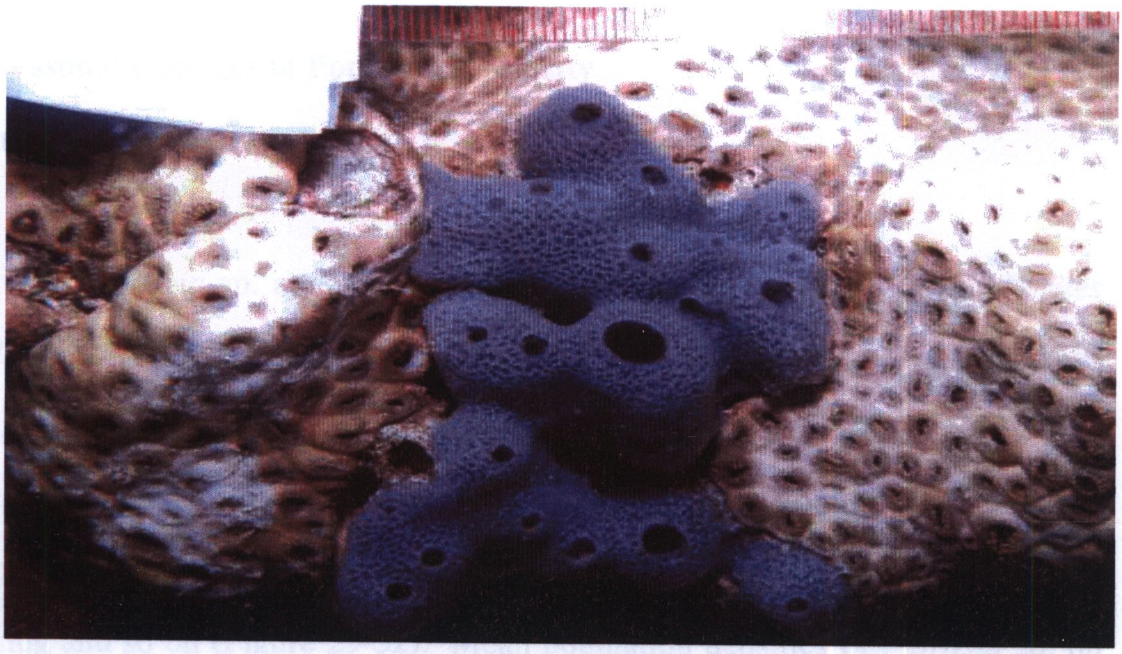

Figure 17 A Sponge, *Petrosia sp.* was Affected by the Severe Coral Bleaching Phenomenon at Nok Island.

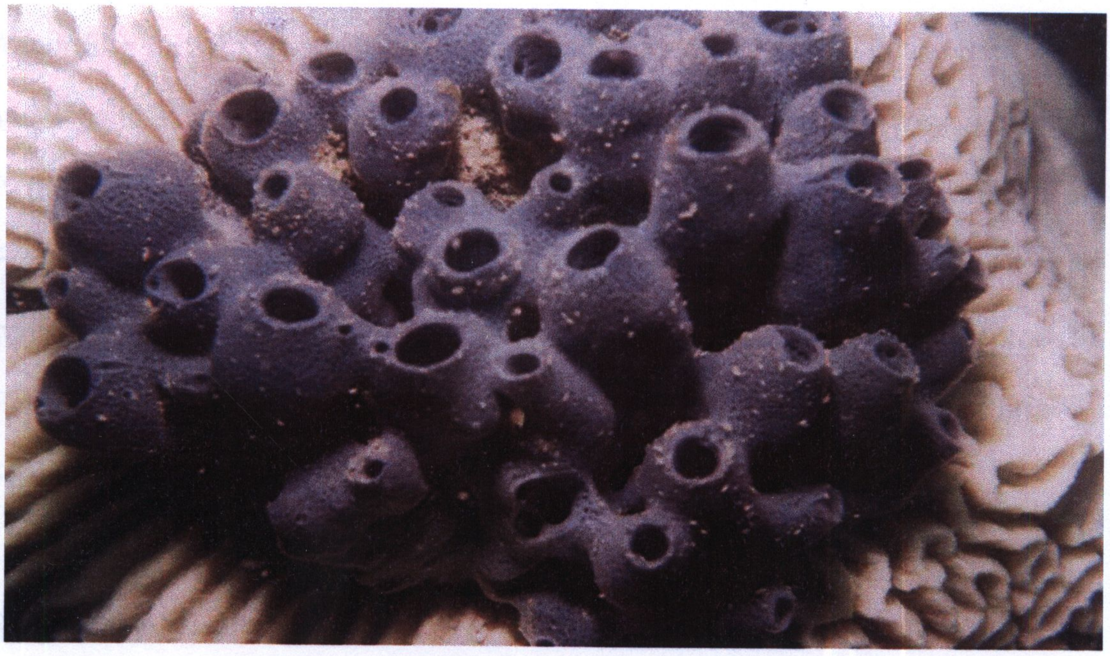

Figure 18 A Sponge, *Petrosia* sp. was Affected by the Severe Coral Bleaching Phenomenon at Khang Khao Island.

Seasonal Changes in Population Density and Distribution Pattern

Petrosia sp. attached mostly on three types of substrate, i.e., *Porites lutea* , *Palythoa* sp. and rock (Figure 19-24). In addition, *Petrosia* sp. also grew on many types of substrate, e.g. *Porites* sp., *Goniopora* sp., *Pavona decussata* and dead coral (Figure 25-28) in the shallow zones (2-5 m in depth) of the coral communities. Natural enemies of *Petrosia* sp. were sea urchin, sea slug and so on (Figure 29-32). Mean population densities of *Petrosia* sp. on *Porites lutea*, *Palythoa* sp. and rock at Nok Island were 0.37±0.03, 0.13±0.02 and 0.07±0.02 colonies/m^2, respectively while those at Khang Khao Island were 0.20±0.05, 0.10±0.03 and 0.07±0.02 colonies/m^2, respectively. According to the statistical analysis, mean population densities at Nok Island and Khang Khao Island were not statistically different (Two-way ANOVA, $P>0.05$, Figure 33). However, mean population densities of *Petrosia* sp. on *Porites lutea* were significantly higher than those on *Palythoa* sp. and rock substrate (Two-way ANOVA, $P<0.05$, Figure 33). Dispersion patterns of *Petrosia* sp. in all studied locations were clumped (Variance/Mean Ratio>1, Figure 34).

Size versus Maturity

The body size of *Petrosia* sp. at Nok Island (average body size, 416 cm^3) was significantly higher than that at Khang Khao Island (average body

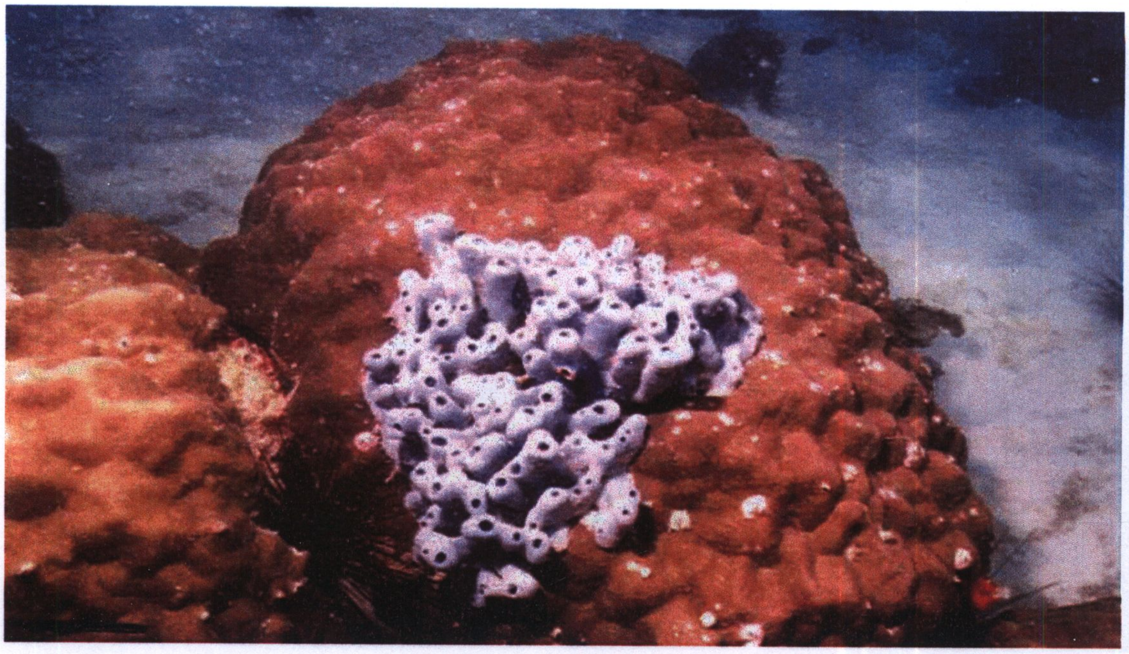

Figure 19 *Petrosia* sp. Attached on a Massive Coral, *Porites lutea* at Nok Island

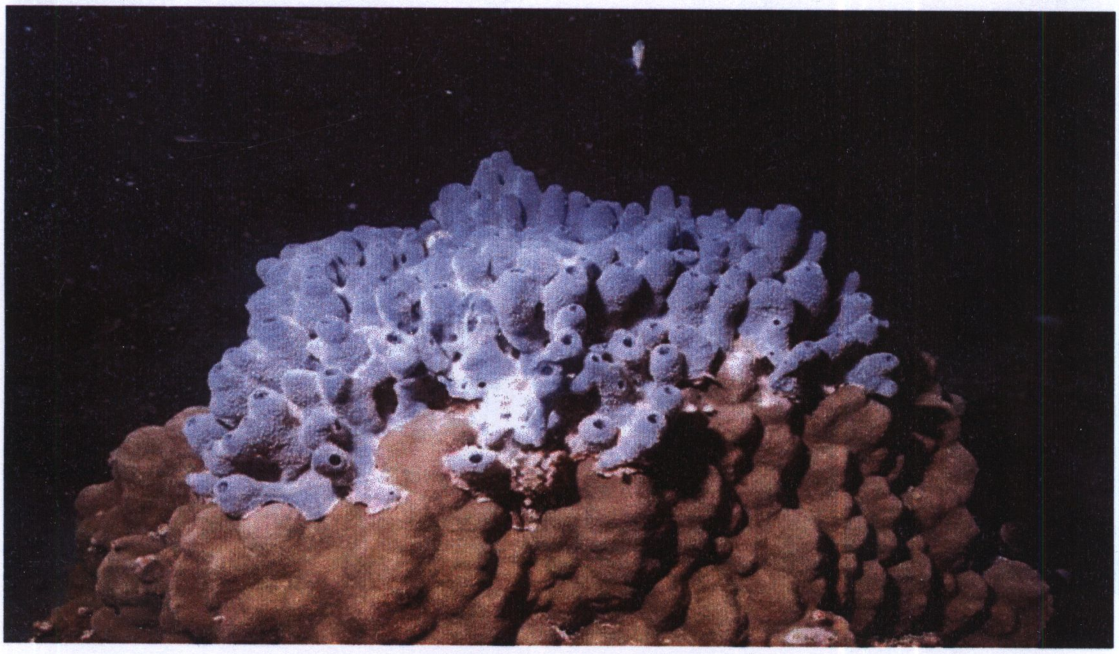

Figure 20 *Petrosia* sp. Attached on a Massive Coral, *Porites lutea* at Khang Khao Island

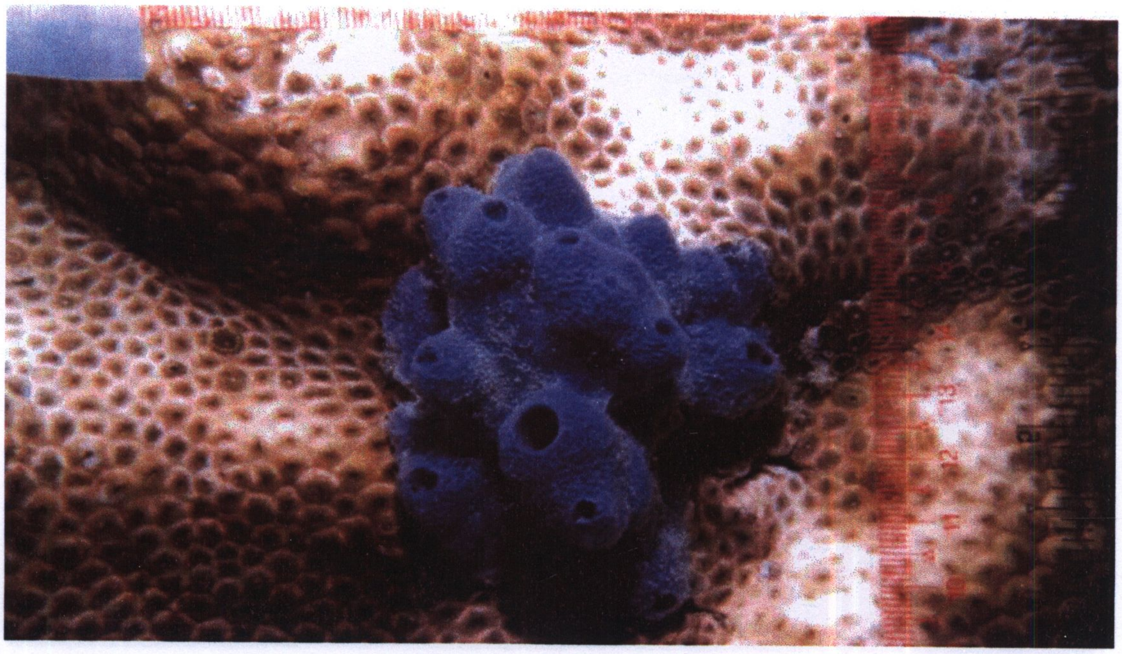

Figure 21 *Petrosia* sp. Attached on a Zoanthid, *Palythoa* sp. at Nok Island

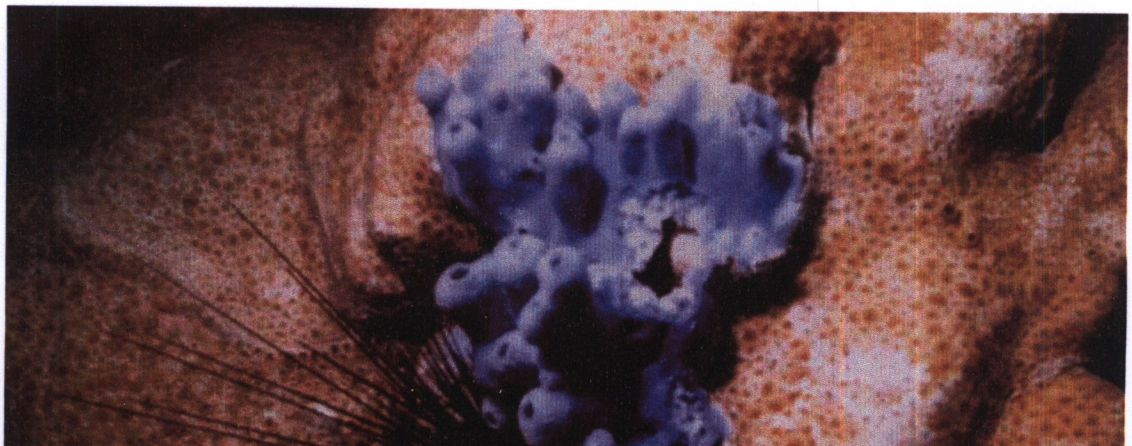

Figure 22 *Petrosia* sp. Attached on a Zoanthid, *Palythoa* sp. at Khang Khao Island

Figure 23 *Petrosia* sp. Attached on Rock Substrate at Nok Island

Figure 24 *Petrosia* sp. Attached on Rock Substrate at Khang Khao Island

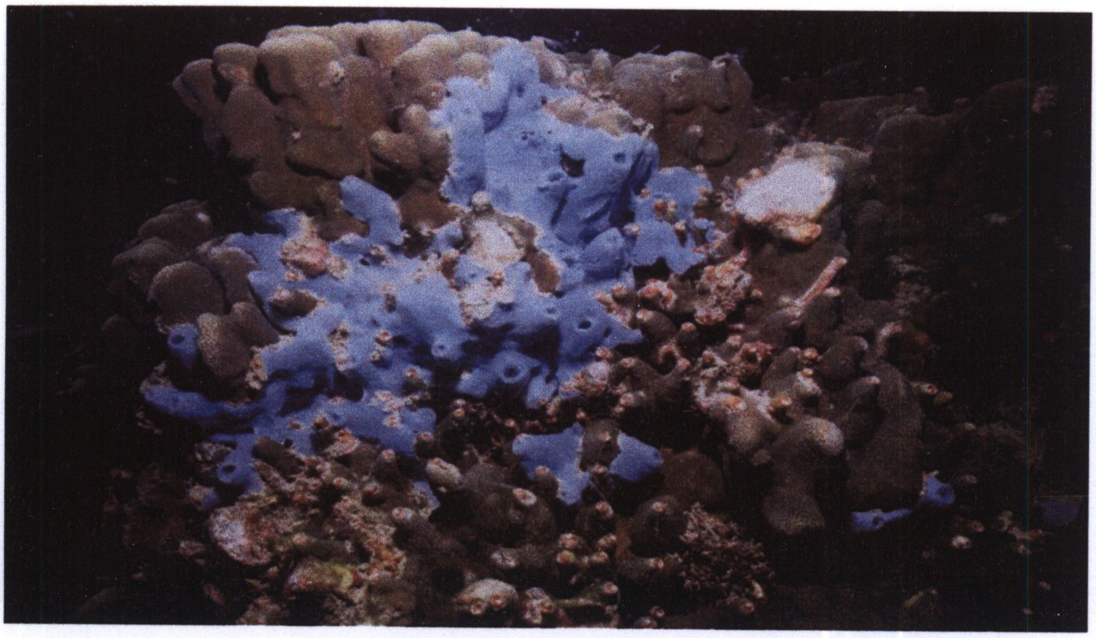

Figure 25 *Petrosia* sp. Attached on a Coral, *Porites* sp.

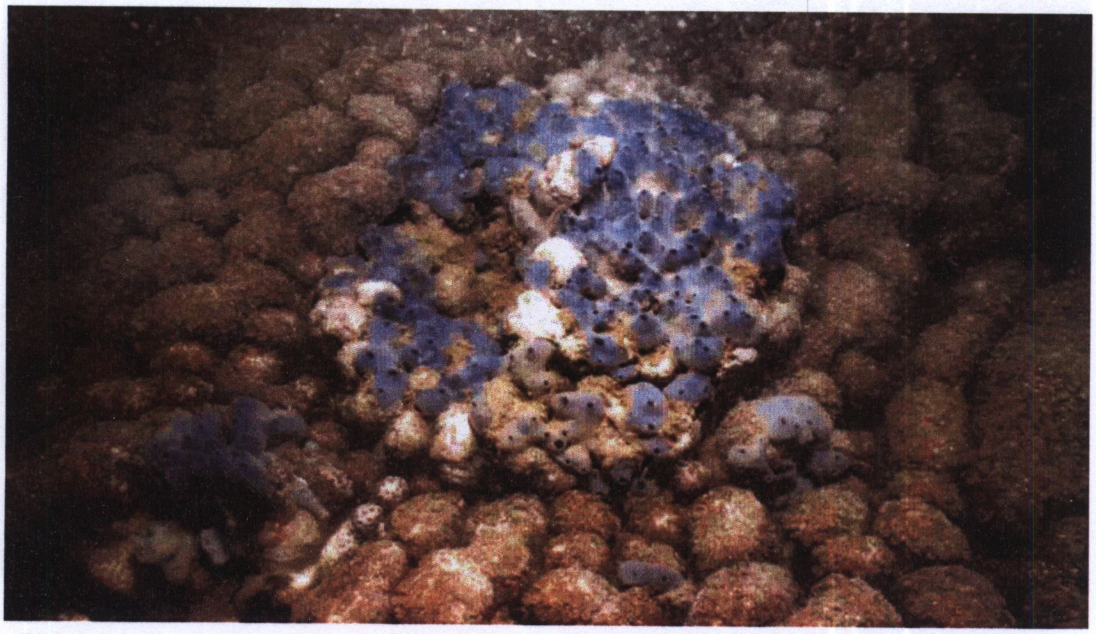

Figure 26 *Petrosia* sp. Attached on a Coral, *Goniopora* sp.

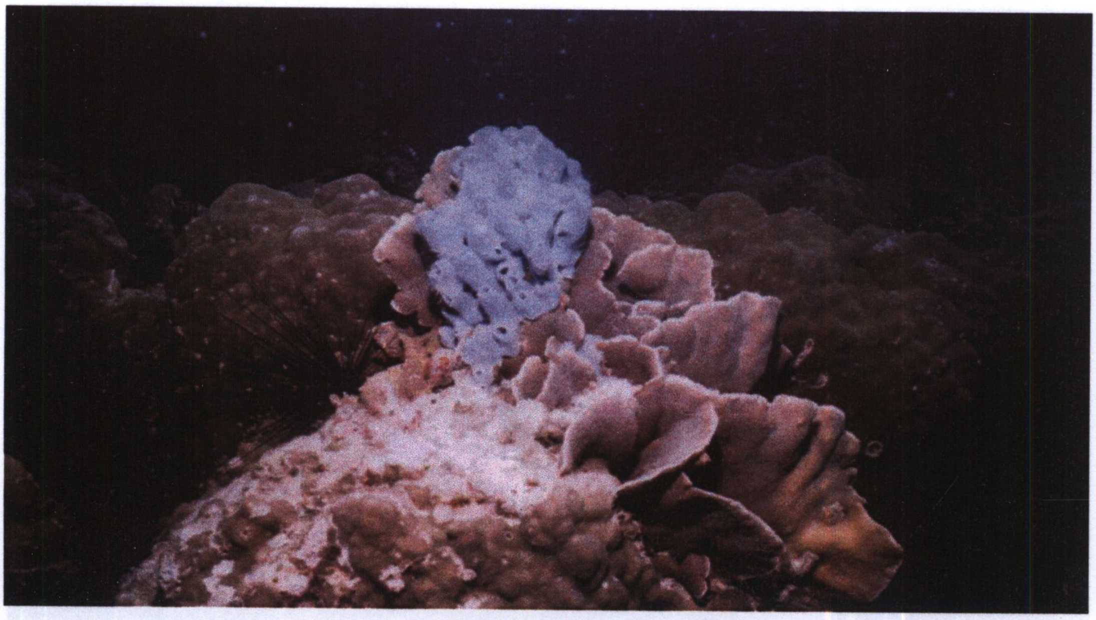

Figure 27 *Petrosia* sp. Attached on a Coral, *Pavona decussata*

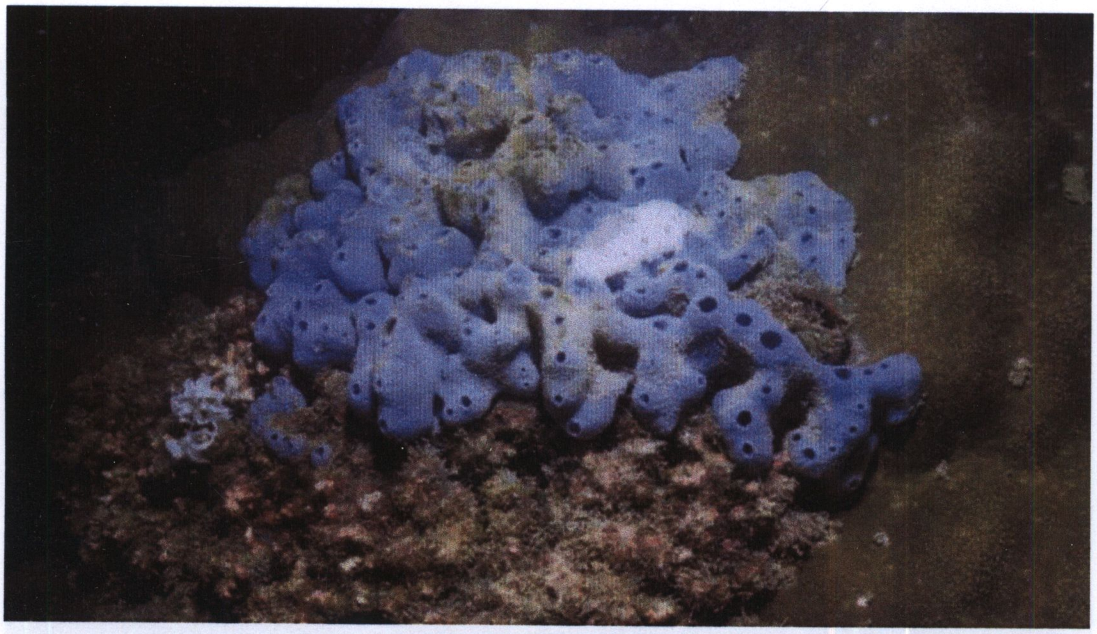

Figure 28 *Petrosia* sp. Attached on a Dead Coral Colony

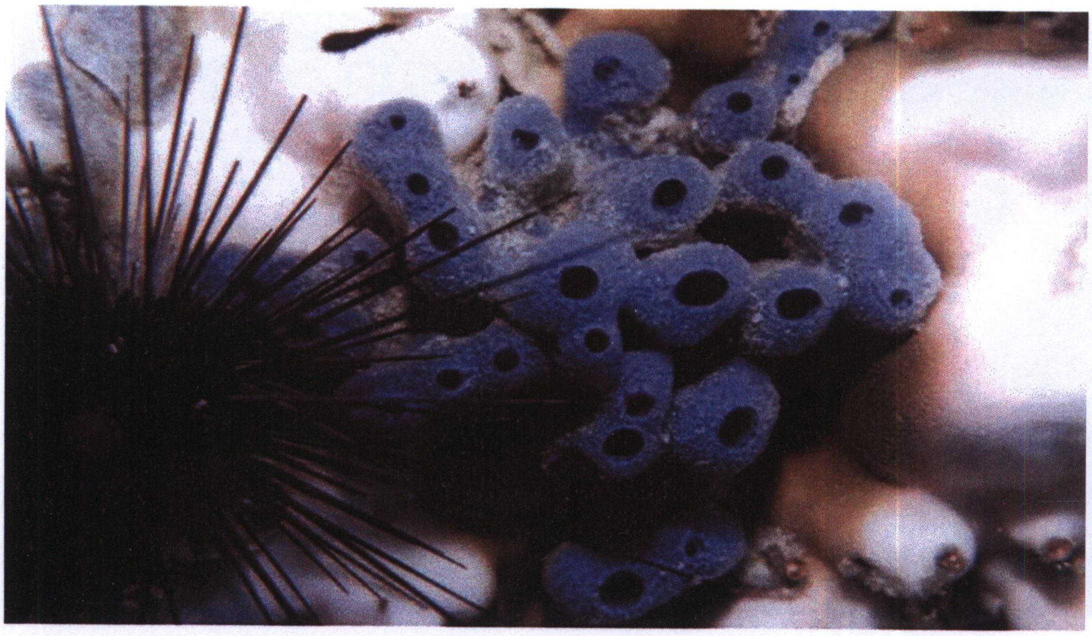

Figure 29 A Sponge, *Petrosia* sp. was Grazed by a Sea Urchin, *Diadema setosum*.

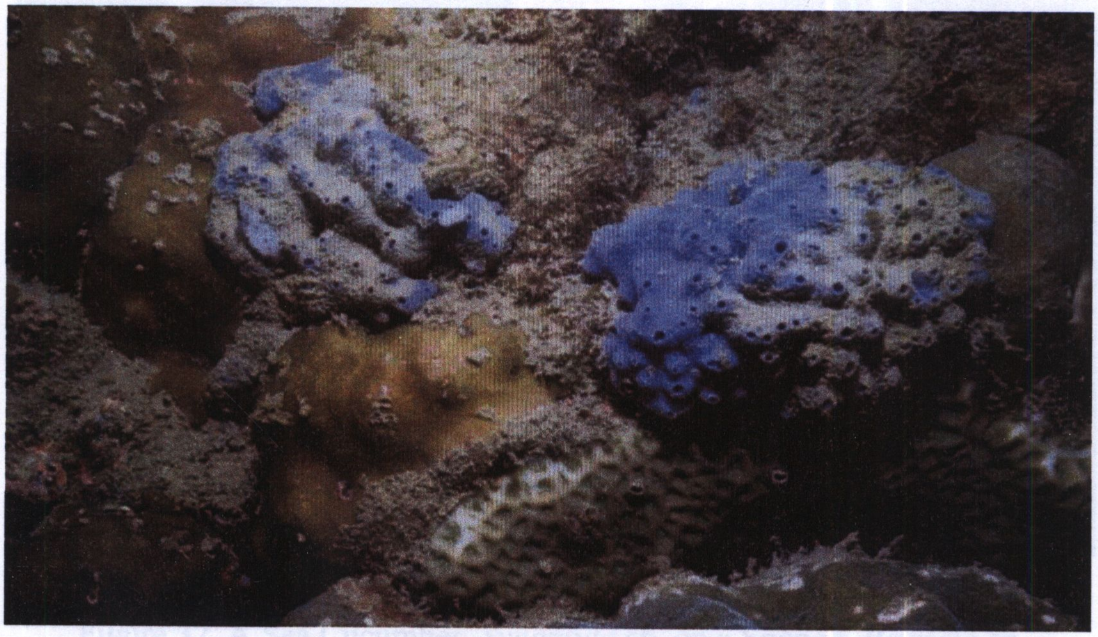

Figure 30 Algae Covered on a Sponge, *Petrosia* sp.

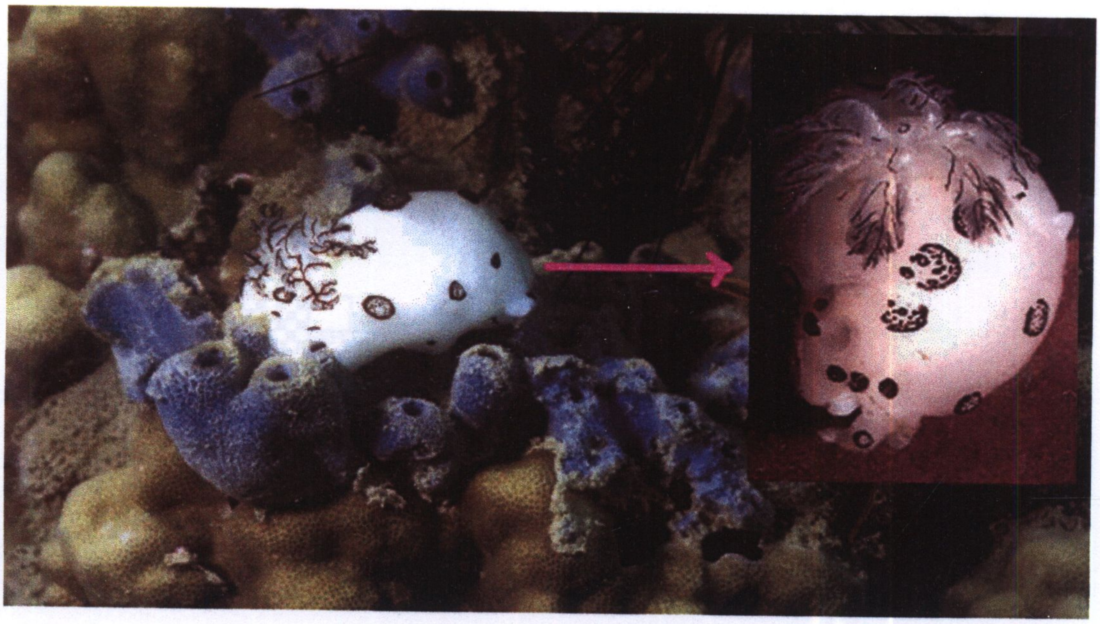

Figure 31 A Sea Slug, *Jorunna funebris* was a Predator of *Petrosia* sp.

Figure 32 A Sea Cucumber, *Synaptula* sp. on a Sponge, *Petrosia* sp.

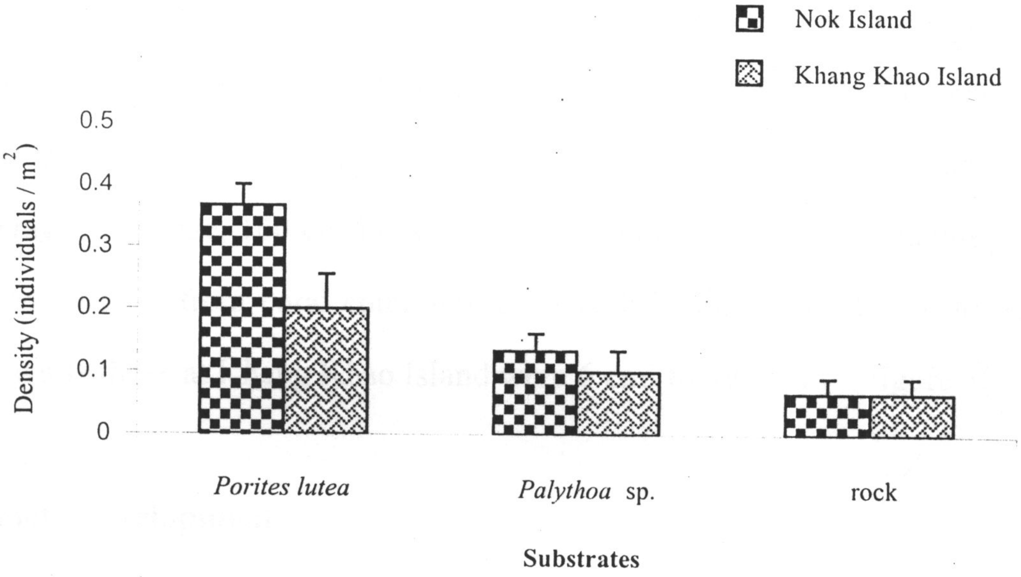

Figure 33 Mean Population Densities of *Petrosia* sp. on Different Types of Substrate at Nok Island and Khang Khao Island

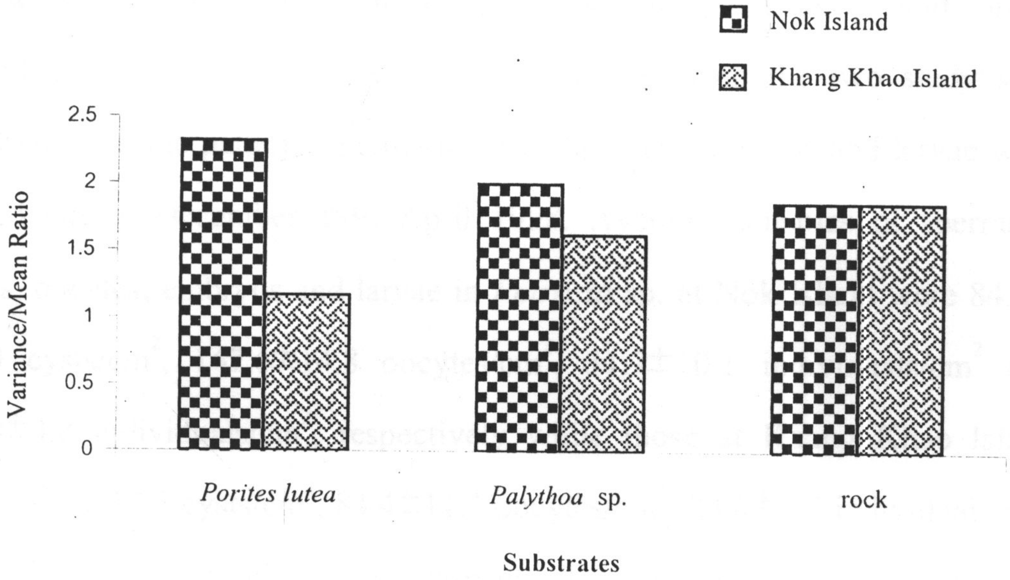

Figure 34 Dispersion Patterns of *Petrosia* sp. on Different Types of Substrate at Nok Island and Khang Khao Island

size, 222 cm^3) (Two-way ANOVA, $P<0.05$). While those sizes of *Petrosia* sp. on *Porites lutea* (average body size, 482 cm^3) was significantly higher than those on *Palythoa* sp. (average body size, 315 cm^3) and on rock substrates (average body size, 160 cm^3) (Two-way ANOVA, $P<0.05$). The mature sizes of *Petrosia* sp. from coral communities at Nok Island were 50 cm^3 to 3500 cm^3 while those at Khang Khao Island were 6 cm^3 to 3920 cm^3 (Figure 35).

Gamete Development

Petrosia sp. was a viviparous hermaphrodite. Spermatic cysts and oocytes were clearly observed in the same specimen. Oocytes, embryos and larvae were developed asynchronously within choanoderm and brood chambers. Gamete development of *Petrosia* sp. from the two study sites showed the same pattern. Spermatic cysts, oocytes, embryos and larvae were found during December 1997-April 1998. Average densities of spermatic cysts, oocytes, embryos and larvae in *Petrosia* sp. at Nok Island were 84.6± 28.0 cysts/cm^2, 147.6±14.8 oocytes/cm^2, 31.7±10.1 individuals/cm^2 and 22.0±4.8 individuals/cm^2, respectively while those at Khang Khao Island were 67.8±12.3 cysts/cm^2, 84.4±11.3 oocytes/cm^2, 23.0±5.7 individuals/cm^2 and 17.0±5.4 individuals/cm^2, respectively. According to the statistical analysis, average densities of gametes (spermatic cysts, oocytes, embryos and larvae) at Nok Island and Khang Khao Island were not statistically different (Two-way ANOVA, $P>0.05$. Figure 36). Mean diameters of spermatic cyst, oocyte, embryo and larva at Nok Island were 14.5 ± 3.8 μm, 18.1 ± 7.6 μm,

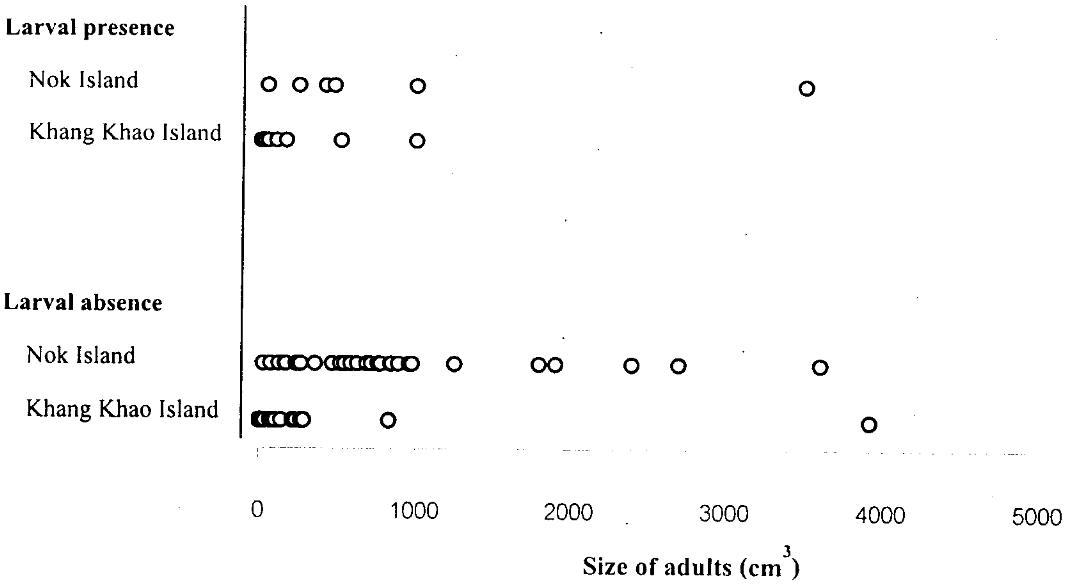

Figure 35 Size versus Maturity in *Petrosia* sp. by Presence or Absence of Parenchymella Larvae in Individuals at Nok Island and Khang Khao Island

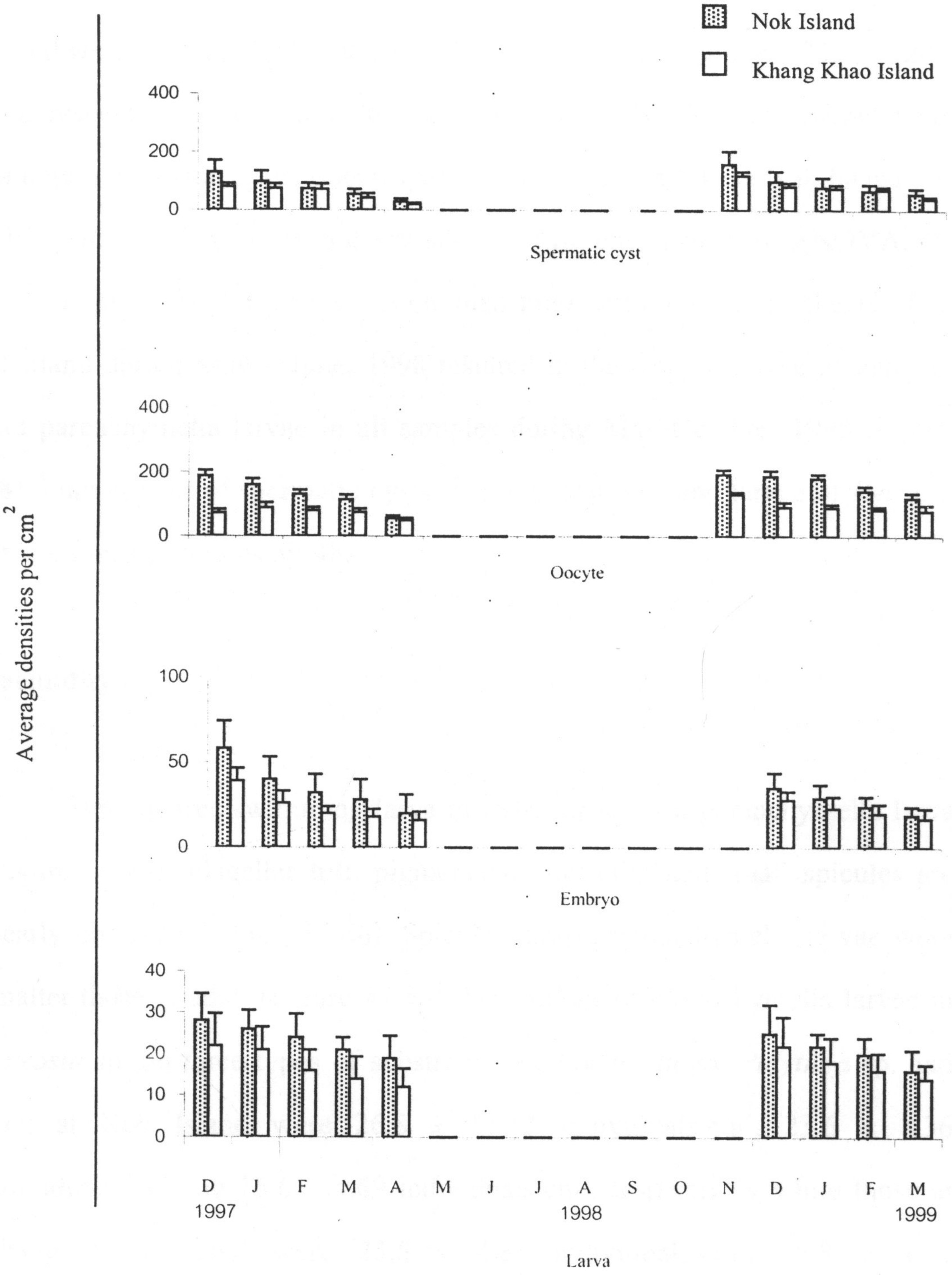

Figure 36 Gamete Development of *Petrosia* sp. at Nok Island and Khang Khao Island During December 1997 – March 1999

173.9 ± 9.4 µm and 241.9 ± 9.2 µm, respectively while those at Khang Khao Island were 11.2 ± 4.7 µm, 29.3 ± 12.1 µm, 217.6 ± 12.6 µm and 232.3 ± 20.0 µm, respectively. According to the statistical analysis, mean diameters of gametes (spermatic cysts, oocytes, embryos and larvae) of the Nok Island and Khang Khao Island were not statistically different (Two-way ANOVA, $P> 0.05$, Figure 37). The severe coral bleaching phenomenon in the Gulf of Thailand during May – June, 1998 resulted in the disappearance of gametes and parenchymella larvae in all samples during May-October, 1998 (Figure 38). Photographs of spermatic cysts, oocytes, embryos and larvae of *Petrosia* sp. are given in figures 39-48.

Fecundity

Type of free-swimming larva in *Petrosia* sp. is a parenchymella larva (Figure 51-54). Flagellar tuft, pigmentary ring, cilia and small spicules are clearly observed (Figure 55-56). Spicule sizes in parenchymella larvae were smaller those in adult (Figure 57-58). Fecundities of parenchymella larvae in *Petrosia* sp. on three types of substrate, i.e., *Porites lutea, Palythoa* sp. and rock at Nok Island were 26.8 ± 11.12 individuals/cm^2, 23.6 ± 8.36 individuals/cm^2 and 16.6 ± 4.69 individuals/cm^2, respectively while those at Khang Khao Island were 23.5 ± 4.81 individuals/cm^2, 15.8 ± 4.99 individuals/cm^2 and 14.3 ± 3.54 individuals/cm^2, respectively. According to the statistical analysis, the fecundities of parenchymella larvae in *Petrosia* sp. of both Nok Island and Khang Khao Island were not statistically different

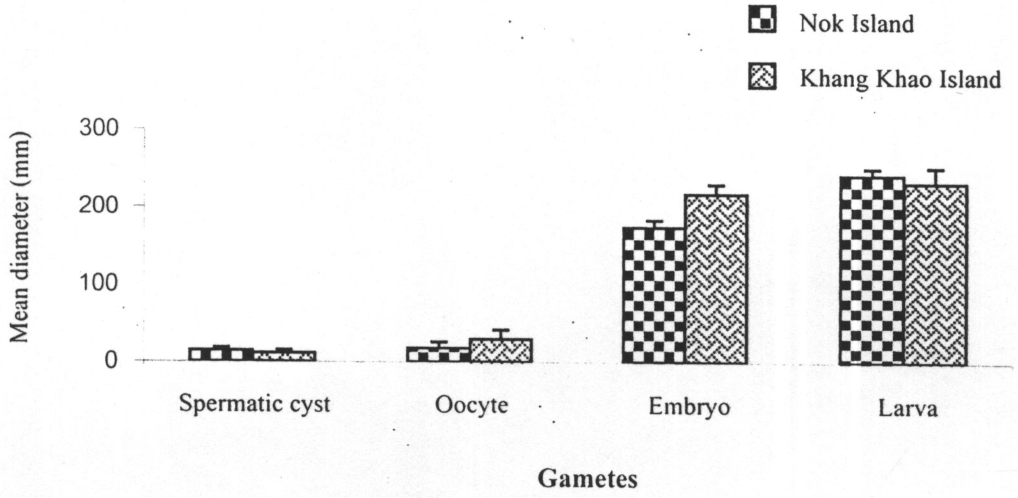

Figure 37 Mean Diameters of Spermatic cysts, Oocytes, Embryos and Larvae in *Petrosia* sp. at Nok Island and Khang Khao Island in December 1998

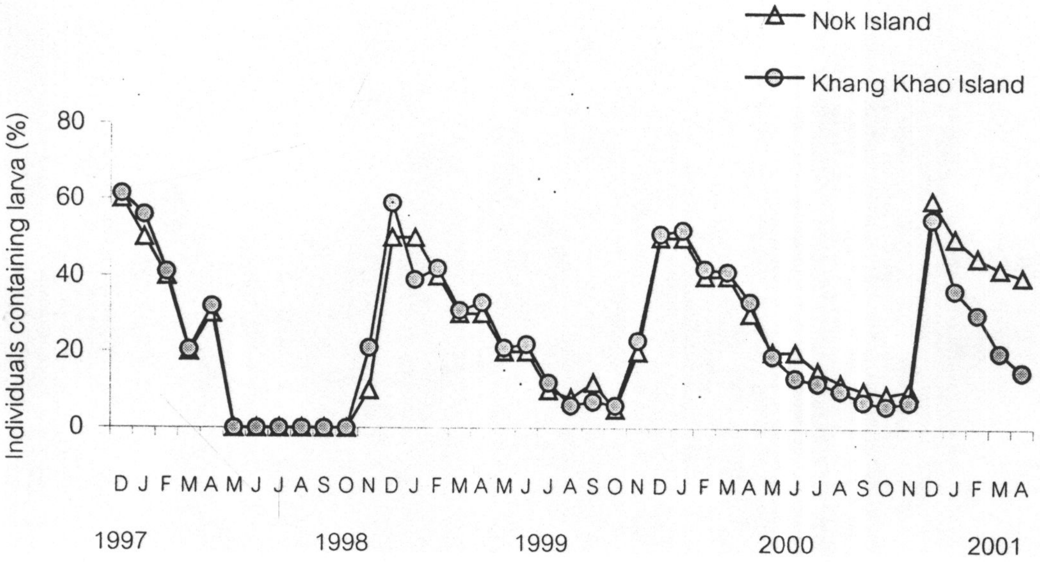

Figure 38 Presence of Parenchymella Larvae in Marked Individuals of *Petrosia* sp. at Nok Islamd and Khang Khao Island During December 1997 – April 2001

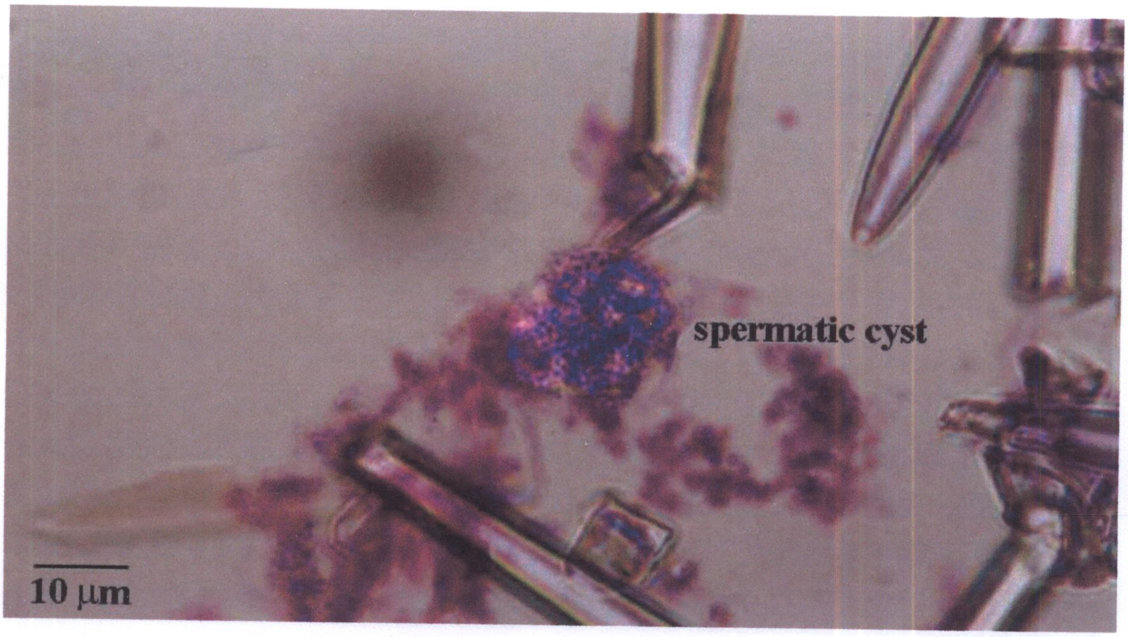

Figure 39 A Spermatic Cyst of *Petrosia* sp. at Nok Island in January 1999

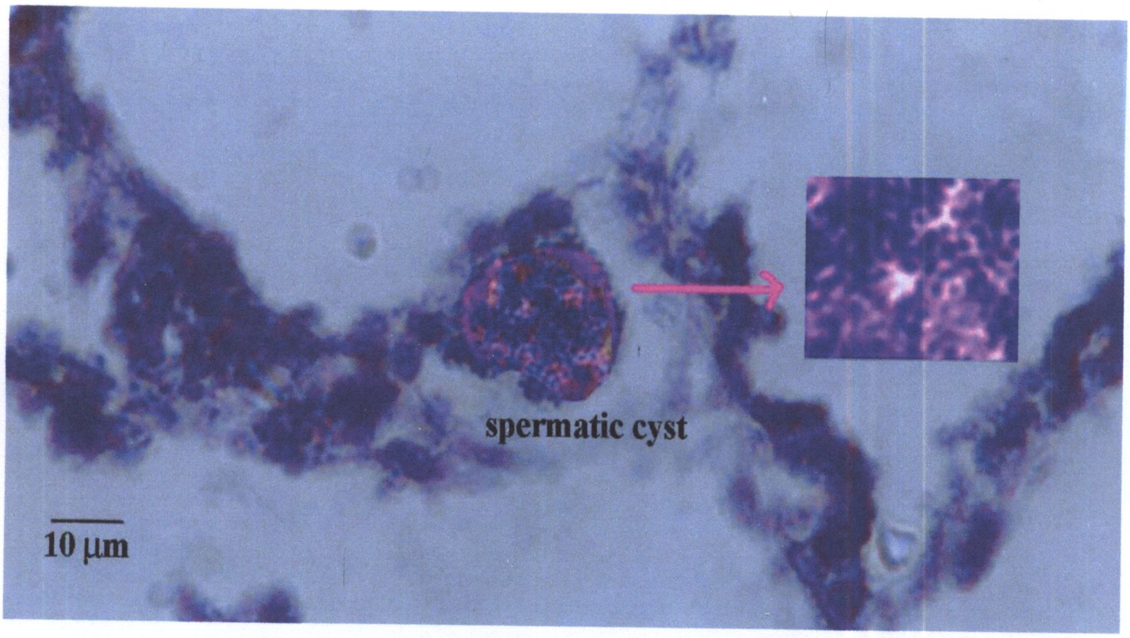

Figure 40 A Spermatic Cyst of *Petrosia* sp. at Khang Khao Island in December 1998

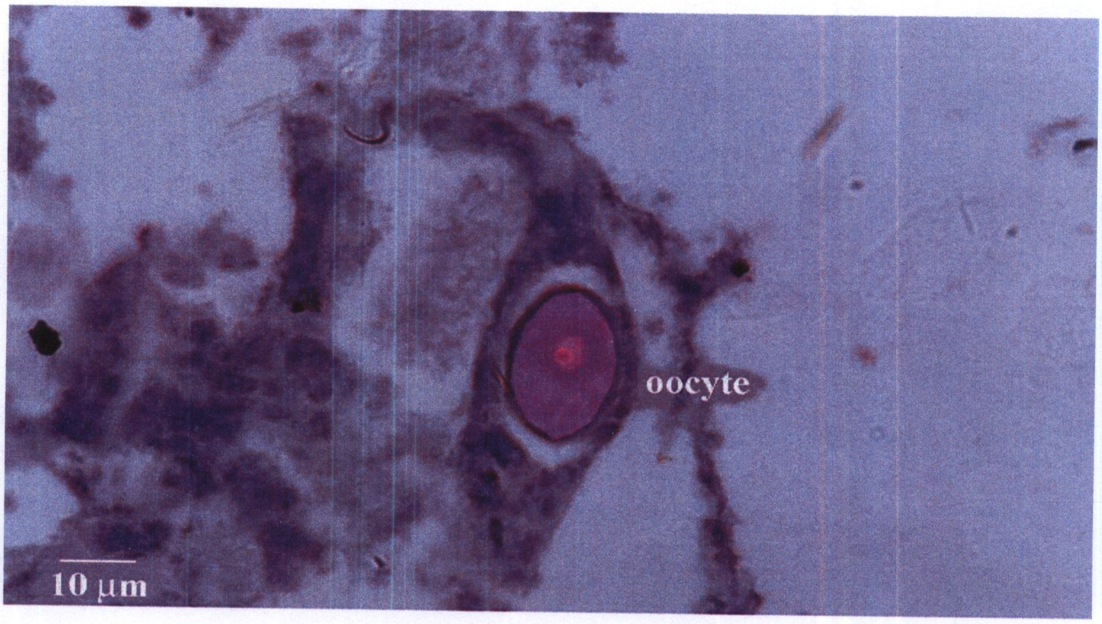

Figure 41 An Oocyte of *Petrosia* sp. at Nok Island in January 1999

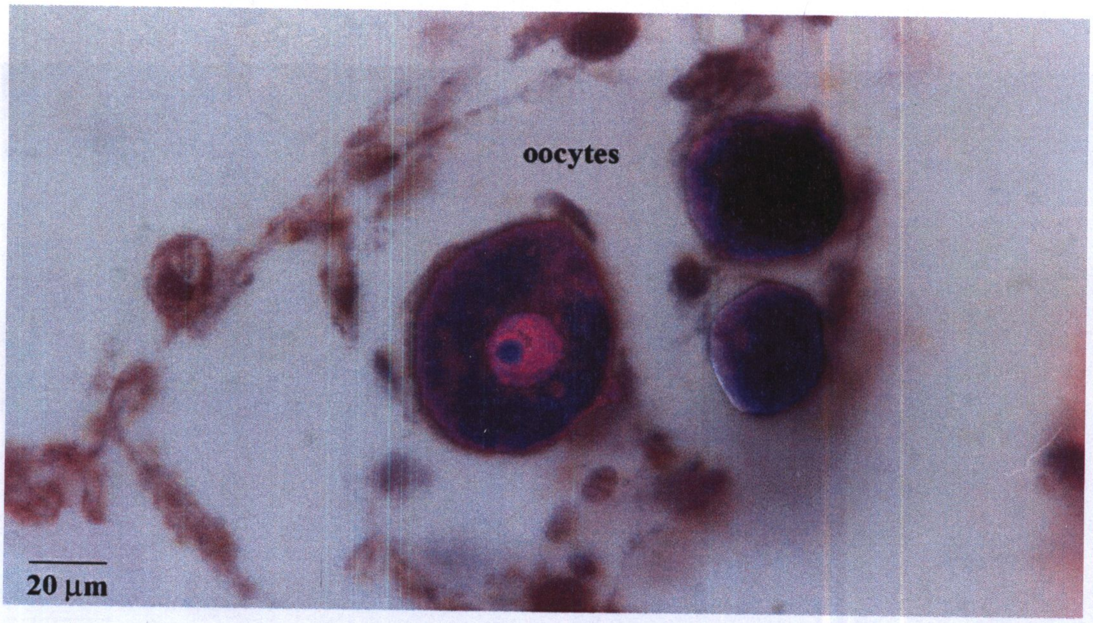

Figure 42 Oocytes of *Petrosia* sp. at Khang Khao Island in December 1998

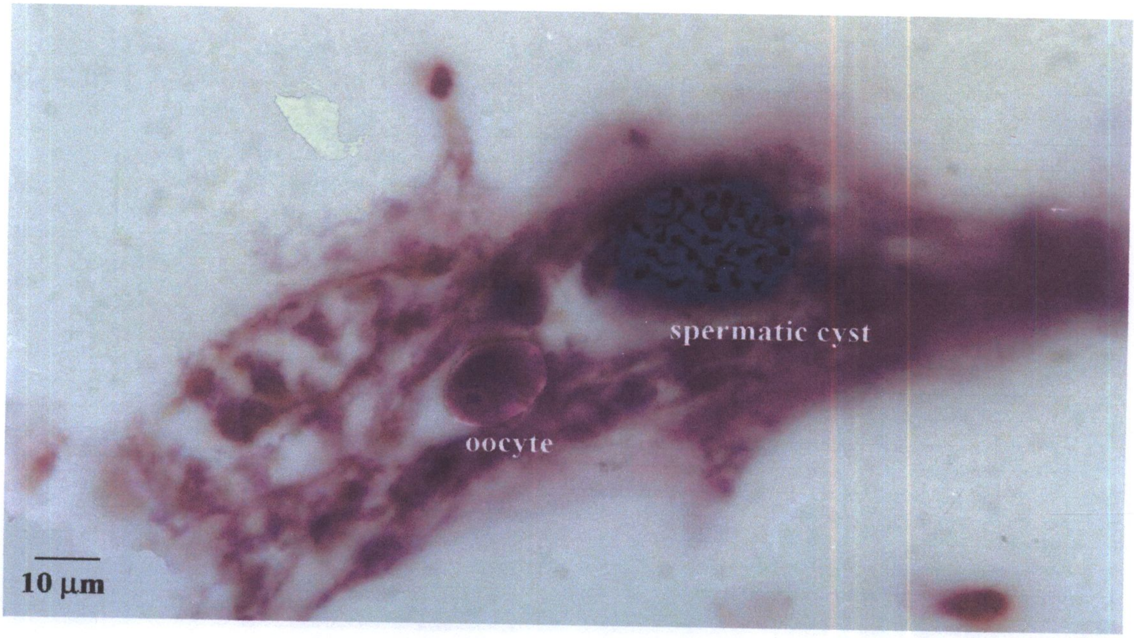

Figure 43 A Spermatic Cyst and an Oocyte of *Petrosia* sp. at Nok Island in January 1999

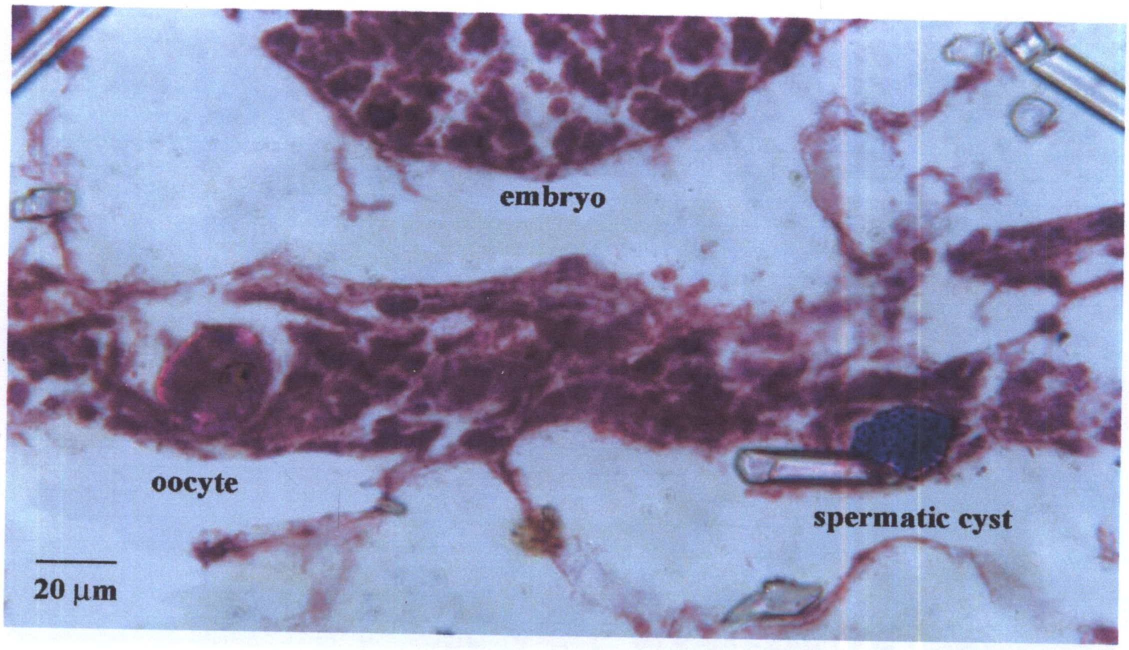

Figure 44 A Spermatic Cyst, an Oocyte and an Embryo of *Petrosia* sp. at Khang Khao Island in December 1998

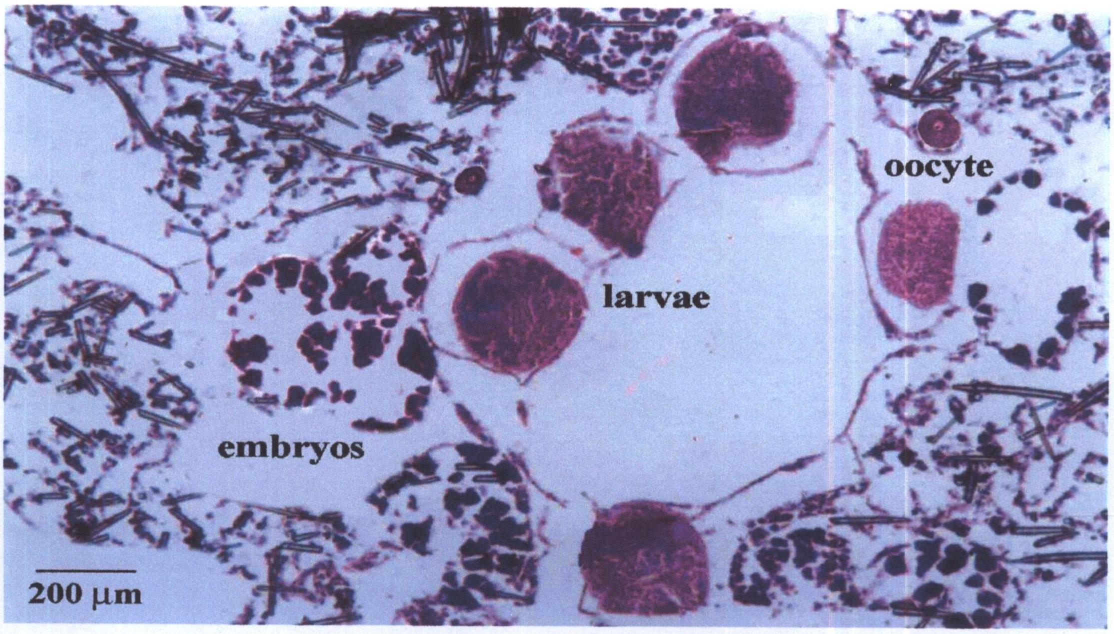

Figure 45 Oocytes, Embryos and Parenchymella Larvae of *Petrosia* sp. at Nok Island in January 1999

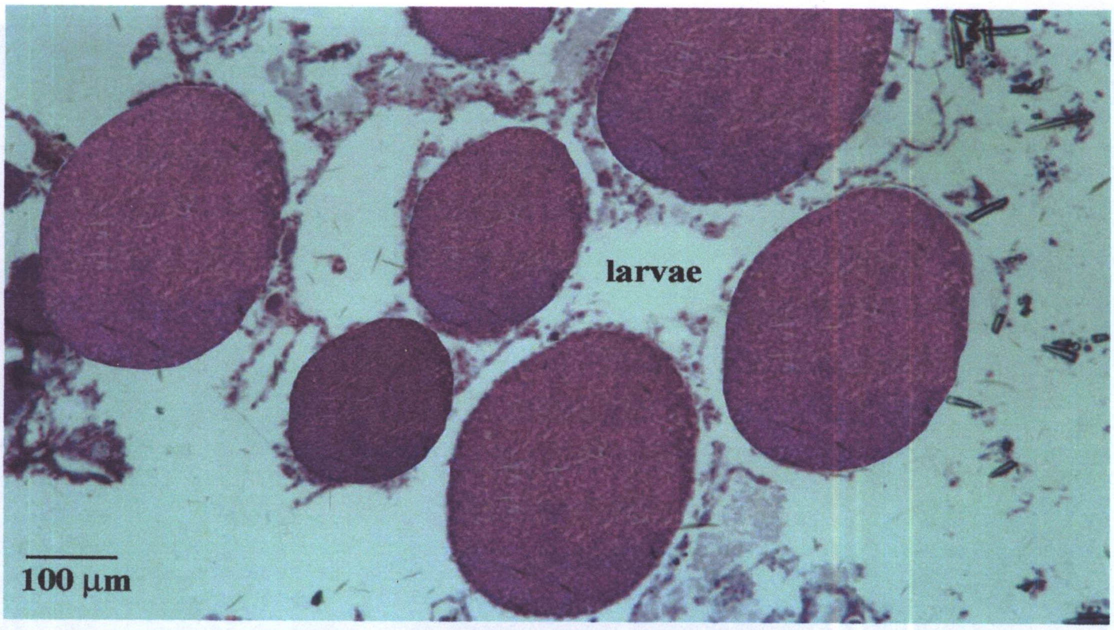

Figure 46 Different Sizes of Parenchymella Larvae in *Petrosia* sp. at Khang Khao Island in February 1999

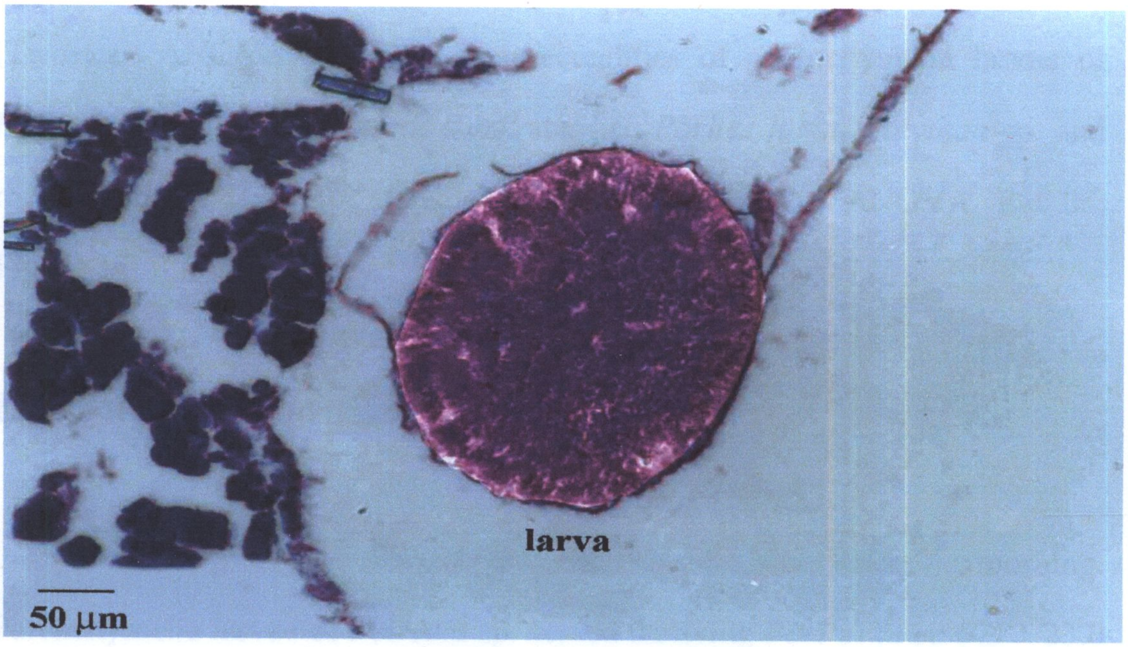

Figure 47 A Parenchymella Larva of *Petrosia* sp. at Nok Island in December 1997

Figure 48 Parenchymella Larvae of *Petrosia* sp. Developed in a Brood Chamber at Khang Khao Island in December 1998

(Two-way ANOVA, $P>0.05$). The fecundities of parenchymella larvae of *Petrosia* sp. on three types of substrate, i.e., *Porites lutea*, *Palythoa* sp. and rock substrates were not statistically different (Two-way ANOVA, $P>0.05$, Figure 49).

Larval Releasing and Settlement

Releasing times of parenchymella larvae were observed in the morning. Parenchymella larvae of *Petrosia* sp. settled on living *Porites lutea* (43 ± 23.0 %), significantly higher than other substrates, dead coral, *Palythoa* sp., rock, gypsum plate and gypsum plate+sediment (One-way ANOVA, $P<0.05$). Average settled number of parenchymella larvae of *Petrosia* sp. on six types of substrates, i.e., living *Porites lutea*, dead coral, *Palythoa* sp., rock, gypsum plate and gypsum plate+sediment were 43 ± 23.0 %, 0 %, 2.5 ± 0.8 %, 1.1 ± 0.4 %, 2.5 ± 1.6 % and 0 %, respectively (Figure 50).

Vegetative Growth

Average growth rate of *Petrosia* sp. at Nok Island (0.7 cm^3/day) was significantly higher than that at Khang Khao Island (0.6 cm^3/day) (Two-way ANOVA, $P<0.05$). Average growth rates of *Petrosia* sp. on three types of substrate, i.e., *Porites lutea*, *Palythoa* sp. and rock substrate were not statistically different (Two-way ANOVA, $P>0.05$). Average growth rates of *Petrosia* sp. on three types of substrate, i.e., *Porites lutea, Palythoa* sp. and rock at Nok Island were 0.8 ± 0.1 cm^3/day, 0.7 ± 0.2 cm^3/day and 0.6 ± 0.1 cm^3/day, respectively while those at Khang Khao Island were 0.7 ± 0.1 cm^3/day, 0.6 ± 0.1 cm^3/day and 0.5 ± 0.1 cm^3/day, respectively (Figure 59, 61-64).

Mortality Rate

Average partial mortality rates of *Petrosia* sp. at Nok Island and Khang Khao Island were not statistically different (Two-way ANOVA, P>0.05, Figure 44). Average partial mortality rates of *Petrosia* sp. on three types of substrate, i.e., *Porites lutea*, *Palythoa* sp. and rock substrates were also not statistically different (Two-way ANOVA, P>0.05). Average partial mortality rates of *Petrosia* sp. on three types of substrate, i.e., *Porites lutea, Palythoa* sp. and rock at Nok Island were 1.1 ± 0.23 cm^3/day, 0.9 ± 0.22 cm^3/day and 0.8 ± 0.16 cm^3/day, respectively while those at Khang Khao Island were 0.9 ± 0.31 cm^3/day, 0.8 ± 0.22 cm^3/day and 0.7 ± 0.19 cm^3/day, respectively (Figure 60, 65-68).

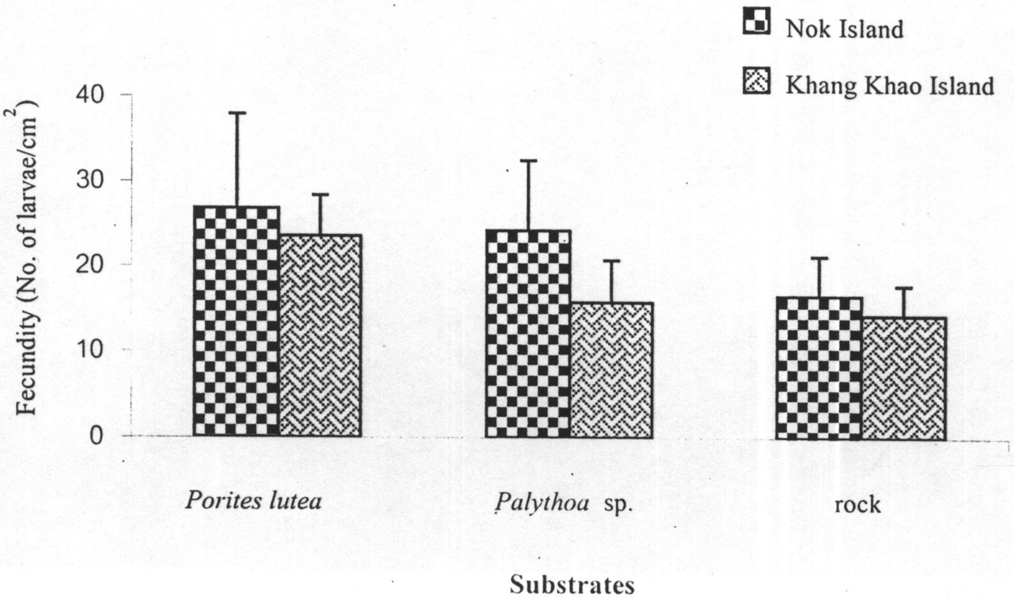

Figure 49 Fecundities of Parenchymella Larvae in *Petrosia* sp. on Different Types of Substrate at Nok Island and Khang Khao Island in December 1998

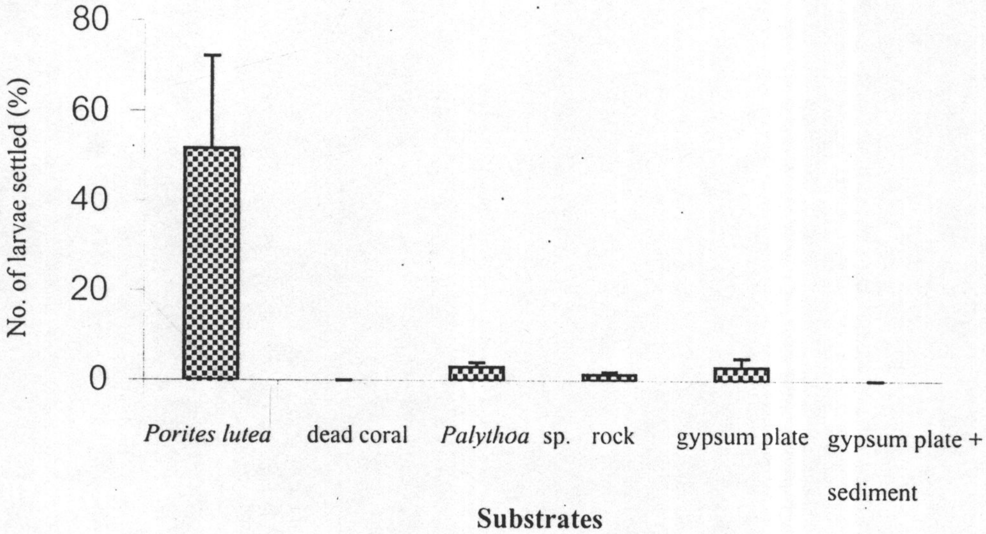

Figure 50 Larval Settlement of *Petrosia* sp. on Six Types of Substrate: *Porites lutea* , Dead Coral, *Palythoa* sp., Rock, Gypsum Plate and Gypsum+Sediment in an Aquarium

Figure 51 Parenchymella Larvae of *Petrosia* sp. Developed in a Brood Chamber at Nok Island in January 1999

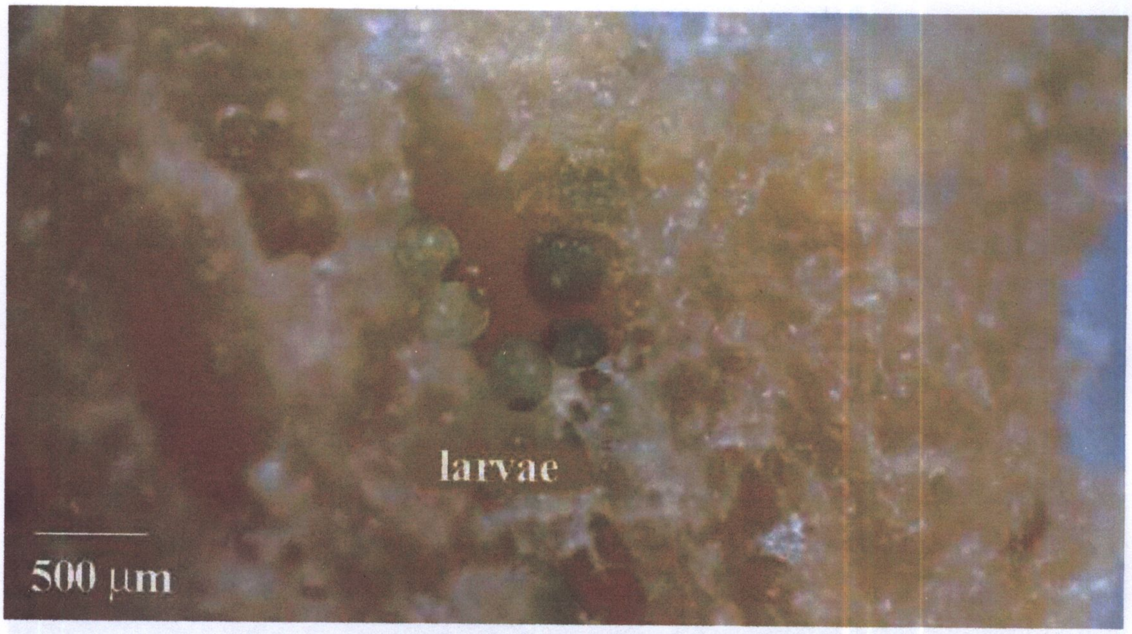

Figure 52 Parenchymella Larvae of *Petrosia* sp. Developed in a Brood Chamber at Khang Khao Island in February 1999

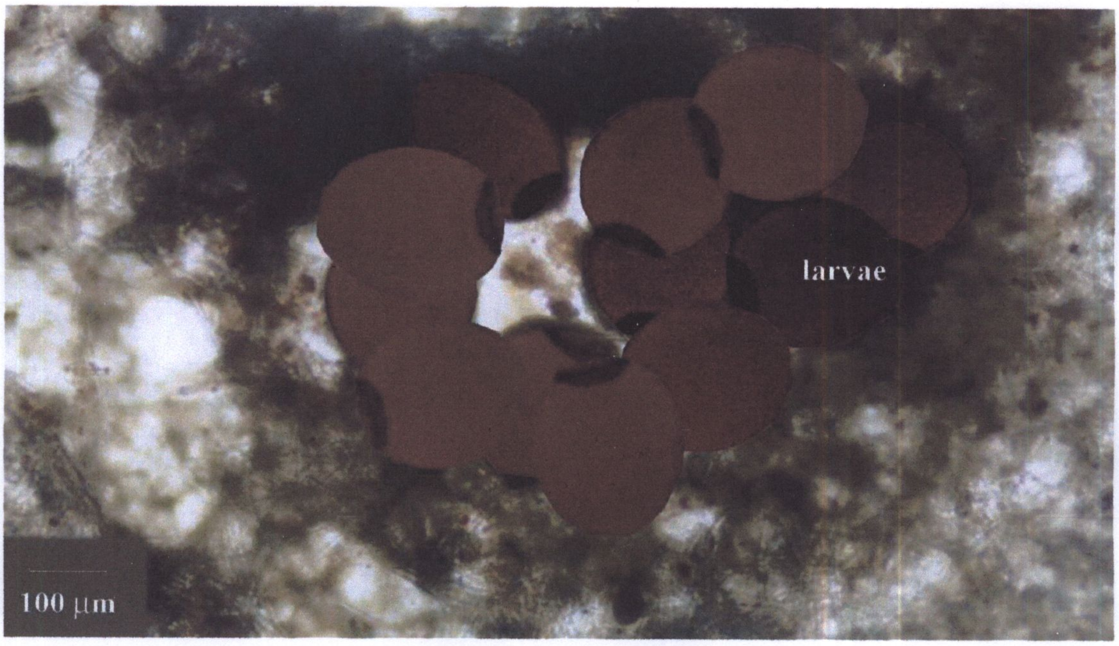

Figure 53 Parenchymella Larvae of *Petrosia* sp. at Khang Khao Island in December 1999

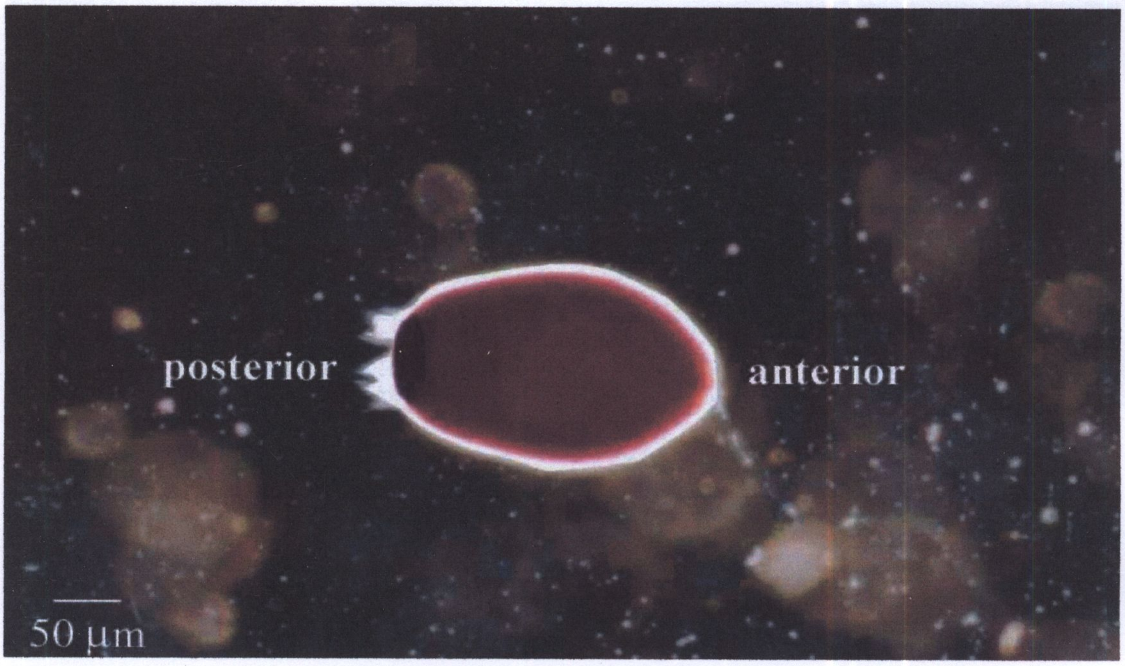

Figure 54 A Parenchymella Larva of *Petrosia* sp. Showing Posterior and Anterior Sides

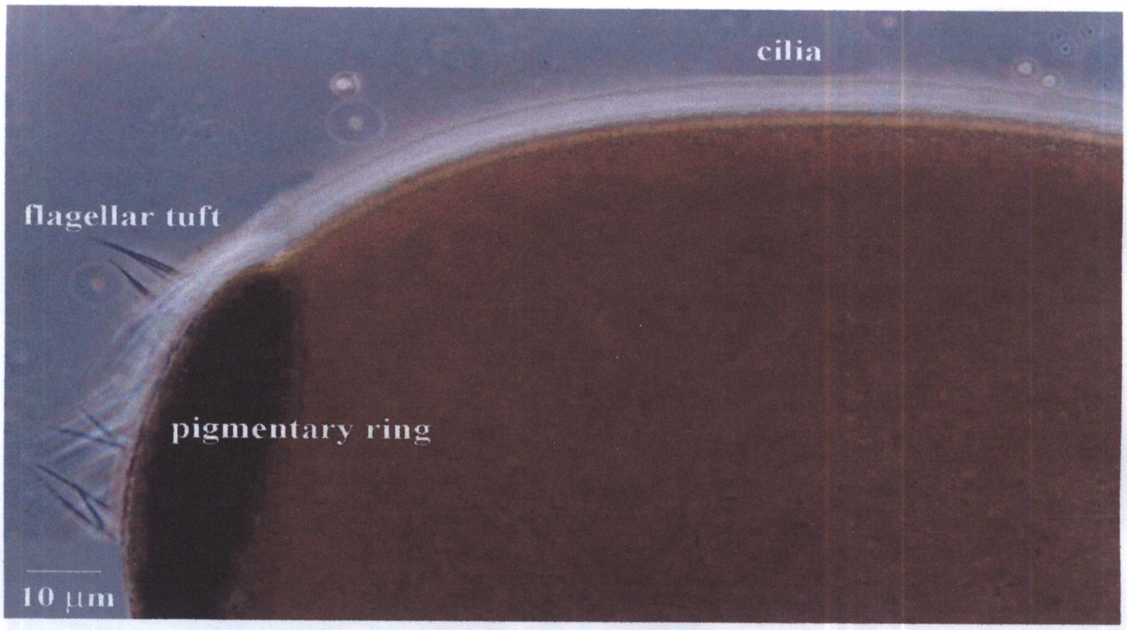

Figure 55 Posterior of a Parenchymella Larva of *Petrosia* sp. at Nok Island Showing Flagellar Tuft, Pigmentary Ring and Cilia

Figure 56 Posterior of a Parenchymella Larva of *Petrosia* sp. at Khang Khao Island Showing Flagellar Tuft, Pigmentary Ring and Cilia

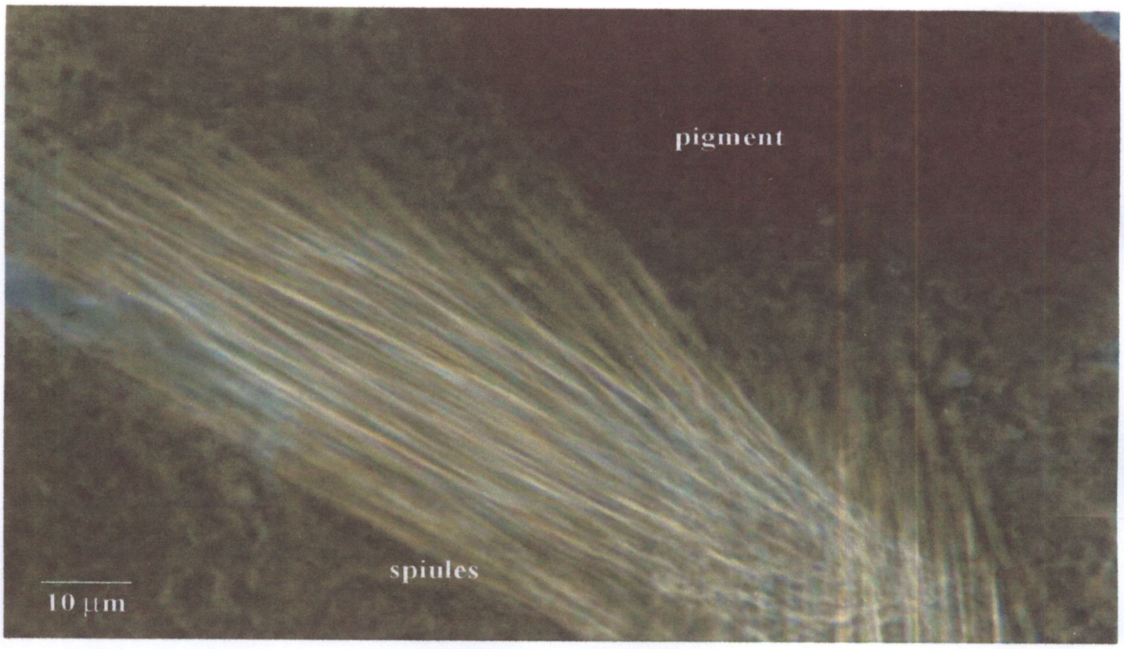

Figure 57 Small Spicules on a Parenchymella Larva in *Petrosia* sp.

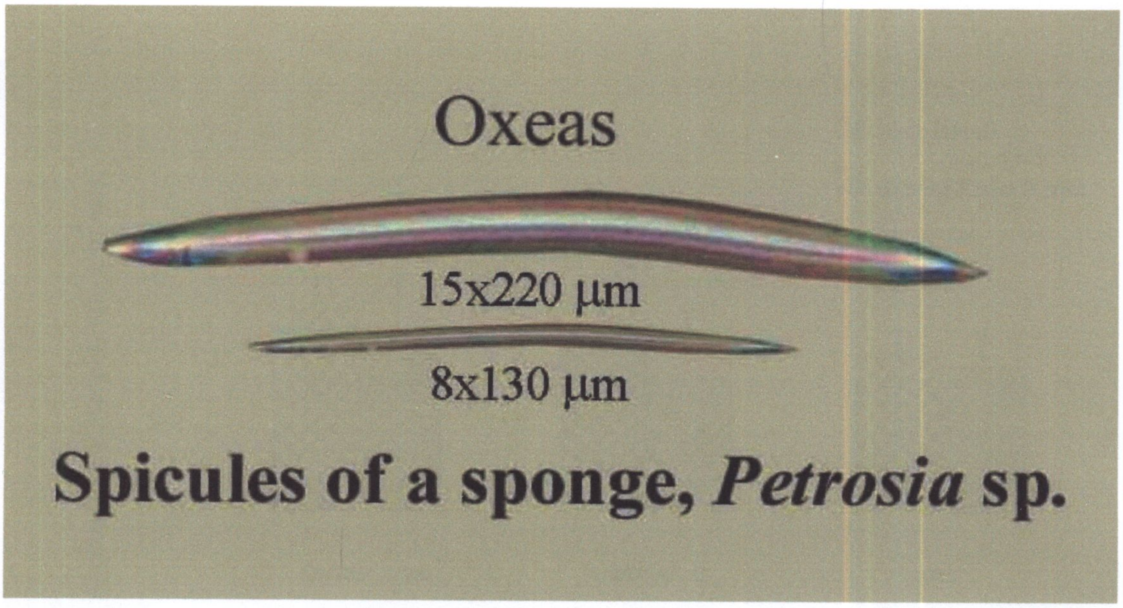

Figure 58 Different Sizes of Oxeas Spicules in Adult of *Petrosia* sp.

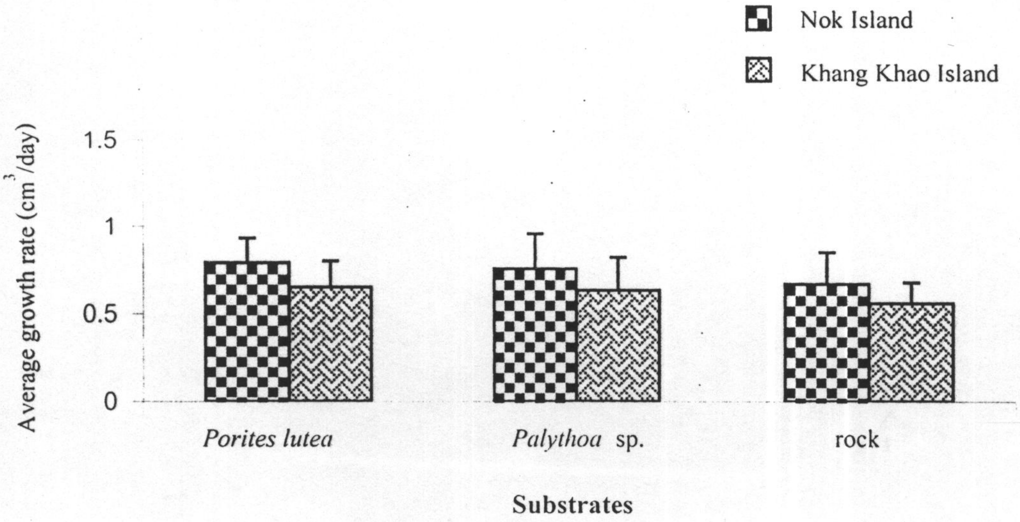

Figure 59 Average Growth Rates of *Petrosia* sp. on Different Types of Substrate at Nok Island and Khang Khao Island

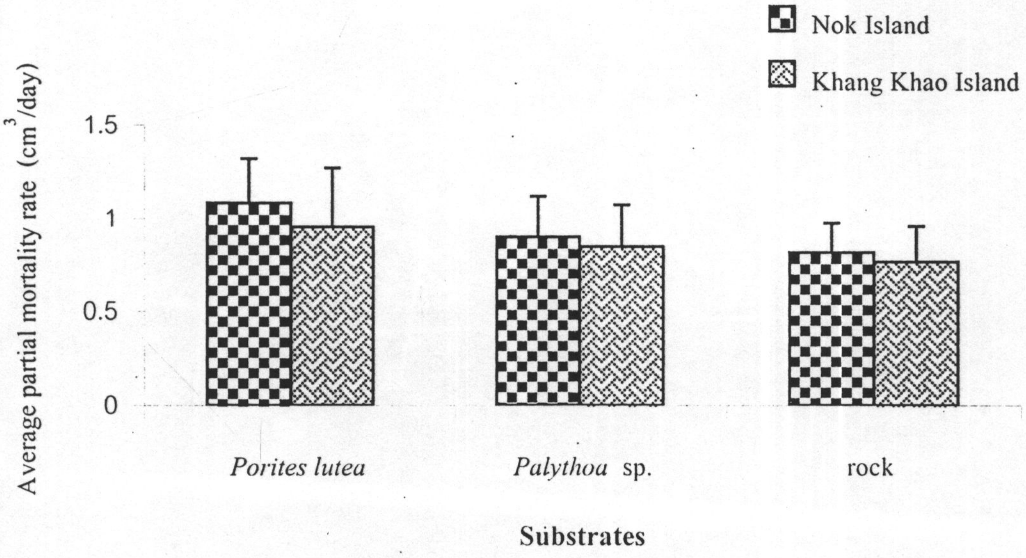

Figure 60 Average Partial Mortality Rates of *Petrosia* sp. on Different Types of Substrate at Nok Island and Khang Khao Island

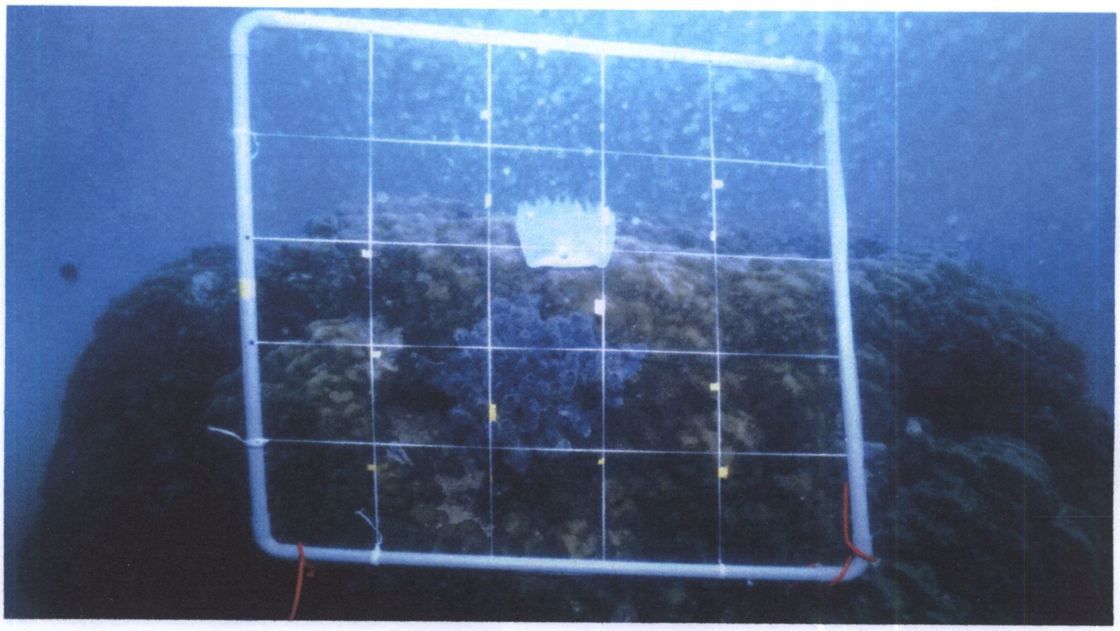

Figure 61 A Marked Individual of *Petrosia* sp. at Nok Island in November 1998

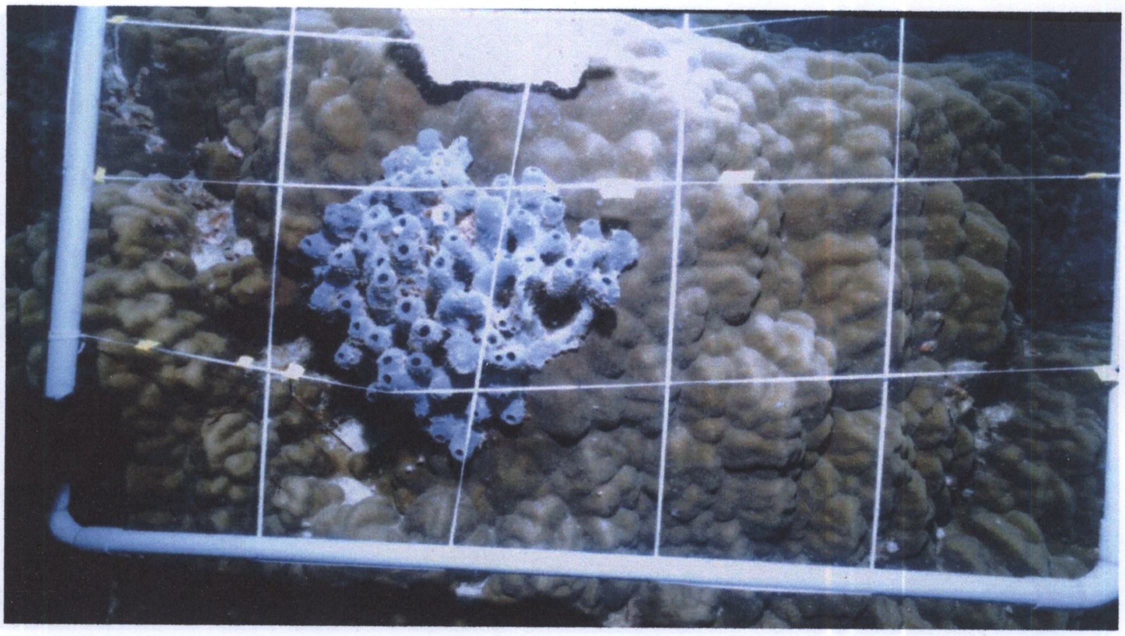

Figure 62 The Same Marked Individual (Figure 61) of *Petrosia* sp. Observed in May 1999

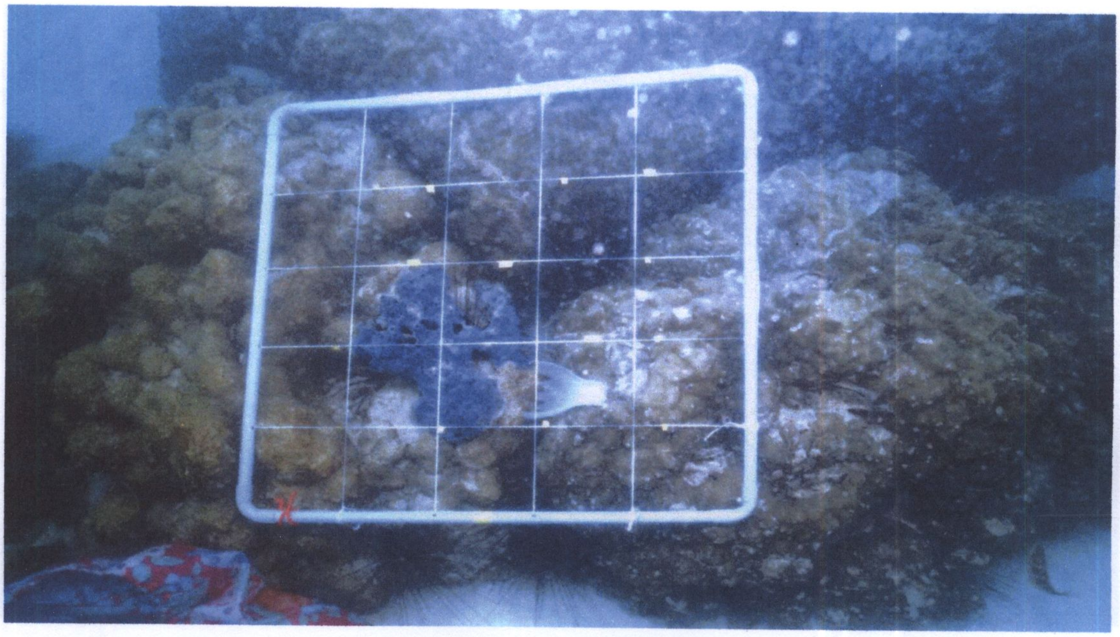

Figure 63 A Marked Individual of *Petrosia* sp. at Khang Khao Island in November 1998

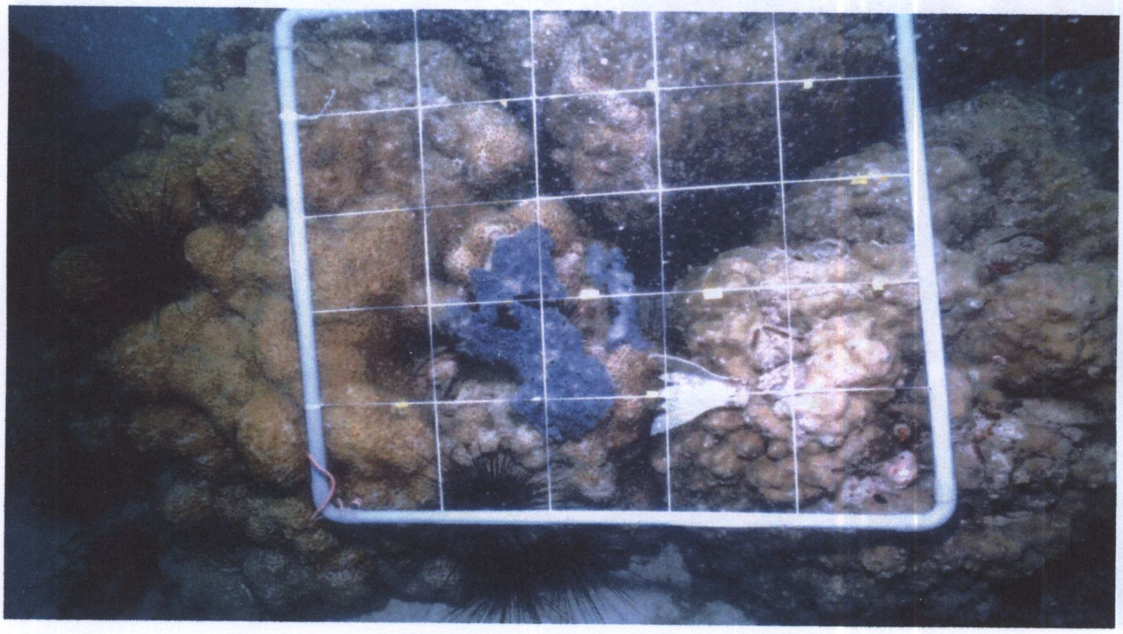

Figure 64 The Same Marked Individual (Figure 63) of *Petrosia* sp. Observed in May 1999

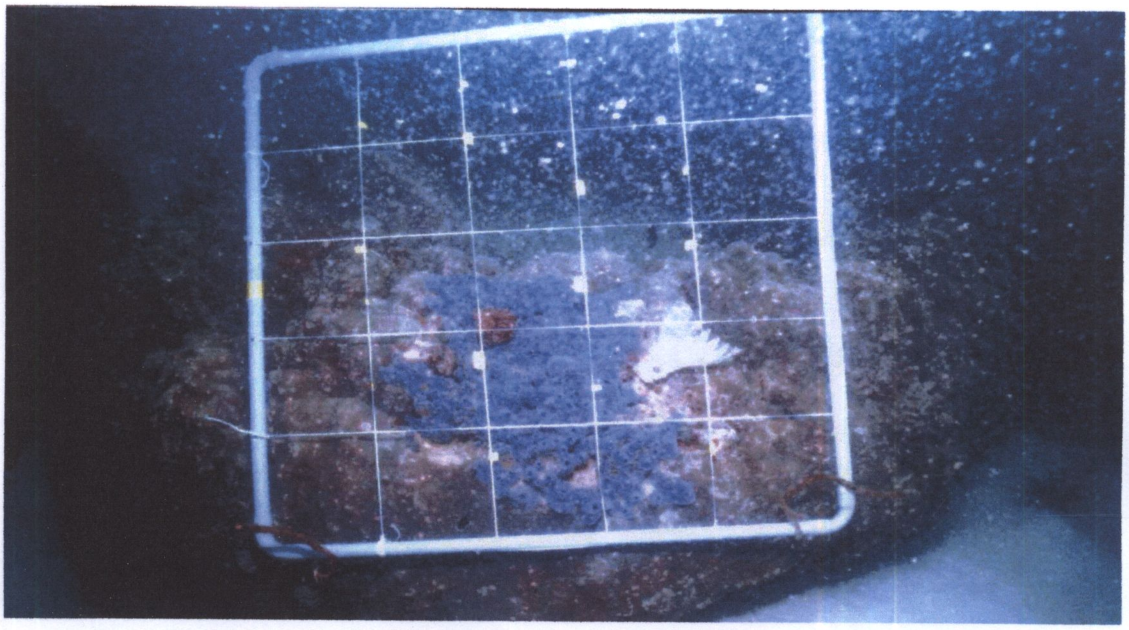

Figure 65 A Marked Individual of *Petrosia* sp. at Nok Island in November 1998

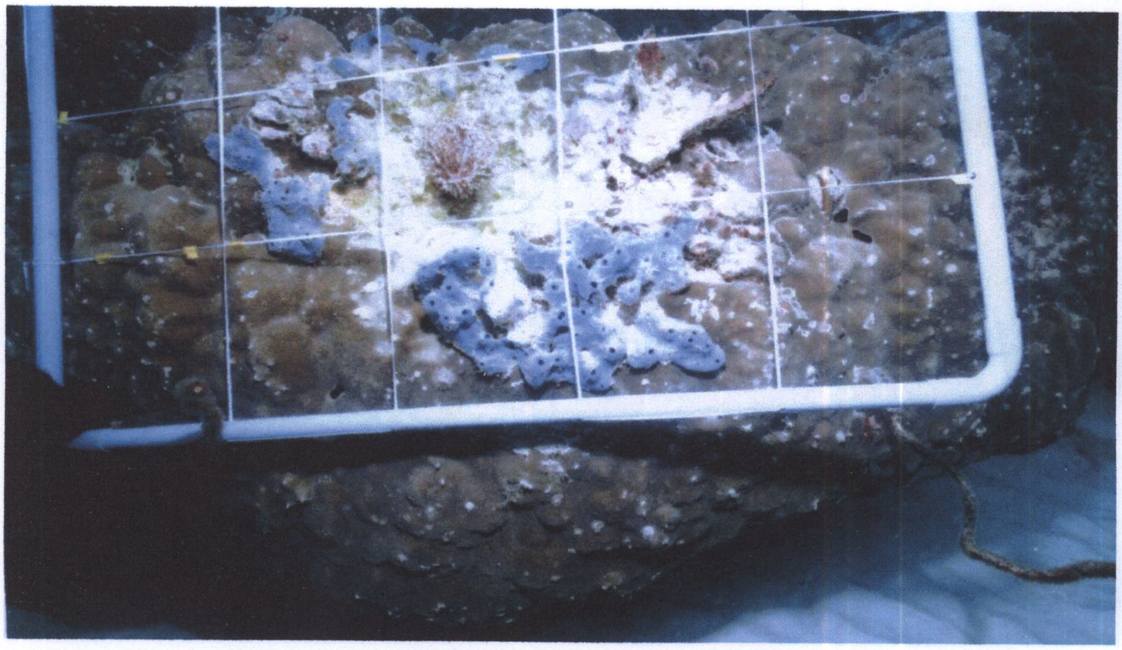

Figure 66 The Same Marked Individual (Figure 65) of *Petrosia* sp. Observed Partial Mortality in May 1999

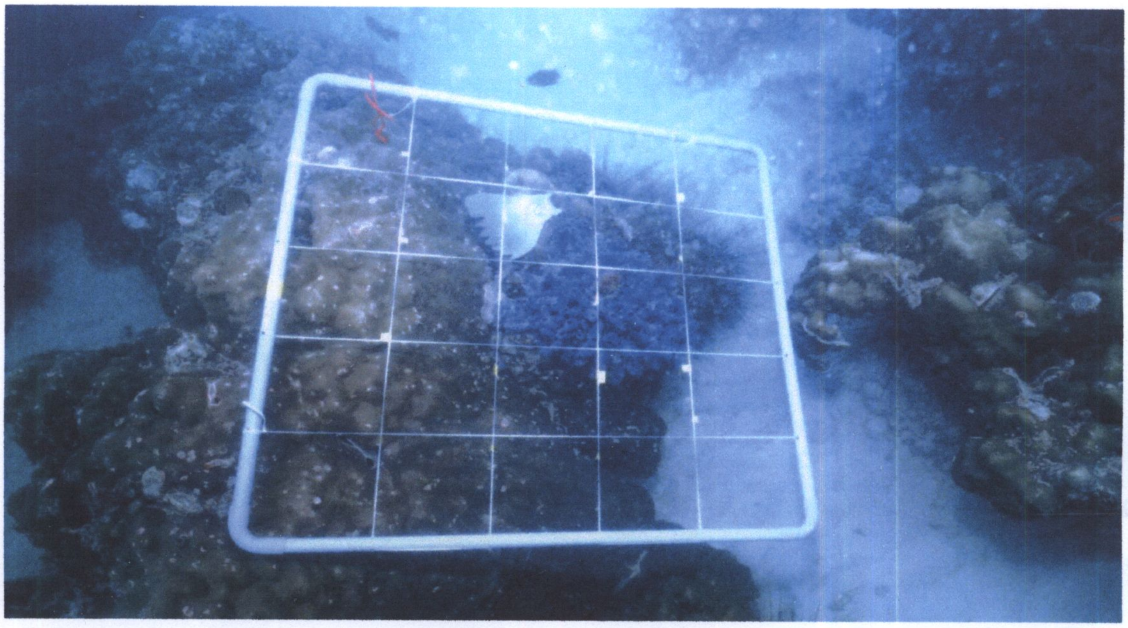

Figure 67 A Marked Individual of *Petrosia* sp. at Khang Khao Island in November 1998

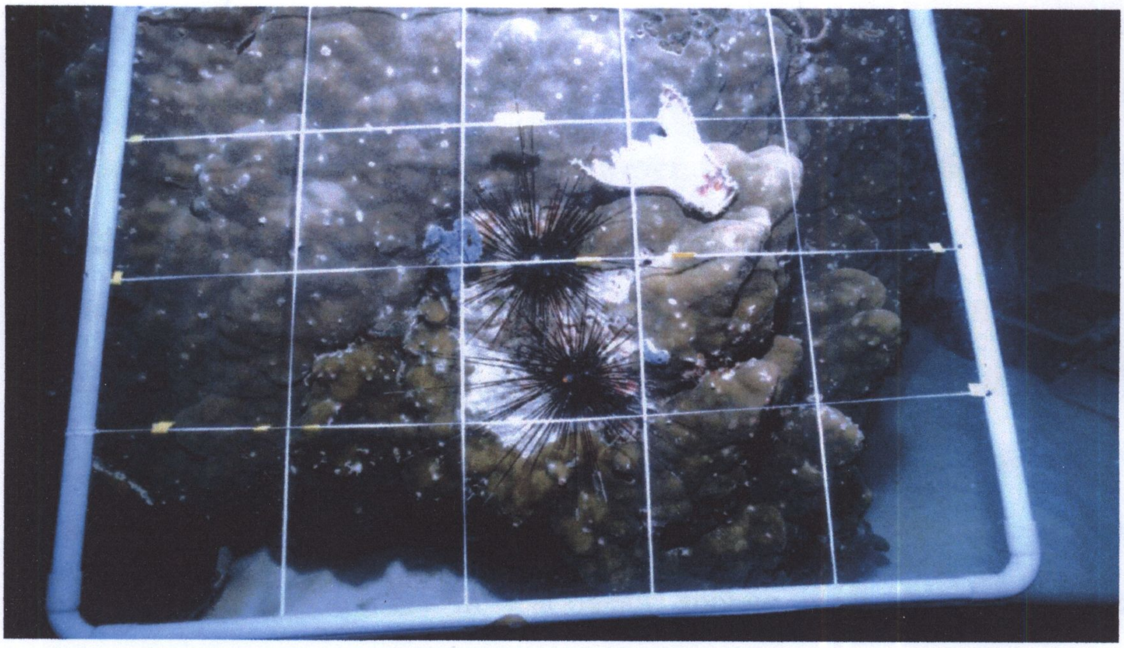

Figure 68 The Same Marked Individual (Figure 67) of *Petrosia* sp. was Grazed by a Sea Urchin, *Diadema setosum* in May 1999.

CHAPTER 5

DISCUSSIONS

Population Density and Distribution Pattern

Mean population densities of *Petrosia* sp. at both Nok Island and Khang Khao Island (3-7 m. in depth) were in the same range (0.07- 0.37 individuals/m^2). Densities of *Tethya microstella* in the Gangehi Island were 0.27 individuals/m^2 (Sara et al. 1993, 351). Population densities of *Xestospongia exigua* at 30 m depth in Great Barrier Reef were 0.051-0.15 individuals/m^2 (Wilkinson and Evans 1989, 4). Moreover, population densities *Tethya crypta* at 3 m depth near Discovery Bay, Jamaica were 0.16 individuals/m^2 (Reiswig 1973, 202). However, population densities of a small sponge, *Aplysina fulva* in the San Blas Island, Panama were 0.02 individuals/m^2 (Wulff 1995, 58). Densities of *Tethya aurantium* in the Mediterranean Sea were 6.59 individuals/m^2 (Corriero et al. 1989, 308). *Petrosia ficiformis* was abundant on vertical or slightly overhanging rock faces below 20 m. at Balearic Island (Uriz et al. 1992, 109). Mean population densities of three species in Caribbean sponge, *Iotrochota birotulata*, *Amphimedon rubens* and *Aplysina fulta* were 17.5 individuals/m^2, 7.8 individuals/m^2 and 16.2 individuals/m^2, respectively (Wulff 1991, 239). Mean densities of four Caribbean sponges, *Tedania ignis*, *Sigmadocia caerulea*, *Haliclona tubifera* and *Halichondria magniconulosa* were 8±6.6, 6±5.5, 5±

6.2 and 0.8±1.6 individuals/m^2, respectively (Maldonado and Young 1996, 172). Obviously, most individuals of *Petrosia* sp. at both Nok Island and Khang Khao Island attached on a massive coral, *Porites lutea*, which was the most abundant coral species in the Gulf of Thailand. Most individuals of *Tethya aurantium* and *T. citrina* were also attached on a seagrass, *Posidonia oceanica* rhizomes, which was the most abundant substrate in the Sicilian lagoon Stagnone di Marsala, Mediterranean (Corriero et al. 1989, 308). Most individuals of *Mycale fistulifera* in the Red Sea was attached on two branching corals, *Stylophora pistillata* and *Acropora* sp. (Meroz and Ilan 1995, 444). However, *Petrosia ficiformis* grew on bottoms with sediment in Mediterranean Sea (Bibiloni et al. 1989, 332). Sponge/coral interactions in the Gulf of Thailand and Caribbean reefs were also the same patterns, i.e., in many cases, sponges overgrowth corals (Aerts and van Soest 1997, 125; Aerts 1998, 241). The dispersion patterns of *Petrosia* sp. at the two study sites were clumped due to patchy distributions of available substrates. Similarly, distribution patterns within a population of *Haliclona cymiformis* in the Great Barrier Reef were also clumped due to the physical characteristics of the habitat (Trautman 1998, 602 and Trautman et al. 2000, 93).

Body Size

Average body sizes of *Petrosia* sp. at Nok Island and Khang Khao Island were 416 cm^3 and 222 cm^3, respectively. However, the body sizes of four species in the Demospongiae from the Great Barrier Reef, i.e., *Xestospongia bergquistia X. exigu*, *Haliclona cymiformis* and *H. amboinensis* were 3405±2000, 121±78, 100±46 and 42±32 cm^3, respectively (Fromont 1994, 310).

Temperature and Reproductive Activity

Reproductive activities of *Petrosia* sp. at Nok Island and Khang Khao Island were lack of evidence of the effects of water temperature on reproduction. However, the onset of reproduction in the Red Sea sponge, *Niphates* sp. is probably triggered by an increasing of water temperature in spring (Ilan and Loya 1988, 747). The onsets of gametes in two sponges, *Xestospongia bergquistia* and *X. testudinaria* from the Great Barrier Reef implied the decreasing of water temperature (Fromont and Bergquist 1994, 124). The onsets of gametes in two sponges, *Haliclona amboinensis* and *H. nitida* from the Great Barrier Reef occurred when water temperatures were increasing (Fromont 1994, 127).

Salinity and Reproductive Activity

Reproductive activities of *Petrosia* sp. at Nok Island and Khang Khao Island were not related to the salinity levels. However, reproductive activity of *Microciona prolifera* in the explants appeared to be normal at 20 ppt, somewhat slowed at 15 ppt and inhibited at 10 ppt (Knight and Fell 1987, 253).

Reproductive Mode

Petrosia sp. from coral communities at Nok Island and Khang Khao Island were viviparous and contemporaneously hermaphroditic species. Gametes were found all year round. However, it has not been yet established, whether they exist self or non-self fertilization. So far, all species studied in the family Petrosiidae are oviparous gonochoric species and synchronous development of gametes, e.g. *Xestospongia bergquistia*, *X. exigua* and *X. testudinaria* (Fromont and Bergquist 1994), *Petrosia ficiformis* (Liaci et al. 1973 cited in Sara 1992, 6, see Table 1). Spermatic cysts, oocytes, embryos and parenchymella larvae of *Petrosia* sp. were asynchronously developed. Fromont (1994, 131) also noted that gametes of *Haliclona amboinensis, Haliclona cymiformis* and *Niphates nitida* were asynchronously developed. These patterns were reported in *Niphates* sp. from the Red Sea as well (Ilan and Loya 1988, 745). *Mycale* sp. in the South Mole, Fremantle, Western Australia is a viviparous and contemporaneously hermaphroditic. Gametes were found in the field during December and January in 1997 and 1998 (Fromont 1998, 187). A Mediterranean sponge, *Mycale contarenii*, is viviparity (Corriero et al. 1998, 319). A viviparous sponge *Halisarca dujardini* from the White Sea, Arctic was asynchronous development of gametes (Ereskovsky 1998, 160). Three species of the Petrosida in the Great Barrier Reef, i.e., *Xestospongia bergquistia*, *X. testudinaria* and *X. exigua* were oviparous and contemporaneously gonochoric. Oocytes and spermatic cyst were evenly distributed throughout the mesohyl (Fromont 1994, 307). A sponge, *Aplysina cauliformis*, from the Barbados is oviparity and dioeciousness (Tsurumi and Reiswig 1997, 1).

Timing of Sexual Reproduction

Gametes of *Petrosia* sp. were developed all year round, while the peak of gamete development occurred during December-January of each year. However, gametes of *Xestospongia bergquistea, X. exigua* and *X. testudinaria* in the Great Barrier Reef were not developed all year round (Fromont and Bergquist 1994, 119). *Negombata magnifica* in the Red Sea had a peak of sexual reproduction during the summer (Ilan 1998, 248).

Gamete Development

Spermatic cysts and oocytes of *Petrosia* sp. were derived from the choanocyte. Compatible spermatic cysts and oocytes in the Demospongiae were derived from the transformation of choanocyte chambers (Sasa 1992, 47). Fertilization of *Petrosia* sp. was lack of evidence in its presence. However, in all cases in which internal fertilization has been observed, it appears that spermatozoa are transported to the oocytes by cellular intermediaries(Fell 1974, 97). The spermatozoa are substantially diversified. The sequential changes of the structure of spermatozoa in the choanocytes are believed to be the process by which the spermatozoa transform to spermiocysts before fertilization. The involvement of two carrier cells are

Table 1 Sexual Characteristics, Oviparity and Viviparity in the Haplosclerida (G, gonochorism; Hc, contemporaneous hermaphroditism; Hs, successive hermaphroditism; o, oviparity; v, viviparity)

Order	Family	Species	Sex phenotype	Oviparity/ Viviparity	References	
Haplosclerida	Haliclonidae	*Haliclona ecbasis*	Hs?	v	Fell 1969	Cited by Sara 1992, 7
		Haliclona permollis	G?	v	Elvin 1976	
		Haliclona loosanoffi	G?	v	Fell 1976b	
		Haliclona oculata	H?	v	Wapstra and van Soest 1987	
		Haliclona xena	H?	v	Wapstra and van Soest 1987	
		Haliclona amboinensis	G	v	Fromont 1994, 127	
		Haliclona cymiformis	G	v	Fromont 1994, 127	
	Petrosiidae	*Xestospongia bergquistia*	G	o	Fromont and Bergquist 1994, 119	
		Xestospongia testudinaria	G	o	Fromont and Bergquist 1994, 119	
		Xestospongia exigua	G	o	Fromont and Bergquist 1994, 119	
		Petrosia ficiformis	G?	o	Scalera Liaci et al. 1973a (Cited by Sara 1992, 7)	
		Petrosia sp.	Hc	v	This study	

carrying the sperm to the oocyte (Nakamura et al. 1998, 179; Watanabe and Okada 1998, 193). The fertilization process of the Barents Sea sponge, *Leucosolenia complicata*, begins with the protein capsule penetrating the oocyte and gradually resolving in its ooplasm (Ankina 1999, 44).

Density of Gametes

Average densities of gametes, i.e., spermatic cysts, oocytes, embryos and larvae in *Petrosia* sp. from the Gulf of Thailand were 76.2±20.15 cysts/cm^2, 116±13.09 oocytes/cm^2, 27.3±7.91 individuals/cm^2 and 19±5.15 individuals/cm^2, respectively. However, average density of gametes, i.e., spermatic cysts, oocytes, embryos and larvae in *Niphates nitida* from the Great Barrier Reef were 126.4 cysts/cm^2, 49.2 oocytes/cm^2, 125.6 individuals/cm^2 and 156 individuals/cm^2, respectively (Fromont 1994, 130-131). Average density of spermatic cysts and oocytes in *Xestospongia testudinaria* from the Great Barrier Reef were 1940 cysts/cm^2 and 372 oocytes/cm^2, respectively (Fromont 1994, 309). Densities of spermatic cysts and embryos in *Pellina semitubulosa* from Italian coasts were 314±129.5 cysts/mm^3 and 31±18 individuals/mm^3, respectively (Mercurio et al. 2000, 91).

Size of Gametes

Mean diameters of spermatic cysts, oocytes, embryos and larvae in *Petrosia* sp. were 14±3.8 μm, 18±7.6 μm, 173±9.4 μm and 241±9.2 μm, respectively. However, diameter of spermatic cysts, oocytes, embryos and larvae in *Haliclona amboinensis* from the Great Barrier Reef were 26±4 μm, 10-30 μm, 240 μm and 340 μm, respectively (Fromont 1994, 128-129). Diameter of spermatic cysts, oocytes and embryos in the Hawaiian sponge, *Alectona wallichii*, were 25-24, 90, 200-320 μm, respectively (Vacelet 1998, 631). Diameter of spermatic cysts in the Caribbean sponge *Spongia barbara* was about 40 μm (Kaye and Reiswig 1991, 6). A head and long flagellum of the spermatozoa of *Verongia archeri* at Jamaica were 2.6 and 37-44 μm, respectively. However, head structures of sponge spermatozoa contain a normal condensed nucleus of 0.3-2.5 μm (Reiswig 1970, 539; Reiswig 1983, 4). The egg diameters of *Tethya tenuisclera* and *T. seychellensis* at Maldive Island were 60 and 80 μm, respectively (Gaino and Sara 1994, 100). The egg sizes of a Mediterranean sponge, *Oopsacas minuta*, were 70-110 μm in diameter (Boury-Esnault and Vacelet 1994, 410). Diametical oocytes of *Halichondria panicea* in Kiel Bight were 90-120 μm (Witte and Barthel 1994, 297). Diameter of oocytes in *Petrosia ficiformis* was 300 μm (Liaci et al. cited by Fell 1983, 3). Diameter of oocyte in the Caribbean sponge, *Spongia lachne*, was about 300 μm (Kaye 1991, 18). The results from the present study progress certain debates concerning life history strategies and taxonomy in marine sponges.

Type and Size of Free-Swimming Larva

Sizes of parenchymella larvae in *Petrosia* sp. at Nok Island and Khang Khao Island were 241.9 and 232.3 µm, respectively. The free-swimming parenchymella larvae of *Haliclona permollis* were approximately 250 µm in length and 180 µm in width (Amano and Hori 1996, 162). However, average length of *Halichondria panicea* larva in Kiel Bight was 180-250 µm (Witte and Barthel 1994, 303). Parenchymella larvae of *Niphates* sp. settled about 30% per day on substrates. The process of larval settlement started by the anterior part attached to the substrate (Ilan and Loya 1988, 746). The sizes of the amphiblastula larvae in the two calcareous sponges, *Leucandra abratsbo* and *Sycon* sp. from rafts at the Breeding Center of Aomori Prefecture in northern Japan were 60x45 and 65x50 µm, respectively (Amano and Hori 1992, 82). Early developmental stages of the parenchymella larvae of *Haliclona tubifera* are retained within the parent. Larvae are released through oscula and motile larvae are strongly negatively phototactic (Woollacott 1993, 303).

Fecundity

Average fecundities of parenchymella larvae in *Petrosia* sp. on three types of substrate, i.e., *Porites lutea* , *Palythoa* sp. and rock in the Gulf of Thailand were 24.5±7.96, 19±6.67 and 15±4.11 individuals/cm^2, respectively. However, the average fecundity of parenchymella larvae in

Haliclona cymiformis from Great Barrier Reef was 16.4 individuals/cm^2 (Fromont 1994, 130).

Pattern and Timing of Larval Releasing

Parenchymella larvae of *petrosia* sp. were released in the new moon during 2-5 days (lunar age). However, gametes of *Agelas clathrodes* were released all lunar age (Hoppe 1988, 48, Table 2). Upon releasing, larvae of *Scopalina lophyropoda* remained close to parental sponges and stayed in vertical posture with the wider (anterior) pole towards the sea surface and the pointed (posterior) pole towards the sea bottom (Uriz et al. 1998, 141). Parenchymella larvae of *Petrosia* sp. was obviously observed to show a negative phototactic response. However, all motile parenchymella larvae of the three species of Caribbean sponges, *Hippospongia lachne*, *Spongia barbara*, *S. cheris* and *S. graminea* were demonstrated a negative phototactic response to a unidirectional light source applied directly to the experimental chamber (Kaye and Reiswig 1991, 27). Four Caribbean sponges, *Tedania ignis*, *Sigmadocia caerulea*, *Haliclona tubifera* and *Halichondria magniconulosa* were demonstrated a negative phototactic in laboratory conditions (Maldonado and Young 1996, 169). Although larvae of two sponges, *Crambe crambe* and *Scopalina lophyropoda* swam more actively when they received some light, no particular phototaxis was observed during the first hours of free-swimming life and became photonegative at settlement (Uriz et al. 1998, 141).

Table 2 Gamete and Larval Releasing Times in Demospongiae

Species	Date	Time (hours)	Lunar age*(days)	No. of individuals	References	
Agelas clathrodes (gametes)	23 Jul. 1971	16.00	2	1	Reiswig	Cited in Hoppe 1988,48
	20 Apr. 1974	18.15	26	Many	Reiswig	
	13-15 Jul. 1974	16.20	23-25	3-Many	Reiswig	
	16 Jul. 1976	14.00	19	Many	Reiswig	
	22 Jul. 1976	--	25	Population	Reiswig	
	2 Sep.1976	Night	9	Population	Reiswig	
	30 Jul. 1985	08.50	14	2	Hoppe	
Petrosia sp. (larvae)	21-24 Dec. 1998	08.00	2-5	10	This study	

*Lunar age: new moon 1-7 d; first quarter 8-14 d; full moon 15-21 d; last quarter 22-28 d.

Substrate Selection and Settlement of Larva

Most parenchymella larvae of *Petrosia* sp. settled on living massive coral *Porites lutea* (43±23 %). However, all parenchymella larvae of four species in Caribbean sponges, *Hippospongia lachne*, *Spongia barbara*, *S. cheris* and *S. graminea* did not select a specific settement surface of substrates (Kaye and Reiswig 1991, 27). Larval settlement of *Sigmadocia caerulea* was 43.3 % per 24 hours after releasing in the laboratory (Maldonado and Young 1999, 14). Most larvae of *Crambe crambe* and *Scopalina lophyropoda* settled preferentially in the microrefuges of the grooved plates (Maldonado and Uriz 1998, 141). Most larvae of the two sponges, *Crambe crambe* and *Scopalina lophyropoda* settled preferentially on the container bottoms (Uriz et al. 1998, 143).

Asexual Reproduction

A sponge *Petrosia* sp. grew about 0.65 cm^3/day (average growth rate) and lack of evidence of gemmule formation. However, bud formation and gemmule formation of most sponge are observed to develop in archeocytes (Harrison and De Vos 1991; Fell 1993, 73). Growth rates of three Jamaican sponges, *Mycale* sp., *Verongia gigantea, Tethya crypta* were approximately 60, 5 and 15 % per year, respectively (Reiswig 1973, 207-218). The growth rate of *Xestospongia muta* in the Great Barrier Reef was approximately 0.3

cm/month (Schmahl 1998, 532). However, the growth rate of *Anthosigmella varians* in Buttonwood Sound, Florida was 1.46 cm^3/day (Hill 1996, 652).

Mortality Rate

Average partial mortality rates of *Petrosia* sp. at Nok Island and Khang Khao Island were approximately 0.9 and 0.8 cm^3/day, respectively. However, mortality rate of a Jamaican sponge *Mycale* sp. was approximately 34-35 % per year (Reiswig 1973, 211).

Predation

A sponge *Petrosia* sp. was grazed by a sea urchin, *Diadema setosum* and fed by a nudibranch, *Jorunna funebris* in the Gulf of Thailand. However, a sea urchin, *Arbacia punctulata* grazed on a sponge, *Cliona celata* in the Mystic Estuary, Connecticut (Fell et al. 1984, 136). In Guam, the nudibranch *Glossodoris pallida* preferentially fed on the branching sponge *Cacospongia* sp. (Becerro et al. 1998, 187).

Aquaculture

The sponge *Petrosia* sp. at Nok Island and Khang Khao Island have not yet been successfully farmed in an aquarium. However, two New Zealand sponges *Latrunculia brevis* and *Polymastia croceus* were successfully cultured in mesh and on rope in the field (Duckworth et al. 1998, 155).

Scope for Future Studies

The sponge, *Petrosia* sp. from coral communities at Nok Island and Khang Khao Island in the Inner Gulf of Thailand is an abundant species that provides several important sponge resources. In addition to the findings of this study, many important aspects remain to be examined, especially applications of sponge biology for aquaculture in the future.

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